Effective February 26, 2018
Policy Replaced by LCD L34513

BlueCross BlueShield of Alabama

Name of Blue Advantage Policy:

Serum Biomarker Panel Testing for Systemic Lupus Erythematosus and Other Connective Tissue Diseases

Policy #: 565
Category: Laboratory

Latest Review Date: July 2017
Policy Grade: B

Background:

Blue Advantage *medical policy does not conflict with Local Coverage Determinations (LCDs), Local Medical Review Policies (LMRPs) or National Coverage Determinations (NCDs) or with coverage provisions in Medicare manuals, instructions or operational policy letters. In order to be covered by Blue Advantage the service shall be reasonable and necessary under Title XVIII of the Social Security Act, Section 1862(a)(1)(A). The service is considered reasonable and necessary if it is determined that the service is:*

1. *Safe and effective;*
2. *Not experimental or investigational*;*
3. *Appropriate, including duration and frequency that is considered appropriate for the service, in terms of whether it is:*
   - *Furnished in accordance with accepted standards of medical practice for the diagnosis or treatment of the patient's condition or to improve the function of a malformed body member;*
   - *Furnished in a setting appropriate to the patient's medical needs and condition;*
   - *Ordered and furnished by qualified personnel;*
   - *One that meets, but does not exceed, the patient's medical need; and*
   - *At least as beneficial as an existing and available medically appropriate alternative.*

Routine costs of qualifying clinical trial services with dates of service on or after September 19, 2000 which meet the requirements of the Clinical Trials NCD are considered reasonable and necessary by Medicare. Providers should bill* ***Original Medicare* *for covered services that are related to* ***clinical trials*** *that meet Medicare requirements (Refer to Medicare National Coverage Determinations Manual, Chapter 1, Section 310 and Medicare Claims Processing Manual Chapter 32, Sections 69.0-69.11).*

Description of Procedure or Service:

Systemic lupus erythematosus (SLE) is an autoimmune connective tissue disease that can be difficult to diagnose because patients often present with diverse, nonspecific symptoms, that overlap with other connective tissue diseases and commonly used laboratory tests are not highly accurate. Similar symptoms may also be present in patients with fibromyalgia. Currently, differential diagnosis depends on a combination of clinical signs and symptoms and individual laboratory tests. More accurate laboratory tests for SLE and other connective tissue diseases could facilitate a differential diagnosis in many patients. Recently, laboratory-developed, diagnostic panel tests with proprietary algorithms and/or index scores for the diagnosis of SLE and other autoimmune connective tissue diseases have become commercially available.

Connective Tissue Diseases

Systemic Lupus Erythematosus

Systemic lupus erythematosus (SLE) is an autoimmune connective tissue disease that affects approximately 1.5 million individuals in the U.S. It is one of several types of lupus, the others being cutaneous and drug-induced lupus. About 90% of lupus patients are women between the ages of 15 and 45 years. SLE causes inflammation and can affect any part of the body, most commonly the skin, heart, joints, lungs blood vessels, liver, kidneys, and nervous system. Although generally not fatal, SLE can increase mortality, most commonly from cardiovascular disease due to accelerated atherosclerosis. SLE can also lead to kidney failure, which may reduce survival. The survival rate in the U.S. is approximately 95% at five years and 78% at 20 years. The morbidity associated with SLE is substantial. Symptoms such as joint and muscle pain can impact quality of life and functional status. SLE also increases patients' risk of infection, cancer, avascular necrosis (bone cancer death), and pregnancy complications (e.g., preeclampsia and preterm birth). The course of the disease is variable, and patients generally experience periods of illness (called flares) and periods of remission. Flare severity can range from mild to serious.

Other Connective Tissue Diseases

There are a number of other connective tissue diseases that may require differential diagnosis from SLE. These include the following:

Rheumatoid arthritis (RA) is a chronic inflammatory peripheral polyarthritis. RA can lead to deformity through stretching of tendons and ligaments and destruction of joints through erosion of cartilage and bone. RA can also affect the skin, eyes, lungs, heart, and blood vessels.

Graves disease is an autoimmune disorder that leads to over activity of the thyroid gland. The disease arises from thyroid stimulating hormone-receptor antibodies. It is the most common cause of hyperthyroidism. Blood tests may show raised TSI antibodies.

Hashimoto disease, also known as chronic lymphocytic thyroiditis, is an autoimmune disorder and is the most common cause of hypothyroidism second to iodine insufficiency. It is characterized by an underactive thyroid gland and gradual thyroid failure. Diagnosis is confirmed with blood tests for thyroid stimulating hormone, T4, and antithyroid antibodies.

