Nail Fold Capillaroscopy: Normal Findings in Children and Adolescents

Maria Teresa R.A. Terreri, Luis Eduardo C. Andrade, Mario Luis Puccinelli, Maria Odete E. Hilário, and José Goldenberg

Objectives: **Capillaroscopy is a simple diagnostic method that permits noninvasive in vivo study of the capillary network. Studies designed to standardize capillary normality in children are limited. This article presents the capillaroscopic findings in healthy children and adolescents, thus making the application of this methodology viable for patients in this age range.**

Material and Methods: **Healthy children were recruited from a private elementary school and junior high school. Nail fold capillaroscopy was performed using a stereomicroscope at 16 times magnification, addressing the following parameters: capillary morphology, capillary enlargement, devascularization, microhemorrhage, and subpapillary venous plexus visibility (PVS). These parameters were related to age, sex, ethnicity, and local periungal conditions.**

Results: **The sample comprised 329 individuals with mean age of 8.2 years. We observed atypical capillary morphology in 118 of the studied cases (36%), mainly bizarre capillaries in 90 (27%), meandering capillaries in 32 (10%), and bushy capillaries in 20 (6%). The enlarged capillary phenomenon was uncommon, being observed in 30 cases (9%). The number of capillaries per millimeter varied from five to nine. Deletion areas were detected in only seven individuals (2%). The subpapillary venous plexus was not visualized in 13 (4%) cases. Younger children presented higher PVS scores and fewer capillaries/mm as compared with older children. PVS scores were lower in males and in nonwhite children. Other variables were not associated with sex or ethnicity.**

Conclusions: **The normal nail fold capillary network in children resembles that observed in adults with some differences, such as a lower number of loops per millimeter, a higher PVS score, and a higher frequency of atypical loops. This information is important for the diagnostic evaluation of children in the context of autoimmune rheumatic diseases.**

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INDEX WORDS: Nail fold capillaroscopy; scleroderma; microcirculation; Raynaud's phenomenon; children.

CAPILLAROSCOPY IS A SIMPLE diagnostic method that permits noninvasive in vivo study of the capillary network. The nail fold region is convenient for microscopic examination of the capillary network because of its immediate access and because the capillaries are placed horizontally, allowing them to be visualized on their long axis (1, 2). The first nail fold capillaroscopy carried out on

From the Division of Allergy, Clinical Immunology and Rheumatology, the Department of Pediatrics and Division of Rheumatology, Department of Medicine, Sao Paulo Federal University, Escola Paulista de Medicina, Sao Paulo, Brazil.

Maria Teresa R.A. Terreri, MD, PhD: *Assistant Researcher, Division of Allergy, Clinical Immunology and Rheumatology, Department of Pediatrics;* Luis Eduardo C. Andrade, MD, PhD: *Associate Professor, Division of Rheumatology, Department of Medicine;* Mario Luis Puccinelli, MD, PhD: *Associate Professor, Division of Rheumatology, Department of Medicine;* Maria Odete E. Hilário, MD, PhD: *Associate Professor, Division of Allergy, Immunology and Rheumatology, Department of Pediatrics;* José Goldenberg, MD, PhD: *Associate Professor, Division of Allergy, Immunology and Rheumatology, Department of Pediatrics.*

Address reprint requests to Maria Teresa Terreri, MD, PhD, Division of Allergy, Clinical Immunology and Rheumatology, Department of Pediatrics, UNIFESP-EPM, Rua Loefgreen 2381-ap. 141, CEP-04040-004, Sao Paulo, SP. Brazil.

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humans apparently dates from 1911, by Lombard (3). Since then, several technical modifications have been accomplished and considerable experience has been gained with the method. Nail fold capillaroscopy has become an excellent means for capillary examination in different medical specialities, such as dermatology, rheumatology, and angiology (4-10). The accumulated experience in the literature, the technological perfecting of microscopes and light sources, appropriate scientific documentation, and development of more objective and reliable parameters have resulted in this method being used routinely in several centers, especially in the diagnosis of diseases having capillary involvement.

