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A STUDY OF TOPOGRAPHIC AND PHENOTYPIC CHARACTERISTICS OF NORMAL SKIN IN OPTICAL COHERENCE TOMOGRAPHY

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Optical coherence tomography (OCT) is a non-invasive in vivo imaging technique with cellular resolution. OCT provides a cross-sectional evaluation of the epidermis and dermis and allows in vivo imaging of normal skin as well as that of different cutaneous lesions. The aim of this study was to evaluate OCT for its ability to identify architectural patterns and cytological features of normal skin in different body sites and in various skin phenotypes, as a basis for further analysis and interpretation of cutaneous pathology. We analyzed several samples of normal skin, with different locations (head and neck, trunk, limbs, palms and soles), from 25 patients, with light and medium cutaneous tones, describing the visibility of different skin structures as the corneous layer and epidermis, dermo-epidermal junction, dermis. Results showed that there are apparent differences in the ability of structural imaging of normal skin between cutaneous sites, also highlighting several limitations of OCT evaluation in special locations such as palms and soles.

Key words: optical coherence tomography, epidermis, dermis, skin imaging

INTRODUCTION

In dermatology, there are currently several imaging techniques that are being studied and used: ultrasound, dermoscopy, confocal laser scanning microscopy, magnetic resonance imaging, multiphoton microscopy and optical coherence tomography (OCT)[1]. These techniques offer examination of the skin with a greater level of magnification than previously and add functional information[2].

All of these imaging techniques are noninvasive and allow in vivo visualization of various pigmented, non-pigmented tumoral, or non-tumoral skin lesions. They have many advantages, including the lack of any type of adverse effects, are not time-consuming and do not require expensive consumables, thus making them appropriate for repeated examination in the same patient. Regarding the disadvantages, they can't offer an overall suitable resolution or penetration depths to reveal the morphology of the single cells, like the histopathological examination does, which is still considered the gold standard[3].

The appearance of healthy skin images at different anatomical sites is of great importance, in particular in order to understand and interpret images of pathological processes[4].

OCT technology is a non-invasive imaging technique that allows real-time cross-sectional evaluation of biological tissues. The principle of OCT imaging is analogous to that of ultrasound imaging, based on the interference of infrared radiation and living tissues, that allows at high-resolution, 2- or 3-dimensional, crosssectional visualization of microstructural morphology of tissues[5]. It measures the intensity of reflection/backscatter of infrared light from the skin. As optical echoes cannot be measured directly because of the high velocity of light, OCT is therefore based on low-coherence interferometry that correlates reflected/backscattered light from tissue with light that has travelled a known reference path[6].

The image data are displayed by assigning grey scales to each reflection according to the measured signal strength. The light source of the OCT system is a broadband halogen lamp with patented filtered technology, with a center wavelength of 1,300 nm. The resolution is 3 microns in all dimensions. Imaging modes are: en face mode with 1,6x1,8mm/512x640 pixels; slice mode with 1,8x1mm and 3D mode 1,6x1,8x1mm[7]. In comparison to other imaging tools such as in vivo confocal microscopy, OCT has an up to 7 times deeper penetration depth, a much wider field of view, and provides cross-sectional imaging but has a lower resolution; compared to ultrasonography, OCT has an up to 50 times higher resolution at the cost of a lower penetration depth[8]. The aim of this study was to describe normal skin morphology using OCT.

MATERIAL AND METHODS

25 healthy volunteers were enrolled in the study, after signing the informed consent form. This study adhered to the Declaration of Helsinki and was performed in a

university setting (Elias Emergency University Hospital, Bucharest, Romania).

Volunteers were recruited and examined during August 2015-October 2015. OCT imaging was done in 9 predetermined anatomical sites: forehead, cheek, chin, chest, posterior trunk, anterior forearm, calf, palms and soles. Exclusion criteria were history of any dermatological disorder, heavy smoking (>20 cigarettes/day) and significant ultraviolet (UV) exposure within 3 months prior to the study.

There were 25 Caucasian patients with Fitzpatrick skin types I—III, light and medium phenotypes, including 2 subgroups of subjects - aged between 20 and 40 years, and aged between 41 and 70 years.

The OCT system used in this study was developed by AGFA Healthcare. The hand-held OCT probe is quite large and heavy but relatively easy to handle, and it is applied directly to the skin, using ultrasound gel to improve image quality. The scanning lasts a few seconds and provides 2 dimensional images -en face and slice mode or 3 dimensional images – as selected with the foot control panel. At the beginning of the study, we performed repeatability measurements under standardized conditions (room temperature- 22 degrees C, acclimatizing for 10 min) in the first 10 of the study subjects. Then, two subsequent OCT measurements per site were performed, under standardized conditions, on 9 different anatomical sites in lying position in all subjects.