Sjogren syndrome is an autoimmune disorder characterized by dryness of the eyes and mouth due to diminished lacrimal and salivary gland function. Affected individuals may also have symptoms of fatigue, myalgia, and cognitive dysfunction which may be difficult to distinguish

clinically from fibromyalgia or medication side effects. Typical antibodies include anti-nuclear antibody (ANA), anti-SSA and anti-SSB or rheumatoid factor.

Antiphospholipid syndrome is a systemic autoimmune disorder characterized by venous or arterial thrombosis and/or pregnancy morbidity. Antiphospholipid antibodies are directed against phospholipid-binding proteins.

Polymyositis and dermatomyositis are inflammatory myopathies characterized by muscle weakness and inflammation. Dermatomyositis may also have skin manifestations.

Treatment

Treatments for SLE can ameliorate symptoms, reduce disease activity, and slow progression of organ damage, however there is no cure for SLE. Muscle and joint pain, fatigue and rashes are generally initially treated with nonsteroidal anti-inflammatory drugs. Antimalarial drugs such as hydroxychloroquine can relieve some symptoms of SLE including fatigue, rashes, and joint pain. Patients with more serious symptoms, such as heart, lung or kidney involvement, can be treated with corticosteroids or immune suppressants. There are also biologic treatments, such as rituximab, which are U.S. Food and Drug Administration approved for treatment of rheumatoid arthritis and are being evaluated for SLE.

Diagnosis

Patients with SLE often present with nonspecific symptoms such as fever, fatigue, joint pain, and rash, which can make the disease difficult to diagnosis. Other connective tissue diseases with overlapping symptoms include rheumatoid arthritis, Graves disease, Hashimoto disease, Sjogren syndrome, antiphospholipid syndrome, and polymyositis/dermatomyositis. Similar symptoms may also be present in patients with fibromyalgia. In some patients, the diagnosis can be made with certainty, for example when there are typical symptoms of rash and joint symptoms, and laboratory testing shows a high-titer abnormal antinuclear antibody (ANA) in a pattern that is specific for SLE. However, in many other patients, the symptom patterns are less clear and ANA testing is equivocal, and as a result, cascade testing with additional serologic tests may be ordered. In addition, ANA testing alone can result in false positives due to low specificity.

Classifications

The diagnosis of SLE has been based on a combination of clinical symptoms and laboratory results. In 1997 the American College of Rheumatology (ACR) updated 1982 criteria for classification of SLE. The ACR classification criteria are as follows:

1. Malar rash
2. Discoid rash
3. Photosensitivity
4. Mouth or nose ulcers (usually painless)
5. Arthritis (nonerosive) in two or more peripheral joints, along with tenderness, swelling, or effusion.
6. Serositis: pleuritis or pericarditis
7. Renal disorder: excessive protein in the urine, or cellular casts in the urine
8. Neurologic disorder: seizures and/or psychosis, in the absence of offending drugs or known metabolic derangements

9. Hematologic disorders: hemolytic anemia, leukopenia, lymphopenia or thrombocytopenia
10. Immunologic disorder: antibodies to double-stranded DNA (anti-dsDNA), antibodies to Smith nuclear antigen (anti-Sm), positive antiphospholipid antibody or false positive serologic test for syphilis known to be positive for at least 6 months.
11. ANA test in the absence of drugs known to induce it.

These criteria were originally developed for research, but they have been widely adopted in clinical care. Individuals who meet four or more of the 11 criteria are diagnosed with SLE. If a patient meets fewer than four of criteria, lupus can still be diagnosed by clinical judgment; it is generally recommended that a rheumatologist confirm the diagnosis. ANA testing is usually performed for patients who present with signs and symptoms involving two or more organ systems, and individuals who test positive are recommended for additional laboratory testing. Assessments of the 1982 ACR criteria have reported sensitivities ranging from 78% to 95% and specificities ranging from 89% to 100%, with lower accuracy in patients with mild disease.

In 2012, the Systemic Lupus International Collaborating Clinics (SLICC), an international group of researchers, developed revised criteria for diagnosing SLE. These criteria include more laboratory tests than the earlier ACR criteria, including elements of the complement system. Patients are classified as having SLE if they satisfy four or more of the 18 criteria below, including at least one clinical criterion and one immunologic criterion or they have biopsy-confirmed nephritis compatible with SLE and with ANA or anti-dsDNA antibodies. In a sample of 690 patients, the SLICC criteria had a sensitivity of 97% and a specificity of 84% for diagnosing SLE, whereas the ACR criteria applied to the same sample had a sensitivity of 83% and a specificity of 96%. It is not clear how well-accepted the SLICC recommendations are in the practice setting. The SLICC criteria are outlined in Table 1.