A few studies have been performed in children with rheumatic fever, dermato/polymyositis, and scleroderma (11-14). However, studies planned to standardize capillary normality in children are limited. Mayer (15), in 1921, reported that the nail fold capillary network develops rapidly, attaining its final aspect during the first 3 months of life, and that, after that age, no major modification is observed. However, it should be emphasized that in this work Mayer made reference only to the subpapillary venous plexus and did not focus on the distal capillary row. Marico (16), in 1964, compared children with healthy adults and concluded that the subpapillary venous plexus visibility score (PVS) is greater in younger children. These findings were confirmed by Whitson and Jones (17). All studies on the normal capillaroscopic pattern in children had an eminently morphological and subjective connotation, with emphasis on the subpapillary venous plexus. However, it has been shown that changes in morphology, size, and distribution pattern of capillary loops at the distal nail fold capillary row are more informative than the study of the subpapillary venous plexus. Moreover, prior studies in adults have shown the advantage of an analytical and objective approach of the microvascular phenomena perceptible by capillaroscopy (18). Therefore, the current work was designed to study the capillaroscopic standard in healthy children and adolescents, focusing on the distal row capillary loops and the venous plexus using an analytical and objective approach.

PATIENTS AND METHODS

The sample comprised 329 children and adolescents of both sexes, without any chronic or acute illness and not using any medication, recruited from private schools. Informed consent for nail fold examination was given by the children or their parents. Given the difficulty of ethnic classification in our country, the children were simply classified as white and nonwhite.

The techniques of microscopy, illumination, skin diaphanization, and photomicrography were similar to those described by Marico (19). Briefly, a microscope with 10× or 16× magnification was equipped with a graduated ruler attached to the right eyepiece, to allow counting the number of capillary loops per millimeter in the distal row. Epi-illumination was provided by incandescent tungsten lamp with a green filter, so as to better highlight the capillaries against the tissue background. Skin diaphanization was obtained with a transparent oleaginous agent.

Naked eye examination of the nail fold region was accomplished to detect trauma, wounds, or swelling, as well as to determine the nail fold color. A film of oil was applied to the nail fold region just before the microscopic examination. All examinations were performed by the same observer (M.T.T.). The methodology used for analysis and interpretation of the various microvascular phenomena was that proposed by Andrade et al (18), which focuses on microhemorrhage, capillary morphological abnormalities, capillary enlargement, capillary devascularization, and visibility of the subpapillary venous plexus. Other microscopic parameters included background color, general capillary visibility, and dominant morphological pattern.

Microhemorrhage was evaluated by counting the number of micropetechiae, represented by red or brown amorphous structures. These were classified according to their distribution pattern: focal pattern, when grouped within limited areas; and diffuse pattern, when distributed throughout the periungal region.

The phenomenon of capillary enlargement was registered as enlarged capillaries and megacapillaries (19). Since wide field nail fold capillaroscopy is not intended to measure individual capillary loops, capillary enlargement was considered in relation to neighbor normal loops in the same individual. Enlarged capillaries were defined as those having four or more times the width of normal neighbor loops in the three limbs, ascendant, transition, and descendant. Megacapillaries were defined as aneurysm-looking loops with limbs about 10 times or

more the width of normal neighbor loops. Morphological anomalies were classified into three types: meandering, bushy, and bizarre capillaries. Meandering capillaries were defined as tortuous loops where the limbs were intertwined and crossed on themselves. Bushy capillaries were defined as those with small branches in different directions. Bizarre capillaries were those presenting with atypical structure, although not conforming to the types just described.