RESULTS AND DISCUSSIONS

25 healthy volunteers (18 females and 7 males) were included and analyzed in this study, with a mean age of 40 years, and a 2,57:1 female:male ratio. There were 2 subgroups of patients – 15 volunteers aged between 20-40 years and 10 volunteers aged between 41-70 years. All patients were Caucasians, 9 with Fitzpatrick skin phototype II, 12 with phototype III and 4 with phototype I. The skin tone was light in 13 patients and medium in 12 patients (Table 1). Visualization of the skin appendages was also possible in several cases (Table2).

Healthy Volunteers	Gender	Age (years)	Fitzpatrick phototype	Skin tone
1.	F	27	I	light
2.	F	29	I	light
3.	F	35	II	light
4.	F	28	III	medium
5.	M	27	II	light
6.	F	25	II	light
7.	F	29	III	medium
8.	M	37	II	light
9.	F	38	I	light
10.	M	34	III	medium
11.	F	25	I	light
12.	M	29	III	medium
13.	F	28	II	light
14.	F	35	II	light
15.	F	34	III	medium
16.	M	64	II	light
17.	F	59	III	medium
18.	M	65	III	medium
19.	F	50	II	light
20.	F	58	III	medium
21.	F	43	II	light
22.	M	57	III	medium
23.	F	56	III	medium
24.	F	44	III	medium
25.	F	47	III	medium

Table 1. In most of the images, OCT reflected the well-known layered architecture of the skin, represented by the stratum corneum, the epidermis, the demarcation between epidermis and dermis and the dermis at different depths, depending on the site.

ANATOMIC SITES		OCT CHARACTERISTICS			
	Visible corneous layer	**Clear DEJ**	**Visible papillary/reticular dermis demarcation**	**Visible blood vessels in the dermis**	**Adnexal structure (hair follicles)**
Forehead	7,12%	50,00%	14,28%	21,41%	23,80%
Cheek	7,12%	71,42%	28,57%	14,28%	42,91%
Chin	7,12%	71,42%	42,91%	14,28%	38,16%
Chest	7,12%	86,20%	28,57%	21,41%	42,91%
Posterior trunk	14,28%	57,14%	35,71%	42,91%	76,33%
Anterior forearm	14,28%	71,42%	35,71%	57,14%	86,20%
Calf	21,41%	78,74%	35,71%	93,45%	92,59%
Palms	100%	93,45%	28,57%	7,12%	No visible adnexal structures
Soles	93,45%	71,42%	21,41%	No visible vessels	No visibile adnexal structures

Table 2. Visualization of the skin appendages.

The stratum corneum appears as a dense, well-defined, homogenous, low-scattering band, practically a dark grey layer located over the epidermis[9,10]. The skin of the palms and soles offered a very clear image of the stratum corneum in almost all 25 patients (Figure 1) In other anatomical sites where the horny layer was visible, there was a clear border between the stratum corneum and the living epidermis, but this was noticed in a small percentage of cases varying between7-12-21,41%.