Table 1. Clinical and Immunologic Criteria

Clinical Criteria
• Acute cutaneous lupus (including but not limited to lupus malar rash)
• Chronic cutaneous lupus (including but not limited to discoid rash)
• Oral ulcers
• Non-scarring alopecia in the absence of other causes
• Synovitis involving ≥2 joints, characterized by swelling or effusion or and ≥30 min of morning stiffness
• Serositis
• Renal: excessive protein in the urine or cellular casts in the urine
• Neurologic disorder: seizures, psychosis, mononeuritis complex, or peripheral, or cranial neuropathy
• Seizures
• Hemolytic anemia
• Leukopenia or lymphopenia
• Thrombocytopenia
Immunologic Criteria
• Antinuclear antibody above laboratory reference range
• Antibodies to double-stranded DNA above laboratory reference range
• Antibodies to Smith nuclear antigen
• Antiphospholipid antibody
• Low complement (low C3, low C4, or low CH150)
• Direct Coombs tests in the absence of hemolytic anemia

As previously noted, the SLICC classification system includes a wider range of laboratory tests than the ACR criteria. To date, the most common laboratory tests performed in the diagnosis of SLE are serum ANA, and, if positive, tests for anti-dsDNA and anti-Sm. ANA tests are highly sensitive (i.e., with a high negative predictive value) but have low specificity and relatively low positive predictive value, particularly when the ANA is positive at a low level. Specificity of testing can be increased by testing for specific antibodies against individual nuclear antigens (extractable nuclear antigens [ENAs]) to examine the "pattern" of ANA positivity. These include antigens against single- and double-stranded DNA, histones, Sm, Ro, La, and RNP antibodies. The presence of anti-dsDNA or anti-Sm is highly specific for SLE because few patients without SLE test positive; however, neither of these tests has high sensitivity. The presence of other antibody patterns may indicate the likelihood of other diagnoses. For example, the presence of Ro and La antibodies suggests Sjögren syndrome, while the presence of antihistone antibodies suggests drug-induced lupus.

Better diagnostic tests for SLE and other connective tissue diseases would be useful in clinical practice. A variety of biomarkers, including markers associated with the complement system, are being explored to aid in the diagnosis of lupus. The complement system is part of the immune system and consists of 20 to 30 protein molecules that circulate in the blood in inactive form until activated by a trigger. When activated, as by an infection, a sequence of events known as the complement cascade is initiated. This cascade involves the proteolysis of a complement protein into a smaller protein and a peptide. The smaller protein is able to bind to the complex at the surface of the invading microorganism and the peptide diffuses away. For example, in the first step, complement protein C3 is cleaved into C3b and C3a. C3b binds to the surface of the microorganism and activates the next step in the cascade, the proteolysis of C5, and the small peptide, C3a diffuses away. The precursors C3 and C4 and the complement activation products (CAPs) (e.g., C3a, C5a, C4d) have been considered as SLE biomarkers. More recently, cell-bound complement activation products (CB-CAPs), which live longer than circulating CAPs, have been investigated as biomarkers of SLE.

In addition to exploration of individual biomarkers with higher accuracy than accepted markers (e.g., ANA, anti-dsDNA), there is interest in identifying a panel of tests with high sensitivity and specificity for SLE diagnosis. At least one multibiomarker test to aid diagnosis of SLE and other connective tissue diseases is commercially available. This panel, Avise CTD (Exagen Diagnostics), contains 22 different tests. It combines two smaller panels, a 10-marker panel that includes common SLE tests, as well as CB-CAPs (known as Avise Lupus) and a 12-marker panel that focuses on connective tissue diseases other than SLE (known as Avise CTD [connective tissue disease]). Avise CTD includes nuclear antigen antibodies markers to help distinguish connective tissue disease, a RA panel to rule-in or rule-out RA, an antiphospholipid syndrome panel to assess risk for thrombosis and cardiovascular events, and a thyroid panel to help rule-in or rule-out Graves' disease and Hashimoto disease. Specific biomarkers in the panel are listed in Table 2.

