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Line-field confocal optical coherence tomography: Characteristic hints for the diagnosis of scarring alopecia due to lupus erythematodes: A preliminary study

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Abstract

Introduction: Lupus erythematosus (LE) is an inflammatory autoimmune disease, that can affect the skin to varying degree. In particular, discoid LE (DLE) and the rare form of lupus panniculitis/profundus are associated with scarring alopecia. The heterogeneity of the clinical, dermatoscopic, and histologic presentation poses a major challenge to the clinician in the diagnosis and differential diagnosis of other forms of scarring alopecia.

Objective: While noninvasive imaging techniques using optical coherence tomography (OCT) and reflectance confocal microscopy (RCM) have proven to be helpful in the diagnosis of scarring alopecia in the context of LE, this study aimed to investigate line-field confocal OCT (LC-OCT) to identify characteristic features of cicatricial alopecia in LE.

Methods: Fifteen patients with cicatricial alopecia in LE were included and the most affected/inflamed areas of the scalp were prospectively examined. In analogy to histopathology and previously reported criteria in RCM, all images were evaluated according to seven established criteria and underwent descriptive analyses.

Results: LC-OCT revealed characteristic features of cicatricial alopecia, such as lymphocytic interface dermatitis (14/15; 93.3%) and basal cell vacuolization (13/15; 86.7%). The most impressive feature was the occurrence of prominent hyperreflective fibers in 14/15 patients (93.3%).

Conclusion: LC-OCT imaging can noninvasively detect morphologic criteria such as lymphocytic and vacuolar interface dermatitis of cicatricial alopecia due to LE. In particular, the presence of hyperreflective collagen fibers appears to be a characteristic easily recognizable feature that may facilitate differential diagnosis with other forms

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of cicatricial alopecia. Further studies are mandatory to differentiate other forms of scarring alopecia.

KEYWORDS
CDLE, chronic-discoid lupus erythematodes, cicatricial alopecia, LC-OCT, noninvasive imaging, OCT, optical coherence tomography, RCM, reflectance confocal microscopy

1 | INTRODUCTION

Lupus erythematosus (LE), an inflammatory autoimmune disease, shows different subtypes with multiple clinical manifestations. Depending on the subtype, the skin can be affected to varying degree. While systemic LE (SLE) shows the involvement of multiple organs of the body with skin involvement in up to 75%, other subtypes of LE are limited solely to the skin (= cutaneous LE, CLE).[1,2]

Skin involvement in LE is also associated with alopecia in up to 85% of the cases. While nonscarring alopecia, usually in the form of diffuse effluvium, may occur nonspecifically during the course of the disease, scarring alopecia is associated with certain subtypes of CLE and results in a significant reduction in quality of life.[1–3]

SLE (– 20%, mainly as an overlap with DLE), and especially the cutaneous forms discoid LE (DLE) (– 60%) as well as the rare form lupus panniculitis/profundus (- 16.4%) are associated with scarring alopecia, whereby the various forms may overlap.[1,2,4]

The lesions occur primarily on the vertex, the most sun-exposed region of the scalp. They typically present as solitary or multiple plaques with marginal scaling and central scarring. Follicular plugging, telangiectasias, atrophy, depigmentation, and hyperpigmentation are also frequently seen.[5] Dermatoscopy shows follicular hyperkeratosis, follicular red dots and large yellow dots, thick arborizing vessels, and scattered dark-brown discoloration of the skin.[6] The diagnosis is confirmed by means of scalp biopsy and subsequent histological assessment. In addition to general characteristics of scarring alopecia such as fibrosis, in CDLE vacuolar interface dermatitis, lymphocytic inflammatory infiltrates, dilated infundibula with laminar keratin and interstitial mucin in the dermis are typically seen. Direct immunofluorescence (DIF) shows granular deposits of IgG, IgM, and C3 along the basement membrane zone.[5,7,8]

However, the diagnosis of scarring alopecia in the context of LE is often a major challenge for the practitioner. This is due to the heterogeneity of the clinical, dermatoscopic, and histologic presentation, which makes it difficult to differentiate LE from other forms of scarring alopecia like lichen planopilaris (LPP), which can present clinical and histological mimicry.[7,9,10]