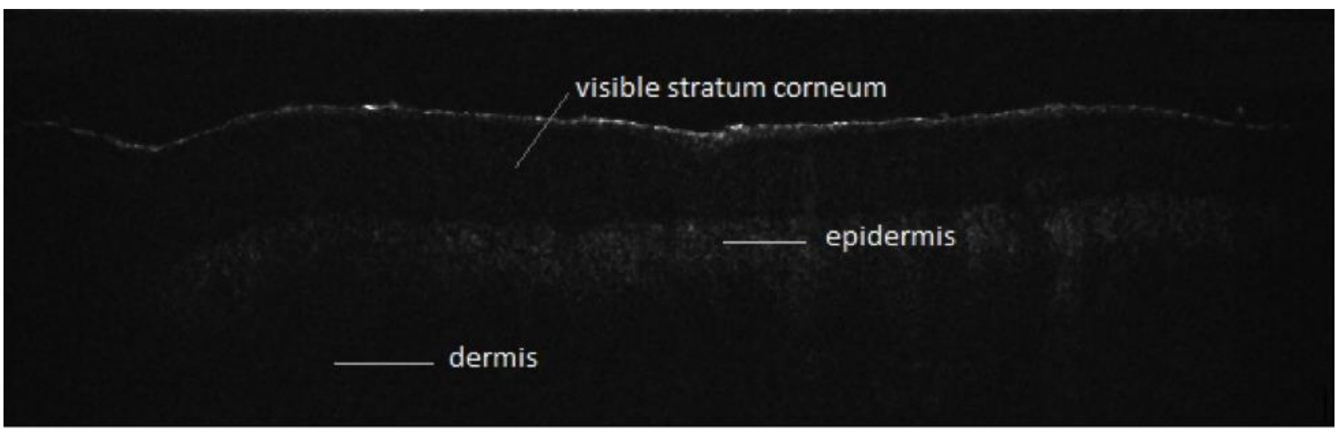

Figure 1. OCT scan of palm region

At other skin sites on the face and on the body, were the stratum corneum is not thick enough to be visualized independently, the epidermis is the first distinguishable layer in the OCT images. The face (e.g. forehead, cheek) region has a thinner epidermis, compared to other regions (Figure 2).

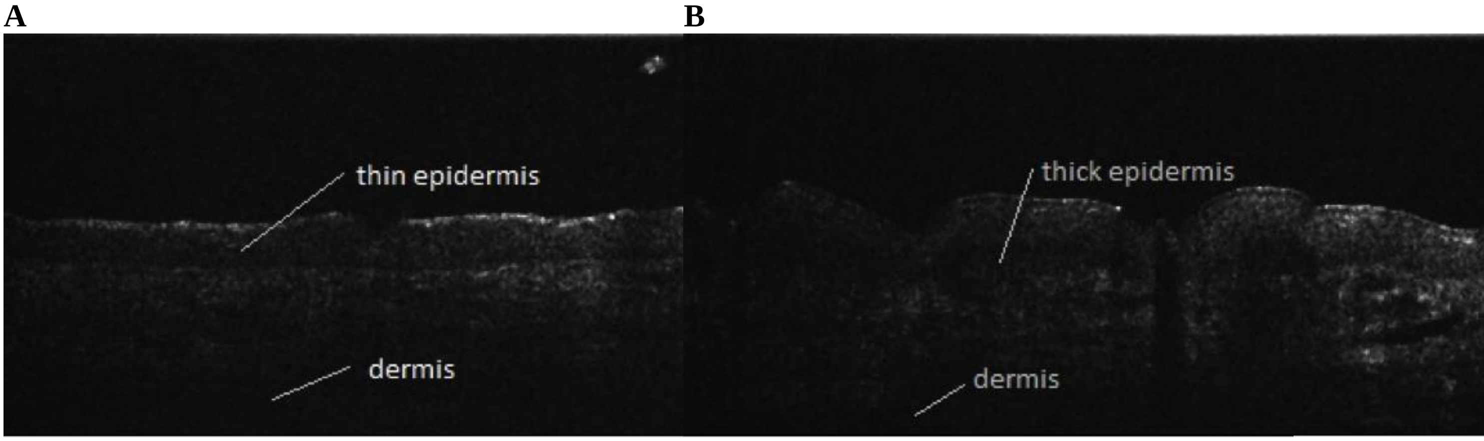

Figure 2. (A) OCT scan face region, (B) OCT scan calf region

The epidermis shows less intense signals than the dermis and it is represented by the layer located between the entrance signal of the OCT and the first increase in image intensity, which is the dermo-epidermal junction. DEJ was visible in most of the analyzed cases: 50-93,45%, with the lowest rate on the face and the highest visibility on the palms (Figure 3).