Table 2. Avise Systemic Lupus Erythematosus Tests

Systemic Lupus Erythematosus Tests
10-marker Avise Lupus test
Auto-antibodies: ANA, anti-dsDNA, anti-mutated citrullinated vimentin, C4d erythrocyte-bound complement fragment, C4d lymphocyte-bound complement, anti-Sm, Jo-1, Sci-70, CENP, SS-B/La
Avise CTD test
Avise Lupus test plus the following:
Auto-antibodies: U1RNP, RNP70, SS-A/Ro
Rheumatoid arthritis auto-antibodies: rheumatoid factor IgM, rheumatoid factor IgA, anticyclic citrullinated peptide IgG
Anti-phospholipid syndrome auto-antibodies: cardiolipin IgM, cardiolipin IgG, β2-glycoprotein 1 IgG, β2-glycoprotein 1 IgM
Thyroid auto-antibodies: thyroglobulin IgG, thyroid, thyroid peroxidase

ANA: antinuclear antibody; anti-dsDNA: Antibodies to double-stranded DNA; anti-Sm: antibodies to Smith nuclear antigen; Ig: immunoglobulin

All 22 markers are assessed when the Avise CTD is ordered. Avise CTD uses a three-step process. The ten-marker panel is done in two tiers, and the add-on 12-marker panel is done in a third step to further assist with the differential diagnosis of connective tissue disease. In addition, ANA testing is done by enzyme-linked immunosorbent assay and by indirect immunofluorescence (IIF). The two-tiered testing approach to the 10-marker panel is described next.

> Tier 1: Tests for anti-Sm, EC4d, BC4d, and anti-dsDNA. If any tests are positive, the result is considered suggestive of SLE and no further testing is done. Cutoffs for positivity are greater than 10 U/mL for anti-Sm, greater than 75 U/mL for EC4d, greater than 200 U/mL for BC4d, and greater than 301 U/mL for anti-dsDNA. Positive findings for anti-dsDNA are confirmed with a Crithidia luciliae assay.
>
> Tier 2: If the tier 1 tests are negative, an index score is created, consisting of results of tests for ANA, EC4d/BC4d, anti-MCV, anti-Jo-1, Anti-Sci-70, Anti-CENP, anti Ss-B/La. (In other words, there are 6 additional markers and the ratio of EC4d to BC4d, both of which were measured in tier 1.)

The index score, calculated using a proprietary algorithm, rates how suggestive test results are of SLE. Although there is information on cutoffs used to indicate positivity for individual markers, information is not available on how precisely the index score is calculated. The score can range from -5 (highly nonsuggestive of SLE) to 5 (highly suggestive of SLE) and a score of -0.1 to 0.1 is considered indeterminate.

Exagen also offers the Avise Lupus Prognostic test, a ten-marker panel that can be ordered with the Avise Lupus/Avise CTD panels. The prognostic test focuses on patients' risk of lupus nephritis, neuropsychiatric SLE, thrombosis, and cardiovascular events. The test includes anti-C1q, anti-ribosomal P, anti-phosphatidylserine/prothrombin immunoglobulin (Ig) M and IgG, anti-cardiolipin IgM, IgG, and IgA and anti-β2-glycoprotein 1 IgM, IgG, and IgA. Four of the 10 markers are included in both panel tests.

Policy:

Effective for dates of service prior to February 26, 2018:

Blue Advantage will treat **serum biomarker panel testing** with proprietary algorithms and/or index scores for the diagnosis of systemic lupus erythematosus as a **non-covered benefit** and as **investigational.**

Blue Advantage does not approve or deny procedures, services, testing, or equipment for our members. Our decisions concern coverage only. The decision of whether or not to have a certain test, treatment or procedure is one made between the physician and his/her patient. Blue Advantage administers benefits based on the members' contract and medical policies. Physicians should always exercise their best medical judgment in providing the care they feel is most appropriate for their patients. Needed care should not be delayed or refused because of a coverage determination.

Key Points:

The most recent literature review was updated through April 25, 2017.

Assessment of a diagnostic technology typically focuses on three categories of evidence: 1) technical performance (test-retest reliability or interrater reliability); 2) diagnostic accuracy (sensitivity, specificity, and positive and negative predictive value) in relevant populations of patients; and 3) clinical utility (demonstration that the diagnostic information can be used to improve patient outcomes). In addition, subsequent use of a technology outside of the investigational setting may also be evaluated.

Serum Biomarker Panel Testing for Systemic Lupus Erythematosus and Other Connective Tissue Diseases

Clinical Context and Test Purpose

The purpose of a serum biomarker panel is to inform the differential diagnosis of connective tissue diseases that share similar symptoms. This may allow earlier appropriate treatment and reduce organ damage.

The question addressed in this evidence review is: Does a serum biomarker panel for SLE and other connective tissue diseases improve diagnosis compared to established clinical criteria and laboratory tests?

The following PICOTS were used to select literature to inform this review.

Patients

The relevant population(s) of interest is patients who have signs and/or symptoms of systemic lupus erythematosus (SLE) or other connective tissue diseases but have not been diagnosed. Most of the initial clinical features of SLE are non-specific and include fatigue, joint and muscle pain, rash, and headaches. Initial laboratory features may also be non-specific.