In the past, noninvasive imaging techniques using optical coherence tomography (OCT) and especially reflectance confocal microscopy (RCM) have proven to be helpful in diagnosing cicatricial alopecia associated with LE.[11–18] Referring to Ardigò et al. and Micali et al., specific morphologic RCM features of scalp have been reported, that suggest its suitability for monitoring treatment, selecting the correct biopsy site, and aiding in the differential diagnosis with other forms of cicatricial alopecia such as LPP. However, due to their general physical limitations, RCM and OCT are limited in the diagnostic value of cicatricial alopecia, as the penetration depth of RCM is only up to 250 μm and the optical resolution of OCT is only up to 7.5 μm.

Line-field confocal optical coherence tomography (LC-OCT) is a high-resolution imaging technique, that enables real-time imaging up to cellular level (resolution 1–2 μm) with an increased penetration depth up to 500 μm. While LC-OCT imaging has proven its potential for in vivo diagnosis of various skin tumors, their subtyping and tumor margins, it also showed promising results in the imaging of inflammatory dermatoses.[19–22] Recent work by Rudnicka et al. also described LC-OCT imaging of scarring alopecia in LPP and folliculitis decalvans.[23,24]

Therefore, the aim of this pilot study was to identify characteristic morphologic features of scarring alopecia in patients with LE using LC-OCT and to compare them with histologic findings and previously reported noninvasive methods such as RCM and OCT.

2 | SUBJECTS AND METHODS

2.1 | Subjects

Between November 2023 and April 2024, 15 patients with scarring alopecia due to LE (11 female and four male) with a mean age of 47.7 years (range 21–76 years) were recruited from the trichology and autoimmune disease outpatient department of the Department of Dermatology and Allergy at the University Hospital of the Ludwig-Maximilian University (LMU) in Munich. Twelve (eight female and four male) patients were diagnosed with DLE, one female patient with SLE, one female patient with lupus panniculitis overlapping with DLE and SLE and one female patient with lupus profundus/panniculitis. The diagnosis was made based on clinical examination, trichoscopy, and any appropriate laboratory constellation (ANAs, ENAs, ACR/EULAR criteria). The diagnosis was confirmed by histopathology with DIF in 14 patients.

For each patient, we documented age, sex, disease duration, as well as current therapy.

Twelve patients had systemic anti-inflammatory therapy (6 hydroxychloroquine, 3 hydroxychloroquine + methotrexate (MTX), 2 hydroxychloroquine + mycophenolate mofetil (MMF) + prednisolone, 1 MTX) of whom two had also topical anti-inflammatory therapy. Two patients used topical anti-inflammatory therapy only and one patient was therapy-naive.

TABLE 1 Demographic data of the patients.

Patient	Sex	Age	Lupus phenotype	Scarring alopecia since	Treatment
1	f	21 y	SLE, lupus pannikulitis, DLE	0.5 y	Prednisolone, hydroxychloroquine, MMF
2	f	48 y	Lupus profundus/pannikulitis	0 y	Hydroxychloroquine
3	m	59 y	DLE	0.5 y	Hydroxychloroquine
4	f	76 y	DLE	9 y	Topical antiinflammatory treatment
5	f	38 y	DLE	1.5 y	Hydroxychloroquine, topical antiinflammatory treatment
6	f	56 y	DLE	15 y	Hydroxychloroquine, MTX
7	m	31 y	DLE	5 y	/
8	f	70 y	SLE	4 y	Prednisolone, hydroychloroquine, MMF
9	f	38 y	DLE	10 y	MTX, topical antiinflammatory treatment
10	f	48 y	DLE	12 y	Topical anti-inflammatory treatment
11	f	61 y	DLE	1 y	Hydroxychloroquine
12	f	32 y	DLE	21 y	Hydroxychloroquine, MTX
13	m	46 y	DLE	3 y	Hydroxychloroquine
14	m	32 y	DLE	13 y	Hydroxychloroquine
15	f	60 y	DLE	20 y	Hydroxychloroquine, MTX

Abbreviations: DLE, discoid lupus erythematosus; f, female; m, male; MMF, mycophenolate mofetil; MTX, methotrexate; SLE, systemic lupus erythematosus.