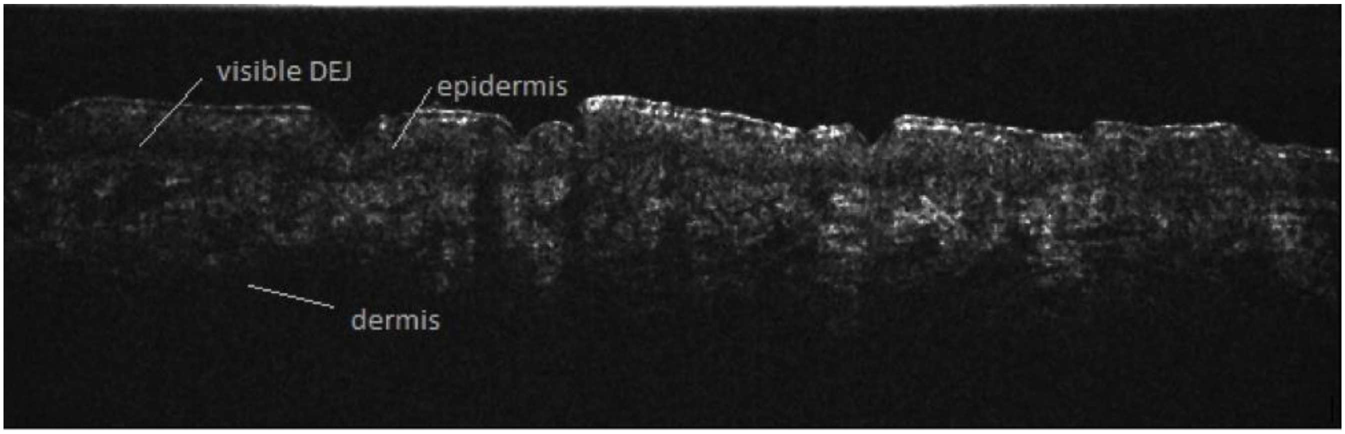

Figure 3. OCT scan of anterior forearm region

A precise demarcation between the two layers of the dermis, the superficial papillary region and the deep thicker reticular dermis can't be distinguishable, as it would be in a high resolution histopathological image[11]. The signal intensity of the dermis is predominantly produced by the extracellular matrix, composed of collagen fibers. The deeper the dermis the intensity of the signal decreases, and the captured images appear progressively darker. In the areas where the epidermis is thinner (e. g forehead, cheek, anterior forearm) the visibility is improved and we can see a larger section of the deep dermis.

However, our results showed a visible papillary/reticular dermis demarcation in 14, 28-42,91% of cases (Figure 4).

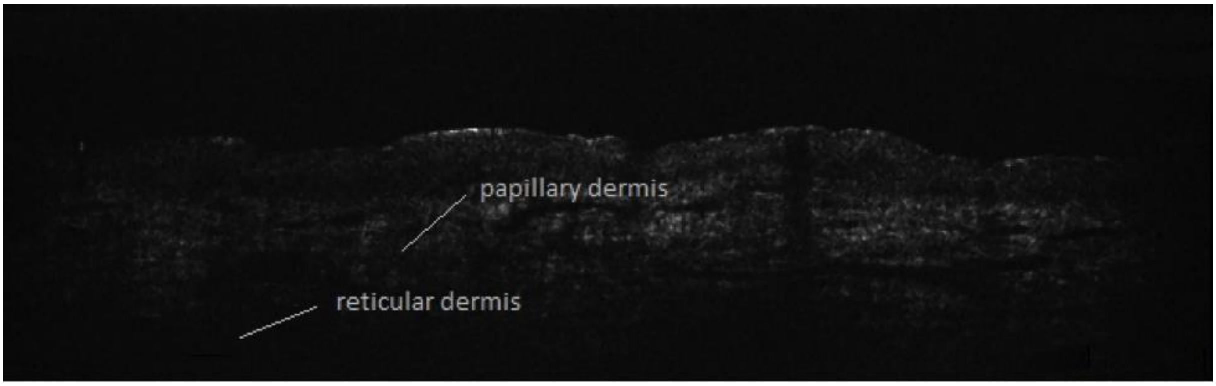

Figure 4. OCT scan anterior forearm- visible demarcation papillary/reticular dermis

The high resolution of this technique permits the visualization throughout the dermis, of different structures like blood vessels and adnexal structures. Vessels appear like elongated uneven black cavities, because they are captured in transversal, longitudinal or oblique section, under different angles[11,12]. Blood vessels with different diameters were especially notable in the calf (93,45%) anterior forearm (57,14%), and posterior trunk (42,91%) regions. (Figure 5). As for other regions with thick epidermis, like palms visible vessels were encountered in only a few cases (7,12%).