Interventions
This evidence review focuses on a commercially available multibiomarker test to aid in the differential diagnosis of SLE and other connective tissue diseases. This panel, Avise CTD, contains 22 different tests. It combines two smaller panels, a ten-marker panel that includes common SLE tests, as well as cell-bound complement activation products (known as Avise Lupus) and a 12-marker panel that focuses on connective tissue diseases other than SLE (known as Avise CTD).

Comparators
The following tests are currently being used to make decisions about diagnosis and treatment of SLE. Diagnosis is based on a combination of clinical symptoms and laboratory results from the 1997 American College of Rheumatology (ACR) criteria (see background).

Outcomes
Beneficial outcomes: differential diagnosis of SLE from other connective tissue diseases and appropriate treatment, leading to a reduction in joint and organ damage.

Specifically, diagnostic accuracy is the outcome of primary interest because the intent of the Avise Lupus and Avise CTD is to rule out fibromyalgia and facilitate the differential diagnosis of SLE from other connective tissue diseases including rheumatoid arthritis, Graves' disease, Hashimoto disease, Sjogren syndrome, antiphospholipid syndrome, and polymyositis/dermatomyositis.

Harmful outcomes: misdiagnosis.

False-positive test results can lead to inappropriate treatment with adverse effects of medications.

False-negative test results can lead to lack of appropriate treatment.

Timing
Follow-up of several years may be needed to assess the accuracy of the diagnosis.

Setting
These tests may be ordered by a specialist in autoimmune disorders and processed in a central laboratory.

Technical Accuracy
Some individual biomarkers (e.g., antinuclear antibodies [ANA] and antibodies to double-stranded DNA [anti-dsDNA]) are considered standard of care in the diagnosis of connective tissue diseases, and, presumably, the technical accuracy of these tests has been established. In 2017, Dervieux et al reported the technical accuracy of the Avise Lupus biomarker panel. Complement split product C4d bound to erythrocytes (EC4d) and B-lymphocytes (BC4d) were stable for two days at ambient temperature and for four days at 4◦ C. The median intra-day coefficient of variation (CV) ranged from 2.9% to 7.8% (n=30) and inter-day CV ranged from

7.3% to 12.4% (n=66). The two-tiered index score was reproducible over four consecutive days when blood was stores at 4◦ C.

Diagnostic Accuracy

Novel Panel Components: Cell-Bound Complement Activation Products

As previously discussed, CB-CAPs are key components of a commercially available biomarker panel test for lupus diagnosis. CB-CAPs include complement C4d levels on erythrocytes, platelets, and B cells. Preliminary investigations of each of these biomarkers have been done by a research team at the University of Pittsburgh.

A study on lymphocyte-bound complement activation products was published by Liu et al in 2009. This cross-sectional study including 224 patients with systemic lupus erythematosus (SLE) (according to American College of Rheumatology [ACR] classification criteria),179 patients with other autoimmune or inflammatory diseases and 114 healthy controls. Levels of lymphocyte-bound complement activation products, T-cell bound C4d and C3d (TC4d and TC3d) and B-cell- bound C4d and C3d (BC4d and BC3d) were measured in all participants. The diagnostic accuracy of these markers was accessed using receiver-operating characteristic (ROC) analysis. The AUC was 0.727 for TC4d and 0.770 for BC4d. TC4d was estimated to be 56% sensitive and 80% specific for differentiating SLE from other diseases. BC4d had 56% sensitivity and 80% specificity.

In addition, the authors compared the CB-CAPs with other, conventionally used, SLE markers. The markers were evaluated as a confirmatory test in patients who tested positive for ANA. This analysis only included the SLE patients, 223 of 224 of whom (99.6%) were positive for ANA. Of the 223 ANA-positive patients, 141 (63%) patients had elevated levels of TC4d and/or BC4d. In contrast, 59 of the 209 ANA-positive patients (28%) tested positive for anti-dsDNA. Moreover, when the more commonly used CAPs, serum C3 and serum C4, were evaluated, 67 of 221 (30%) of ANA-positive patients tested positive for C3 and 82 of 221 patients (37%) tested positive for C4.

Previously, a 2006 cross-sectional study of platelet C4d was published by Navratil et al. It included 105 patients with SLE (according to ACR criteria), 115 patients with other autoimmune or inflammatory diseases, and 100 healthy controls. Abnormal levels of platelet C4d were detected in 18% of SLE patients. False negative rate and sensitivity rates were not reported. The authors reported that the marker was 100% specific for a diagnosis of SLE compared with healthy controls and 98% specific compared with patients who had other diseases.