Scarring alopecia in the context of LE occurred for an average of 7.7 years.

See Table 1 for an overview of the demographic data of all patients.

Patients underwent clinical, dermoscopical, and LC-OCT examination after signed informed consent.

3 | METHODS

3.1 | Imaging tools and imaging protocol

Primarily, clinical and dermoscopic images of the hair follicles were recorded using Fotofinder (FotoFinder GmbH, Germany) and Dermogenius (Dermoscan GmbH, Regensburg, Germany). Following dermoscopy, the scalp was imaged using noninvasive imaging.

In each patient, the most clinically and dermoscopically "inflamed" lesions as well as nonaffected hair follicles were subjected to LC-OCT examination.

LC-OCT was conducted with the deepLive™ device (DAMAE Medical, Paris, France). LC-OCT is a noninvasive optical imaging tool which can generate cell-resolved images of the skin up to a depth of 500 μm in real time. Further technical details have been described elsewhere.[25,26]

For each patient, vertical images and videos as well as three-dimensional (3D) blocks were recorded.

3.2 | LC-OCT criteria

Based on the histological and previously described RCM criteria for scarring alopecia in LE, we evaluated LC-OCT recordings for the presence of the following seven features:

- Hyperreflective collagen bundles
- No rimming of the papillae
- Follicular remnants
- Lymphocytic interface dermatitis
- Basal cell vacuolization
- Melanophages
- Mucin deposits.

All images were analyzed by three experienced examiners (M.D., E.C.S., and M.-C.N.) who agreed on the outcome for each patient.

LC-OCT presentation of the seven microscopic features is summarized in Table 2.

4 | RESULTS

Resembling histopathology, lymphocytic infiltrates were seen with LC-OCT in the superficial dermis in 14 patients (14/15; 93.3%). The lymphocytes were visualized as hyperreflective, roundish small

TABLE 2 Description of the LC-OCT features.

LC-OCT criteria	Description
Hyperreflective collagen bundles	Hyperreflective linear structures in the upper dermis, arranged in different directions, corresponding to thickened collagen bundles
No rimming of the papillae	No visibility of roundish regularly arranged structures at the epidermis–dermis border corresponding to dermal papillae
Follicular remnants	Roundish areas associated with former hair follicles, filled with bright material and necrotic edematous structures
Lymphocytic interface dermatitis	Small, bright, round cellular structures, as singles or tightly aggregated, at the level of the dermo-epidermal junction
Basal cell vacuolisation	Mildly to low refractile round cellular structures at the dermo-epidermal junction, often associated with a complete annular bright ring with polar enhancement surrounding a dark center corresponding to signet ring-like cells
Melanophages	Polygonal bright cellular structures in the upper dermis
Mucin deposits	Dark, hyporeflective, black ribbon-like structures in the superficial and upper dermis, intermingled with undefined bright areas

structures. Another histopathologic criterion characteristic of LE is basal cell vacuolization, which was detected in 13/15 (86.7%) patients by LC-OCT and appears as mild-to-low refractile cells at the dermo-epidermal junction (DEJ). These two inflammatory changes in the area of the DEJ led to complete (12/15) or partial (2/15) obliteration of the rim of the dermal papillae in 14 patients (14/15; 93.3%). In 10 patients (10/15; 66.7%), a typical feature of vacuolar interface dermatitis described by Amini-Adle et al. for RCM, the signet ring-like cells, could also be well identified by LC-OCT.[27] These appear as a complete annular bright ring with polar enhancement surrounding a dark center, with keratinocyte necrosis and apoptotic keratinocytes thought to be responsible for this vacuolar degeneration (Figure 1).

The most impressive feature seen with LC-OCT in 14 patients (14/15; 93.3%) was hyperreflective linear structures arranged in different directions, most likely corresponding to thickened collagen bundles in the sense of perifollicular fibrosis due to the scarring aspect of alopecia (Figure 2). Dark, hyporeflective, black ribbon-like structures were seen in the superficial and upper dermis above and between the thickened collagen bundles in 13 patients (13/15; 86.7%). These structures have not been described for RCM in context with LE. Correlating with histopathology, these structures may represent mucin deposition, which is typical for LE.