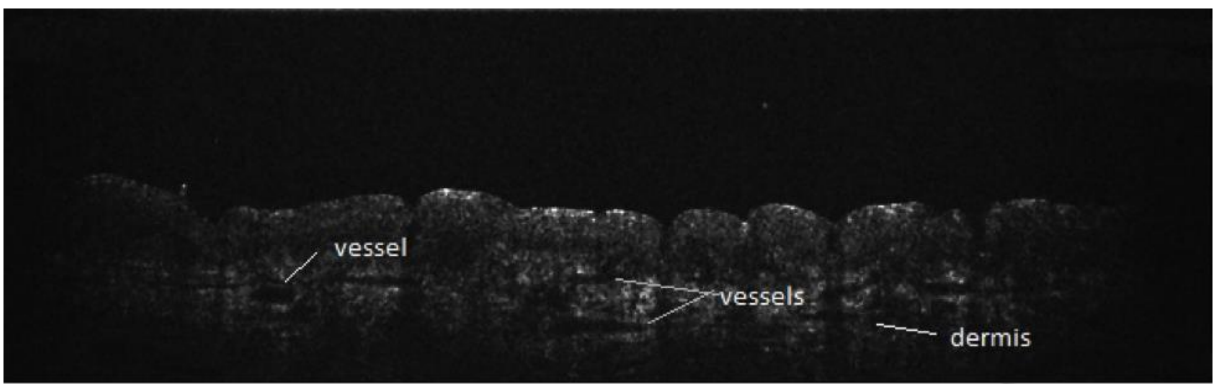

Figure 5. OCT scan of calf region – visible vessels

All three types of imaging modes of the OCT system allows also the visualization of hair follicles in most of the anatomical regions; although small-sized follicles were recognizable in various regions, a higher rate was notable in the calf (92,59%), anterior forearm (86,20%) and posterior trunk (76,33%) areas, compared to other regions of the body, like the forehead (23,80%). Because the hair follicles create a dark shadow, this could impair the visibility of the cutaneous structures in areas with a high number of such appendices (Figure 6).

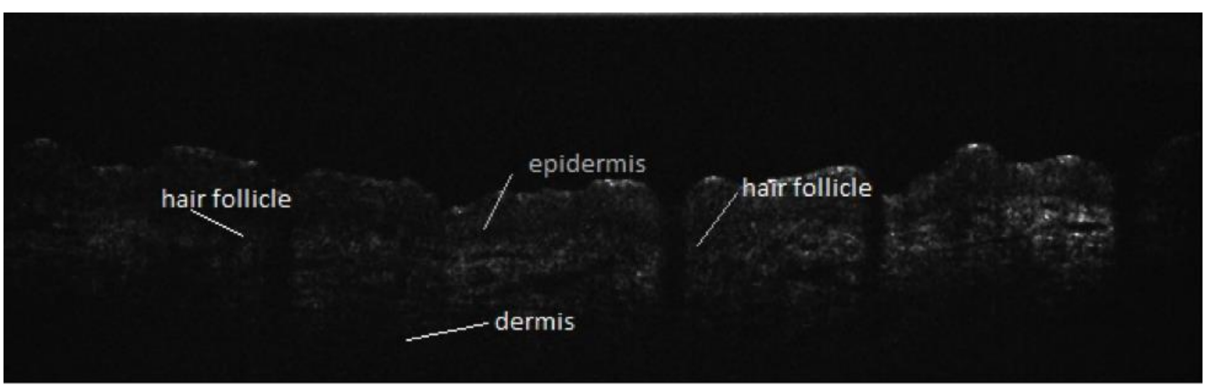

Figure 6. OCT scan of calf region –visible hair follicles

Regarding the aspect of the identifiable structures of the normal skin on OCT, there wasn't a detectable difference between the patients with medium or light skin tones, as it may be possible in other non-invasive imaging techniques like confocal microscopy where due to higher percentage of melanin existing in darker skin tones, well defined pictures are created in the detriment of lighter ones[13, 14].

As in previous studies, our results showed that OCT technique proved to be capable to present morphological features of the epidermis and superficial dermis in almost all analyzed cases.

Also, in most of the images skin appendages including hair follicles were identified by OCT.

While other research showed visible eccrine ducts or sebaceous glands, we were not able to identify these kind of structures in our images, perhaps as a consequence of the system's resolution[8,12,15,16].

OCT images presented some regional differences especially in terms of apparent epidermal thickness and visibility of the dermo-epidermal junction and vessels; thick epidermis was visible in all images of the palms and soles from almost all 25 patients; also the calf and the posterior trunk are other anatomical sites were thick epidermis was distinguishable in most of the patients.

These results are in line with previous studies that showed a clearly visible stratum corneum in the glabrous skin of the palms and increased vascularity in the skin of arms and calves[8,12,15,16].

The uniqueness of the study consists of the fact that not only does the research analyse the structure of the tissue imaged via the OCT, but also offers a direct comparison with different patterns identified in various parts of the body; hence adding to previous papers and at the same time contributing as solid reference for the future study of the pathological tissue.

CONCLUSIONS

The OCT is a reliable non- invasive imaging method that enables the visualization of skin structures, bringing cutaneous in vivo imaging closer to the gold standard represented by the histopathological examination. Analyzing the aspect of the normal skin and the characteristics of different body regions will represent a basis for further study and interpretation of cutaneous pathology.

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