Serum Biomarker Panel Tests

In 2014, Putterman et al published data from a large cross-sectional industry-sponsored study evaluating serum biomarkers for the diagnosis of SLE. This study included an analysis of the ten markers in the Avise Lupus (plus ANA) using two-tier testing logic similar to that used in the commercially available panel (see Background section). The study included two cohorts (total N=794); 593 participants were enrolled between April to August 2010, and 201 participants enrolled between June 2011 and September 2013. Together, the two cohorts consisted of 304 patients who met the ACR classification criteria for SLE, 161 patients diagnosed with other

rheumatic diseases and 205 heathy volunteers. Results of serum testing were available for 764 of 794 (96%) participants.

The diagnostic accuracy of the CB-CAP EC4d and BC4d were compared with reduced complement (C3, C4) and anti-dsDNA. The area under the ROC was significantly higher for EC4d (0.82) and BC4d (0.84) than for C3 (0.73) and C4 (0.72) ($p<0.001$). The area under the ROC curve was significantly higher for BC4d than anti-dsDNA (0.79, $p=0.009$) but there was not a significant difference between EC4d and anti-dsDNA.

A total of 140 patients with SLE (46%), nine patients with other diseases (3%) and one healthy volunteer tested positive for at least one of the four tier 1 markers. Patients testing negative for tier-1 tests underwent tier 2 testing and an index score was calculated. A total of 102 of 164 patients with SLE analyzed in tier 2 (62%) had an index score greater than 0 (i.e., suggestive of SLE). Moreover, 245 of 276 patients with other rheumatic diseases had an index score less than 0 (i.e., not suggestive of SLE). When results of tier 1 and tier 2 testing were combined, the overall sensitivity for SLE was 80% (242/304), and the overall specificity for distinguishing SLE from other diseases was 86% (245/285). The specificity for distinguishing between SLE and health volunteers was 98% (201/205).

As shown in Table 3, specificity and AUC was higher for models including CB-CAPs than without these markers; sensitivity was slightly lower.

Table 3: Diagnostic Accuracy of Various Combinations of Markers

Measures	dsDNA, Sm, and ANA	Ds, Sm, ANA, Plus Antibody Specificity Components But Not CB-CAPs	Two-Tiered Testing Using All Markers, Includes CB-CAPs EC4d and BC4d
Sensitivity	89%	83%	80%
Specificity	53%	76%	86%
Area under the curve	0.78	0.80	0.91

ANA: antinuclear antibodies; CB-CAP: cell-bound complement activation product; dsDNA: double stranded DNA; Sm: Smith nuclear antigen.

Previously, an industry-sponsored study published in 2012 by Kalunian et al reported on the first cohort of 593 individuals included in the Putterman et al analysis. The sample consisted of 210 patients with SLE who met ACR classification criteria, 178 patients with other rheumatic diseases, and 205 healthy volunteers. Authors evaluated the performance of a seven-marker biomarker panel for the diagnosis of SLE; some markers are included in a commercially available panel test. The biomarkers included ANA, anti-dsDNA, and anti-MCV measured by enzyme-linked immunosorbent assay. In addition, the authors assessed the cell-bound complement activation products, EC4d, PC4d, and BC4d, determined by fluorescence-activated cell sorting.

In a multivariate logistic regression, SLE diagnosis was associated with a positive ANA test, a negative anti-MCV test, and elevated values of EC4d and BC4d (area under the curve [AUC]=0.92, $p<0.001$). The weighted sum of these four markers correctly categorized 106 of 148 (71.6%) of SLE patients who were anti-dsDNA negative. (The investigators evaluated the

four-marker index score among individuals who tested negative for anti-dsDNA because of the low sensitivity of this test, 29.5%, and thus high false negative rate). The specificity of the four-marker index was 98.0% (200 of 204 healthy volunteers with test results were correctly classified). When anti-dsDNA was added to the four-marker panel, the test had 80% sensitivity for SLE (168 of 210 SLE patients were correctly classified). Moreover, this five-marker test had 97.6% specificity among healthy individuals (200 of 205 were correctly classified as not having SLE). The five-marker test also had 87% specificity in patients with other rheumatic diseases; the most false positives, nine, were in patients with rheumatoid arthritis. The biomarkers in the five-marker test are part of the ten-marker Avise 2.0 SLE test marketed by Exagen. It is not clear whether the index score reported along with the Avise 2.0 panel is the same or different as the index score reported in the Kalunian study.