Furthermore, follicular remnants containing laminar keratin were seen in 11 cases (11/15; 73.3%), detectable in LC-OCT recordings as roundish or oval areas filled with white amorphous material (Figure 3).

In nine patients (9/15; 60.0%), the upper dermis displayed a dense infiltrate of plump bright, oval-to-stellate cells consistent with the presence of abundant melanophages in the upper dermis.

LC-OCT findings are displayed case by case in Table 3.

5 | DISCUSSION

Noninvasive imaging using RCM and OCT has proven to be useful in the diagnosis of various skin diseases. In recent years, LC-OCT has emerged as an innovative noninvasive imaging method that combines the advantages of both methods due to its higher resolution than OCT and increased penetration depth compared to RCM.[25,26] It has already proven its value in the diagnosis of numerous tumorous and inflammatory dermatoses. Rudnicka et al. have already described LC-OCT as a useful diagnostic tool in two forms of cicatricial alopecia, namely LPP and folliculitis decalvans.[23,24]

Since scarring alopecia in LE is often difficult to differentiate, we investigated whether LC-OCT can be used to visualize characteristic features of this disease, analogous to previous studies with RCM. To this end, we recorded the most clinically and dermoscopically inflamed lesions due to scarring alopecia in LE in 15 patients with LC-OCT and evaluated the images and 3D blocks in correlation with histopathological findings as well as with previously reported RCM and OCT criteria.

Analogous to histopathology, we were able to visualize the characteristic lymphocytic interface dermatitis in 14 patients with LC-OCT (93.3%). These results agree with the findings of LE in RCM described by Ardigo, Agozzino, and Lacarruba et al. In the context of interface dermatitis, LE is often associated with vacuolar degeneration of the basal layer of the epidermis, which was also found in 86.7% of our patients on LC-OCT and in the cited RCM studies. In 14 out of 15 patients (93.3%), this interface dermatitis also led to a blurring of the DEJ, so that the so-called rimming of the dermal papillae is no longer recognizable on LC-OCT.

The signet ring-like cells described by Amini-Adle et al. as an additional indication of interface dermatitis correlating with basal cell vacuolization have also been described by Melo et al. in the context of discoid scalp lupus and RCM.

To our knowledge, this is the first time describing basal cell vacuolization in context with these signet ring-like cells with LC-OCT.

The most impressive feature, present in 93.3% of the patients, was thickened collagen bundles, which were seen with LC-OCT as highly hyperreflective linear structures arranged in different directions in the upper dermis. Lacarruba and Agozzino et al. also described thick collagen bundles in the context of dermal sclerosis in 100% of lupus patients examined with RCM. Adigo and Melo et al. described them as abnormal collagen bundles and found them in 60% of patients with RCM, while Ardigo et al. attributed the missing 40% to the insufficient dissolution of the RCM at the level of the papillary dermis.

Judging from the images shown in the above RCM papers, the collagen fibers are not as impressive and brightly visualized by RCM compared to the LC-OCT images of our patients. The description of "pale" fibers in the RCM also shows the difference to the hyperreflective fibers in our LC-OCT recordings. There are different explanations for this phenomenon.

On the one hand, it could be due to the different imaging methods with LC-OCT in our study and RCM in the previous reports.

