CELL BOUND COMPLEMENT ACTIVATION PRODUCTS DISTINGUISH SYSTEMIC LUPUS ERYTHEMATOSUS FROM OTHER DISEASES AMONG PATIENTS WITH HIGH ANTINUCLEAR ANTIBODY TITERS AND NORMAL COMPLEMENT

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ABSTRACT

PURPOSE

Patients with a suspicion of systemic lupus erythematosus (SLE) are often referred to the rheumatologist because of elevated anti-nuclear antibody (ANA) titers. We sought to evaluate the association of cell bound complement activation products (CB-CAPs), low complement (C3 or C4), and anti-double stranded (ds) DNA with SLE in the context of different ANA titers.

METHODS

The cohort (N=1155, all adults) consisted of 498 SLE with established disease (all fulfilling the 1997 ACR criteria, 91% females, mean age 41 years) pooled from prior studies of complement activation products, and a control group of 657 subjects (86% females; mean age 56 years; inclusive of 314 rheumatoid arthritis and 343 other diseases subjects). Abnormal CB-CAPs consisted of C4d bound to erythrocyte or B-lymphocyte levels above the 99th percentile of normal healthy. Low complement (C3 and C4) and anti-dsDNA (all confirmed using Crithidia luciliae) were determined using immunoassays. ANA titers were determined by indirect immunofluorescence, with subjects classified as having negative (<1:80), intermediate (1:80 to 1:320) or high (≥1:640) ANA status. The association of the markers in distinguishing SLE from non SLE were evaluated using sensitivity, specificity, and Youden's index (J), a measure of diagnostic effectiveness that combines sensitivity and specificity (J= sensitivity+ specificity -1). J differences were tested using t-tests.

RESULTS

The association of abnormal CB-CAPs, low complement and anti-dsDNA in distinguishing SLE from non SLE are presented in the Table. Overall, abnormal CB-CAPs was significantly more associated with SLE (J=0.51) than low complement (J=0.32) and anti-dsDNA (J=0.31) (p<0.01; n=1155). The greater association of abnormal CB-CAPs in comparison to low complement and anti-dsDNA was statistically significant in the group of subjects with high ANA (p<0.03), intermediate ANA (p<0.01) and negative ANA (p<0.02). This association was also seen among subjects with high ANA (J=0.60) compared to intermediate (J=0.45) and negative ANA (J=0.17) (p<0.01). Similar results were observed for low complement and anti-dsDNA (p<0.01). In the group of subjects with normal complement (309 SLE and 619 non SLE), abnormal CB-CAPs was 50% sensitive and 89% specific while anti-dsDNA was 20% sensitive and 99% specific (J=0.39 vs 0.19; p<0.01). In the subset of subjects with high ANA and normal complement (117 SLE and 106 non SLE), abnormal CB-CAPs was 68% sensitive and 82% specific and yielded higher diagnostic value than anti-dsDNA alone (40% sensitive and 93% specific) (J=0.50 vs 0.34; p<0.01).

CONCLUSION

Abnormal CB-CAPs is more significantly associated with the diagnosis of SLE compared with low complement and anti-dsDNA and is a sensitive and specific measure for SLE in subjects with high ANA titers and normal complement levels.

OBJECTIVE

- We decided to evaluate the association of elevated cell-bound complement activation products (CB-CAPs), low complement (low C3 and/or C4), and positive anti-double stranded (ds) DNA antibodies with SLE in the context of different ANA titers: negative (<1:80): intermediate (1:80 to 1:320); high (≥1:640)

METHODS

Cohort:
N= 1155 subjects, all adults:

- 498 SLE with established disease (all fulfilling the 1997 ACR criteria, (91% females, mean age 41 years) pooled from prior studies of complement activation products
- 657 controls (86% females; mean age 56 years) inclusive of 314 rheumatoid arthritis and 343 other diseases.

Biomarkers:

- CB-CAPs: C4d bound to erythrocyte or B-lymphocyte levels were determined using flow cytometry. Abnormal CB-CAPS consisted of levels above the 99th percentile of normal healthy group.
- Low complement levels (C3 and C4) were determined using immunoturbidimetry (Optilite, The Binding site)
- Anti-dsDNA determined by using ELISA (INOVA Diagnostics) and conformed using *Crithidia luciliae*.
- ANA titers were determined by indirect immunofluorescence (NOVA view), with subjects classified as having negative (<1:80), intermediate (1:80 to 1:320) or high (≥1:640) ANA status.

Statistical analysis:

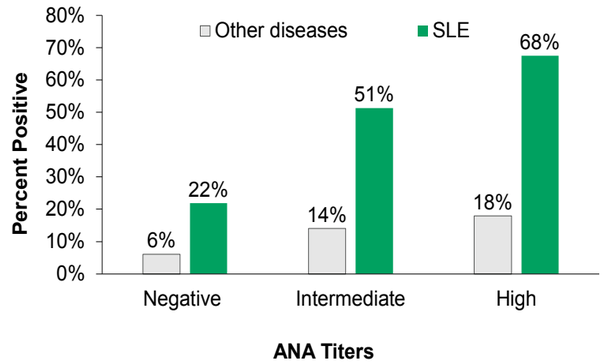
- The association of the markers in distinguishing SLE from non SLE were evaluated using sensitivity (se), specificity (Sp), and Youden's index (J).
- J is a measure of diagnostic effectiveness that combines sensitivity and specificity (J=sensitivity+ specificity -1).
- J differences were compared using t-tests.

RESULTS

Abnormal CB-CAPs was significantly more associated with SLE (J=0.51) than low complement (J=0.32) and anti-dsDNA (J=0.31) irrespective of ANA titers (p<0.01)

	Negative ANA [<1:80] Total=374, 79 SLE	Intermediate ANA [1:80-1:320] Total=430, 181 SLE	High ANA [≥1:640] Total=351, 238 SLE	All ANA [<1:80-≥1:640] Total=1155, 498 SLE
Anti-dsDNA	Se=4±2% Sp=99±0% J=0.03±0.02	Se=18±3% Sp=100±0% J=0.17±0.03	Se=53±3% Sp=94±2% J=0.46±0.04	Se=32±2% Sp=98±0% J=0.31±0.02
Low complement	Se=8±3% Sp=95±1% J=0.02±0.03	Se=34±4% Sp=94±2% J=0.28±0.04	Se=51±3% Sp=94±2% J=0.45±0.04	Se=38±2% Sp=94±1% J=0.32±0.02
Abnormal CB-CAPs	Se=23±5% Sp=94±1% J=0.17±0.05	Se=59±4% Sp=86±2% J=0.45±0.04	Se=77±3% Sp=82±4% J=0.60±0.05	Se=62±2% Sp=89±1% J=0.51±0.02

Abnormal CB-CAPs by ANA Titers Among Subjects with Normal Complement Levels



CONCLUSION

Abnormal CB-CAPs is more significantly associated with the diagnosis of SLE compared with low complement and anti-dsDNA and is a sensitive and specific measure for SLE in subjects with high ANA titers and normal complement levels.