A limitation of the Putterman et al and Kalunian studies is that study populations included patients with SLE who met ACR classification criteria, but not patients with symptoms suggestive of SLE who failed to meet ACR criteria. It is not known how the diagnostic accuracy of the panel test compares with the ACR classification criteria or to concurrent clinician diagnosis (in the Putterman study, the mean time since SLE diagnosis was 11 years). Furthermore, although they are included in the SLICC classification criteria, the complement factors C3 and C4 are not widely used in clinical practice to diagnose lupus and, therefore, the clinical significance of higher diagnostic accuracy for EC4d and BC4d is unclear.

In 2017, Mossell et al reported an industry-sponsored retrospective study with 23 patients who had a positive Avise Lupus result and 23 patients who had a negative result. All of the patients were ANA positive but negative for autoantibodies specific for SLE, representing cases that were difficult to diagnosis. Each positive Avise test case was matched to a control (negative test) from the same clinic with the same ANA level. The chart review was performed by a non-blinded rheumatologist approximately one year after the test results were available. Of the cases with a positive Avise Lupus test, 20 (87%) were diagnosed with SLE during follow-up. This compared with four (17%) individuals who had a negative result on the Avise Lupus test, resulting in sensitivity of 83.3% and specificity of 86.4%. Interpretation of this study is limited due to the retrospective design, relatively short follow-up to monitor progression of disease, and the lack of an independent reference standard, since diagnosis was based in part on the results of the test. The authors noted that prospective studies will be performed.

Clinical Utility

No studies were identified that provide direct evidence on the impact of serum biomarker panel testing for SLE on patient outcomes. A more accurate and timelier diagnosis of SLE (i.e., before multiorgan system involvement) and other connective tissue diseases could lead to better patient management (e.g., more appropriate medical treatment). This, in turn, could improve health outcomes (e.g., less joint or organ damage, improved survival).

Summary of Evidence

For individuals with signs and/or symptoms of SLE or connective tissue disease other than SLE who receive serum biomarker panel testing, the evidence includes several diagnostic accuracy studies. Relevant outcomes are test accuracy, symptoms, and quality of life. One study evaluated a panel similar to a commercially available test; it found that the panel test had somewhat higher

specificity and lower sensitivity than the most commonly currently used biomarkers. The clinical significance of this degree of difference in diagnostic accuracy is unclear. One case-control study found a high sensitivity and specificity of a commercially available test for the diagnosis of SLE, but this retrospective analysis has a number of limitations. Prospective studies are needed. The evidence is insufficient to determine the effects of the technology on health outcomes.

Practice Guidelines and Position Statements
In 2014, an international group including participants in the European autoimmunity standardization initiative and the International Union of Immunologic Societies published recommendations on the assessment of autoantibodies to cellular antigens. The recommendations included the following statements relevant to the diagnosis of SLE:

- The diagnosis of systemic autoimmune rheumatic diseases (SARD) requires a panel of specific laboratory tests (i.e., ANA [antinuclear antibodies], anti-dsDNA [double-stranded DNA], anti-ENA [extractable nuclear antigen] antibodies)
- The detection of ANA is the first-level test for laboratory diagnosis of SARD.
- If the ANA test is positive, testing for anti-dsDNA antibodies is advised when there is clinical suspicion of SLE
- In case of a positive ANA test…it is recommended to perform specific tests for anti-ENA antibodies…."

U.S. Preventive Services Task Force Recommendations
This topic is not a preventive service.

Key Words:
Avise SLE, systemic lupus erythematosus, SLE, serum biomarker panel test

Approved by Governing Bodies:
Clinical laboratories may develop and validate tests in-house and market them as a laboratory service; laboratory-developed tests must meet the general regulatory standards of the Clinical Laboratory Improvement Act (CLIA). The Avise® tests (Exagen Diagnostics) are available under the auspices of CLIA. Laboratories that offer LDTs must be licensed by CLIA for high-complexity testing. To date, the U.S. Food and Drug Administration has chosen not to require any regulatory review of this test.

Benefit Application:
Coverage is subject to member's specific benefits. Group specific policy will supersede this policy when applicable.