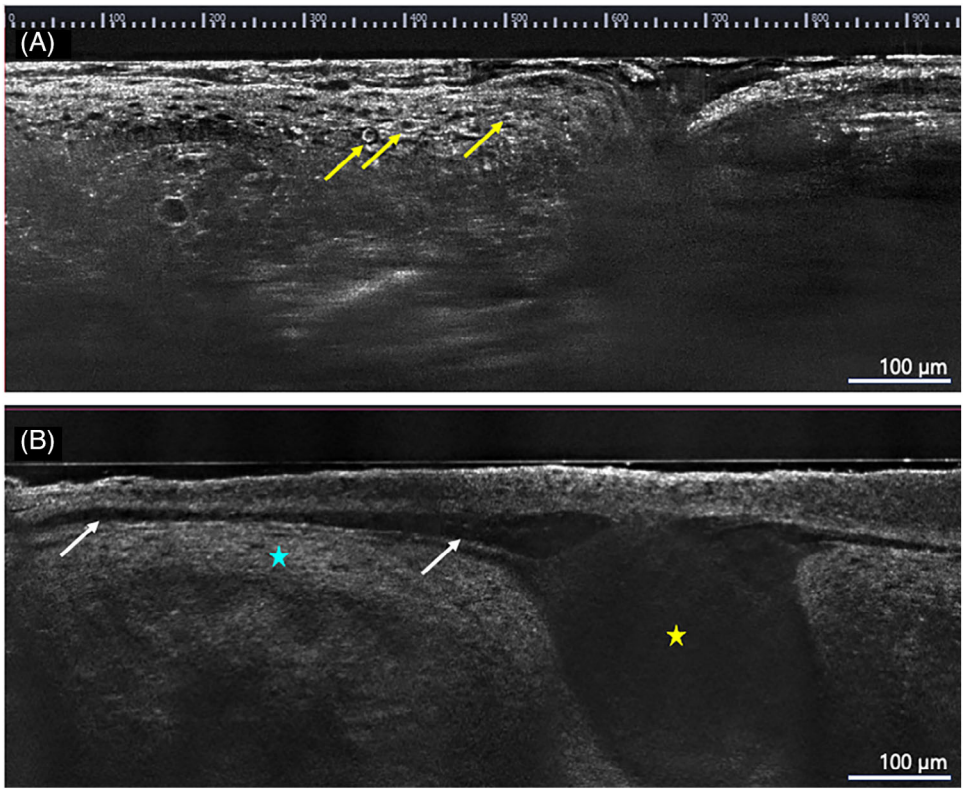

FIGURE 1 (A) Basal cell vacuolization with signet ring-like cells (yellow arrows) appearing as a complete annular bright ring with polar enhancement surrounding a dark center; (B) mucin deposits (white arrows) with lighter areas underneath (blue asterisk) next to a dissolving hair follicle (yellow asterisk).

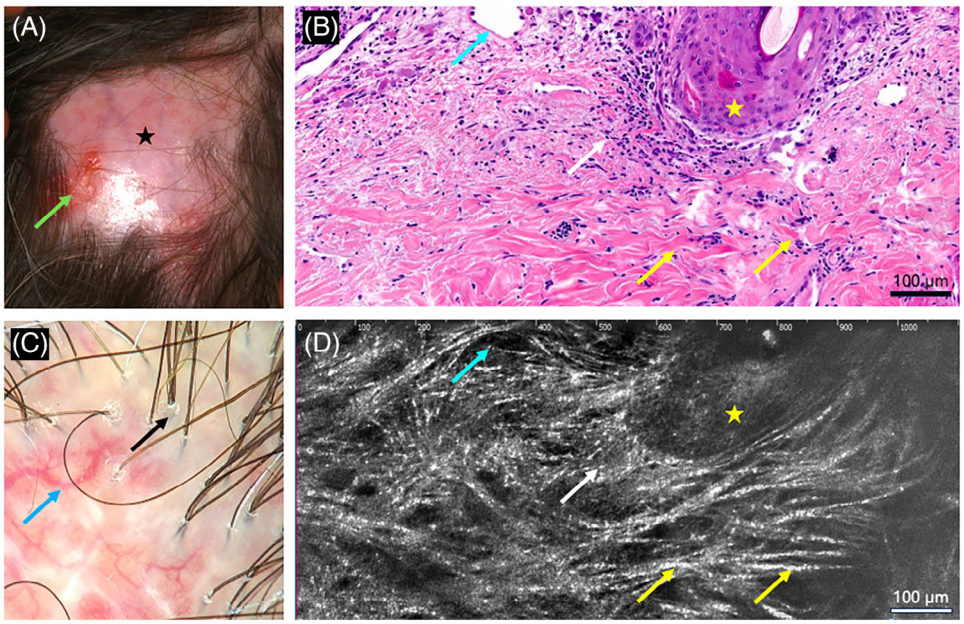

FIGURE 2 (A) Clinical picture with scarring (black asterisk) and erythematous plaque (green arrow) on the scalp of a patient with CDLE; (B) HE-stained section: hair follicle (yellow asterisk) with dermal perifollicular inflammatory cells (white arrow), dermal sclerosis (yellow arrows), and blood vessels (blue arrow); (C) trichoscopic image (×20): perifollicular erythema (blue arrow) and perifollicular cast (black arrow); (D) horizontal LC-OCT image: hair follicle (yellow asterisk), dermal inflammatory cells (white arrow), hyperreflective collagen bundles (yellow arrows), and blood vessels (blue arrow).

