Evaluation of anti-CENP reactivity in samples exhibiting the centromere HEp-2 pattern, which is associated with a better prognosis within the limited cutaneous systemic sclerosis spectrum

Poster

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Introduction/Objectives:
Anti-centromere antibodies (ACA) are among the most specific markers for systemic sclerosis (SSc), particularly the limited cutaneous form (lcSSc), where they are traditionally associated with a more favorable prognosis. The discrete speckled centromere pattern (AC-3) on HEp-2 cells strongly suggests the presence of antibodies directed against centromere proteins, especially CENP-B and CENP-A. However, the precise correlation of AC-3 reactivity with specific CENP antigens, as well as its clinical implications within a homogeneous lcSSc cohort, remains incompletely understood. This study aimed to investigate the frequency and distribution of anti-CENP-A and anti-CENP-B reactivities in sera from lcSSc patients displaying the AC-3 pattern, and to analyze their clinical and demographic associations, particularly in relation to organ involvement.

Methods:

A total of 76 patients fulfilling ACR/EULAR 2013 classification criteria for lcSSc were included. Among them, 38 exhibited the AC-3 pattern on HEp-2 IFA, while 48 were classified as Non-AC-3. Reactivity to CENP-B and CENP-A was evaluated by two solid-phase immunoassays: ELISA (for CENP-B) and line-blot (for both CENP-A and CENP-B). In parallel, clinical data from 68 lcSSc, including age, sex, disease duration, and major organ involvement (interstitial lung disease [ILD], pulmonary arterial hypertension, esophageal dysmotility, arthritis, and cardiac or renal manifestations) were reviewed. Statistical comparisons between groups and correlations between assays were performed.

Results:
Among the AC-3 samples, 92.1% were positive for CENP-B by ELISA and 84.2% by line-blot, while 81.6% reacted with CENP-A. Concordance for CENP-B reactivity between ELISA and line-blot was moderate (78.9%). Overall, 100% of AC-3-positive samples displayed reactivity to either CENP-A or CENP-B, confirming the high sensitivity of HEp-2 IFA in identifying ACA. Combined reactivity to both CENP-A and CENP-B in all platforms was observed in 68.4% of samples, whereas one serum exhibited exclusive anti-CENP-A reactivity, and seven were positive only for CENP-B. Clinically, AC-3-positive patients were older but showed significantly reduced frequency of ILD compared to Non-AC-3 lcSSc patients (10.5% vs. 54.2%; p=0.001). Frequencies of other organ manifestations, including esophageal involvement, arthritis, pulmonary hypertension, or renal and cardiac complications, were similar between groups.

Conclusions:
This study demonstrates that in lcSSc patients, the AC-3 centromere pattern on HEp-2 IFA is strongly predictive of anti-CENP reactivity, with CENP-B being the predominant target but anti-CENP-A also present, including rare cases of exclusive reactivity. Importantly, the presence of AC-3 in this lcSSc cohort was associated with a distinctly lower prevalence of ILD, reinforcing its role as a marker of milder disease course and more favorable prognosis. The incomplete concordance between ELISA and line-blot highlights the variability between solid-phase assays, underscoring the importance of HEp-2 IFA as a primary screening tool. Altogether, these findings support the clinical utility of combining immunofluorescence with antigen-specific assays for the precise characterization of anti-centromere reactivity in systemic sclerosis, and provide evidence that ANA patterns can further stratify lcSSc patients into a subgroup with reduced risk of severe pulmonary involvement.

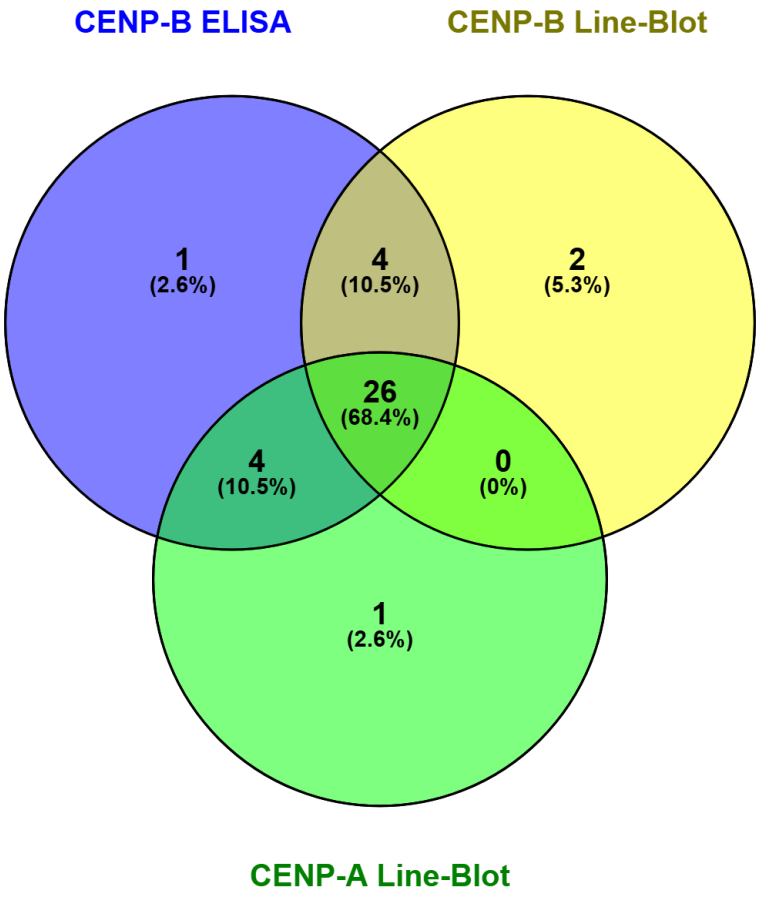
key-points

- In a cohort composed exclusively of limited cutaneous Systemic Sclerosis patients, those with the AC-3 centromere HEp-2 pattern showed significantly lower prevalence of interstitial lung disease compared to those without AC-3.

- CENP-B is the dominant autoantigen in AC-3 samples, but anti-CENP-A reactivity was also prevalent and exclusive anti-CENP-A reactivity was observed in one sample.
- HEp-2 IFA was 100% sensitive for detecting anti-CENP antibodies, meaning all AC-3-positive samples reacted with CENP-B and/or CENP-A in at least one solid-phase immunoassay, reinforcing HEp-2 IFA as a highly reliable screening method.

Palabras Clave

(1) Systemic Scleroderma, (2) Autoantibodies, (3) Fluorescent Antibody Technique, (4) HEp-2 Cells, (5) Centromere.



Venn diagram for anti-CENP-B/A reactivity in ELISA and line-blot assays in samples from the AC-3 group.

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