Current Coding:
CPT Codes:

There is not specific CPT code for this panel of tests. There are codes that would likely be used for some of the component tests such as:

83520	Immunoassay for analyte other than infectious agent antibody or infectious agent antigen; quantitative, not otherwise specified
86038	Antinuclear antibodies (ANA)
86039	Antinuclear antibodies (ANA); titer
86146	Beta 2 Glycoprotein I antibody, each
86147	Cardiolipin (phospholipid) antibody, each Ig class
86200	Cyclic citrullinated peptide (CCP), antibody
86225	Deoxyribonucleic acid (DNA) antibody; native or double stranded
86235	Extractable nuclear antigen, antibody to, any method (e.g., nRNP, SS-A, SS-B, Sm, RNP, Sc170, J01), each antibody
86376	Microsomal antibodies (e.g., thyroid or thyroid-kidney), each
86800	Thyroglobulin antibody
88184	Flow cytometry, cell surface, cytoplasmic, or nuclear marker, technical component only; first marker
88185	Flow cytometry, cell surface, cytoplasmic, or nuclear marker, technical component only; each additional marker (List separately in addition to code for first marker)
88187	Flow cytometry, interpretation; 2 to 8 markers

Some payers might instruct the use of the unlisted chemistry code for the whole panel:

84999	Unlisted chemistry procedure

Due to the reporting of an index score for the entire panel, the test would more accurately be reported with the unlisted multianalyte assay with algorithmic analysis (MAAA) CPT code:

81599	Unlisted multianalyte assay with algorithmic analysis

References:

1. Agmon-Levin N, Damoiseaux J, Kallenberg C, et al. International recommendations for the assessment of autoantibodies to cellular antigens referred to as anti-nuclear antibodies. Ann Rheum Dis. Jan 2014; 73(1):17-23.
2. American College of Rheumatology (ACR). The 1982 Revised Criteria for Classification of Systemic Lupus Erythematosus. www.rheumatology.org/practice/clinical/classification/SLE/sle.asp. Accessed June, 2014.
3. Cabas-Vargas J, Chitkara P, Christianakis S, et al. Finding the best approach to autoimmune connective tissue disease diagnosis (Paid supplement supported by Exagen Diagnostics, Inc). Rheumatology News. 2014; August: 1-8.
4. Dervieux T, Conklin J, Ligayon JA, et al. Validation of a multi-analyte panel with cell-bound complement activation products for systemic lupus erythematosus. J Immunol Methods. Apr 05 2017.
5. Gill JM, Quisel AM, Rocca PV, et al. Diagnosis of systemic lupus erythematosus. Am Fam Physician. Dec 1 2003; 68(11):2179-2186.

6. Guidelines for referral and management of systemic lupus erythematosus in adults. American College of Rheumatology Ad Hoc Committee on Systemic Lupus Erythematosus Guidelines. Arthritis Rheum. Sep 1999; 42(9):1785-1796.
7. Hochberg MC. Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum. Sep 1997; 40(9):1725.
8. Kalunian KC, Chatham WW, Massarotti EM, et al. Measurement of cell-bound complement activation products enhances diagnostic performance in systemic lupus erythematosus. Arthritis Rheum. Dec 2012; 64(12):4040-4047.
9. Kasitanon N, Magder LS, Petri M. Predictors of survival in systemic lupus erythematosus. Medicine (Baltimore). May 2006; 85(3):147-156.
10. Liu CC, Kao AH, Hawkins DM, et al. Lymphocyte-bound complement activation products as biomarkers for diagnosis of systemic lupus erythematosus. Clin Transl Sci. Aug 2009; 2(4):300- 308.
11. Lupus Research Institute. Lupus Fact Sheet. www.lupusresearchinstitute.org/lupus-facts/lupus-fact-sheet.
12. Manzi S, Navratil JS, Ruffing MJ, et al. Measurement of erythrocyte C4d and complement receptor 1 in systemic lupus erythematosus. Arthritis Rheum. Nov 2004; 50(11):3596-3604.
13. Mossell J, Goldman JA, Barken D, et al. The Avise Lupus test and cell-bound complement activation products aid the diagnosis of systemic lupus erythematosus. Open Rheumatol J. 2016; 10:71-80.
14. Navratil JS, Manzi S, Kao AH, et al. Platelet C4d is highly specific for systemic lupus erythematosus. Arthritis Rheum. Feb 2006; 54(2):670-674.
15. Petri M, Orbai AM, Alarcon GS, et al. Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. Arthritis Rheum. Aug 2012; 64(8):2677-2686.
16. Putterman C, Furie R, Ramsey-Goldman R, et al. Cell-bound complement activation products in systemic lupus erythematosus: comparison with anti-double-stranded DNA and standard complement measurements. Lupus Sci Med. 2014; 1(1):e000056.
17. Suresh E. Systemic lupus erythematosus: diagnosis for the non-specialist. Br J Hosp Med (Lond). Oct 2007; 68(10):538-541.

Policy History:

Adopted for Blue Advantage, September 2014
Available for comment September 23 through November 6, 2014
Medical Policy Group, June 2016
Medical Policy Group, July 2017
Medical Policy Group, January 2018