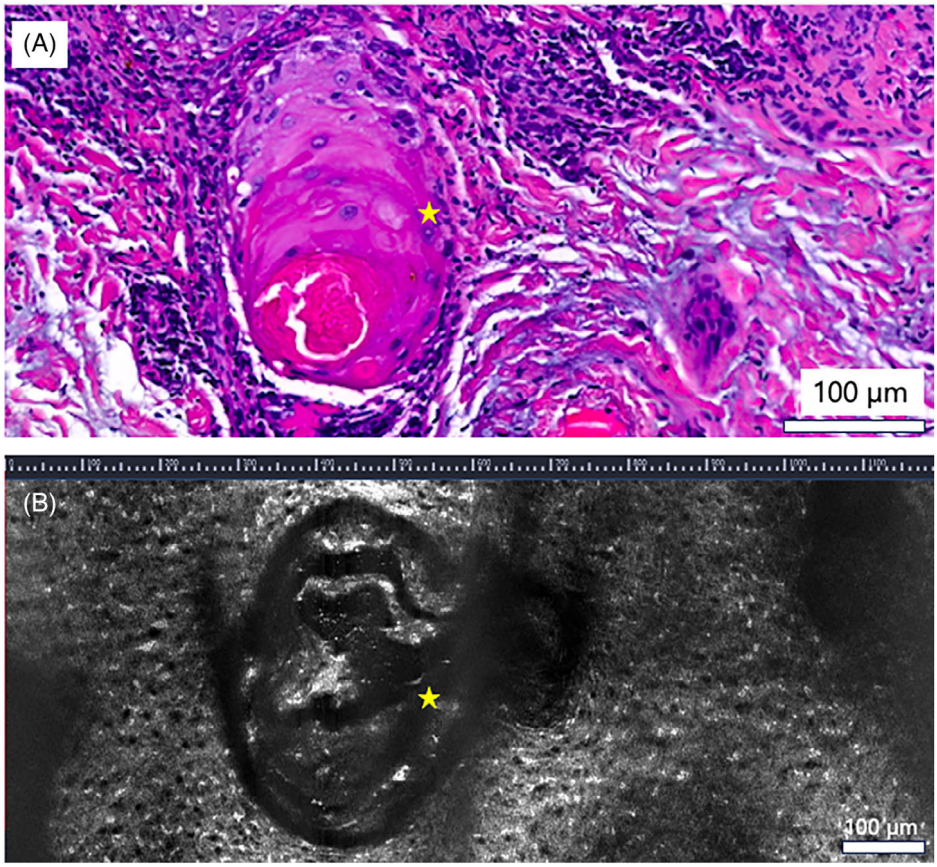

FIGURE 3 (A) HE-stained section of a follicular remnant containing laminar keratin; (B) horizontal LC-OCT image at the same level showing a roundish area filled with highly refractive amorphous material. LC-OCT, line-field confocal optical coherence tomography.

TABLE 3 Incidence of the seven LC-OCT criteria.

Patient	Hyperreflective collagen bundles	No rimming of the papillae	Follicular remnants	Lymphocytic interface dermatits	Basal cell vacuolisation	Melanophages	Mucin deposits
1	*x*	*x*	*x*	*x*	*x* + SRC	*x*	*x*
2	*x*		*x*	*x*	*x* + SRC		
3	*x*	*x*	*x*	*x*	*x*		*x*
4	*x*	*x*	*x*	*x*	*x* + SRC		*x*
5	*x*	*x*	*x*	*x*	*x*	*x*	*x*
6	*x*	*x*		*x*		*x*	*x*
7	*x*	*x*		*x*	*x* + SRC	*x*	*x*
8		*x* partial	*x*	*x*			
9	*x*	*x*		*x*	*x*	*x*	*x*
10	*x*	*x*	*x*	*x*	*x* + SRC	*x*	*x*
11	*x*	*x*		*x*	*x* + SRC	*x*	*x*
12	*x*	*x*	*x*	*x*	*x* + SRC		*x*
13	*x*	*x* partial	*x*		*x* + SRC		*x*
14	*x*	*x*	*x*	*x*	*x* + SRC	*x*	*x*
15	*x*	*x*	*x*	*x*	*x* + SRC	*x*	*x*
Incidence	93.3%	93.3%	73.3%	93.3%	86.7%, SRC 66.7%	60.0%	86.7%

Note: x, criterion present.
Abbreviation: SRC, signet ring-like cells.

On the other hand, the LC-OCT images of scarring alopecia in LPP and folliculitis decalvans published by Rudnicka et al. do not show the collagen fibers appearing as hyperreflective as in our images. This suggests that the explanation for the impressively bright fibers is due to the pathomechanism of LE and is, therefore, specific to LE.

A typical histopathologic feature of LE is mucin deposition in the dermis. According to Palmisano et al., mucin appears on LC-OCT as well-shaped dark linear silhouettes in the dermis containing amorphous material associated with brighter undefined areas. In correlation with these findings and histopathology, we were able to identify these mucin deposits in the dermis in 13/15 (86.7%) of our patients on LC-OCT. Furthermore, in the report by Palmisano et al., the areas below the mucin deposits appear brighter, which is consistent with the hyperreflective collagen fibers in our patients, as these are located directly below and between the mucin deposits in our images.

Amorphous material such as mucin behaves hyporefractively in imaging techniques such as LC-OCT, allowing more light to reach the underlying structures.[26,28] Since collagen fibers behave hyperrefractilvely in LC-OCT,[29] this, combined with the increased light signal, could explain the bright glow of the fibers.

Another explanation could be that the mucin deposits compress the surrounding collagen fibers, resulting in a mass effect. This effect has been described in studies of basal cell carcinoma using LC-OCT.[26,29]

Even if the reason for the hyperreflective collagen bundles cannot be proven, this feature is a helpful tool in diagnosing scalp LE due to its impressive appearance and, thus, potentially easy recognition even by inexperienced examiners. Another reason for the usefulness of this feature in diagnosis is that the presence of collagen bundles is not as sensitive to anti-inflammatory therapy and, therefore, may be a helpful differential diagnostic feature in patients undergoing therapy and in later stages of the disease when even histopathology often does not allow a precise diagnosis.

The next step in investigating LC-OCT for the diagnosis and differential diagnosis of cicatricial alopecia due to LE is, therefore, a blinded comparative study with other forms of cicatricial alopecia, in particular LPP.

6 | LIMITATIONS

Even though LC-OCT has a greater penetration depth than RCM and some features can, therefore, be imaged more clearly, the physically limited penetration depth of LC-OCT up to 500 μm is the greatest limitation of the device. As a result, deeper structures that might be the source of additional information cannot be evaluated. Furthermore, in contrast to histology, it is often not possible to differentiate between cell types due to the lack of different staining. This can only be done based on the size and sometimes the shape of the cells. In addition, DIF staining, which is very helpful in diagnosing LE, cannot be performed compared to histology. A limitation of our study lies in the different durations of disease and therapy of the patients, which can influence the presence of the existing criteria.

7 | CONCLUSION

As only clinical, histologic, and RCM features have been defined for LE to date, this is, to our knowledge, the first description of LE using LC-OCT. In conclusion, LC-OCT can noninvasively detect characteristic criteria for scarring alopecia in LE such as lymphocytic and vacuolar interface dermatitis of cicatricial alopecia due to LE in real time.

In particular, the presence of hyperreflective collagen fibers appears to be a characteristic, easily recognizable feature that may facilitate differential diagnosis with other forms of cicatricial alopecia. This needs to be further investigated in future studies.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ETHICS STATEMENT

The study was approved by the Ethics Committee of the LMU Munich (Project-No: 17-0699). Patients underwent all examinations after the signed informed consent.

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