Capillary devascularization was estimated in two ways: the number of capillary loops per millimeter allowed the detection of diffuse devascularization, and the deletion score measured focal devascularization. The number of capillaries per millimeter was counted through the eyepiece with a graduated ruler. The final figure was the average of the linear density of capillaries in the fingers observed. Deletion areas were defined as the lack of two or more successive capillaries, or avascular areas measuring more than 3 mm in extent. This system was adapted from Lee et al (20), and is graded as follows:

Grade 0: No deletion areas
Grade 1: One or two discontinuous deletion areas
Grade 2: More than two discontinuous deletion areas
Grade 3: Extensive and confluent deletion areas

The final degree corresponded to the mean of the figures ascribed for each finger.

Subpapillary venous plexus visibility was classified according to the score proposed by Wertheimer and Wertheimer (21), as follows:

Grade 0: Absence of plexus visibility; capillaries only are seen
Grade 1: Doubtful visibility or the occasional presence of a venule parallel to the nail fold
Grade 2: Plexus visible only in restricted areas, or throughout the width of the finger but only close to the distal row of capillaries
Grade 3: Plexus visible throughout the width of the finger but not extending proximally, or if extending proximally, not generalized throughout the width of the finger
Grade 4: Plexus visible throughout the width of the finger and extending proximally.

The final PVS was given by the sum of the values obtained in the 10 fingers. Based on previous experience, the visibility of the subpapillary venous plexus was classified as high (PVS > 20), medium ($5 <$ PVS ≤ 20), and low (PVS ≤ 5) (18).

Background color of the subepithelial tissue between capillaries was classified as light or dark. General visibility of the capillaries was graded according to the subjective ease or difficulty with which the capillaries were observed, namely, good, regular, or bad (including nil visibility in the latter). The dominant morphological pattern was defined as the most prevalent morphological loop variant. Common morphological variants found in the general population are open, tortuous, and crossed (18). When the capillaries were either too small or the branches too close to each other, making morphological definition impossible, the pattern was reported as undefined. Cases in which only the top of the capillary loop was visible were denoted as a cuticulitis pattern, given the similarity to what is observed after manicure excision of the cuticle. Finally, the dominant pattern was reported as mixed when more than one variant co-dominated.

The chi-square test or exact Fisher's test, when appropriate, were used to analyze possible association between discontinuous variables. The Mann-Whitney test was employed for comparison of continuous variables between two independent groups. Comparisons involving three or more groups were evaluated by the Kruskal-Wallis test. The limit for rejection of the null hypothesis was established at 0.05 (5%), with significant values being marked with an asterisk.

RESULTS

Demographic and Capillaroscopic Features of the Sample

The sample comprised 329 individuals with mean age of 8.2 years, ranging from 2.1 to 16.7 years, being 51% girls and 66% Caucasian. Two hundred fifty-four children (77%) presented light periungal color, and 75 (23%) presented dark periungal color. Microhemorrhage was observed in 67 (20%), with a mean of 5.3 micropetechiae per individual (range, 1 to 51). In most of these cases (92%), the micropetechiae depicted a focal distribution pattern. We observed atypical capillary morphology in 118 (36%); mainly bizarre capillaries, verified in 90 individuals (27%), meandering capillaries in 32 individuals (10%), and bushy capillaries in 20 (6%). Figure 1 illustrates bushy capillaries and meandering capillaries, respectively. Apart from these morphological variants, several children

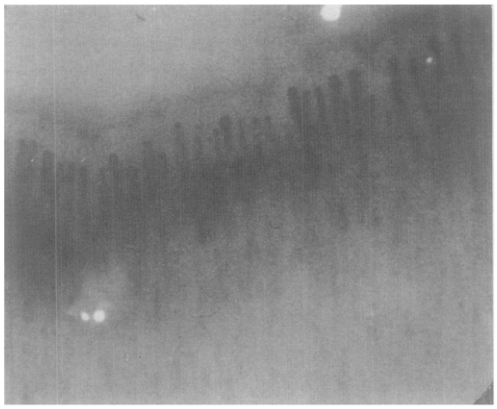

Fig 1. Open-loop dominant morphological pattern: Most loops display non–crossing over limbs with minimal tortuosity.

presented a special capillary morphology characterized by a bifurcated loop with two branches, designated open-ended spanner loop. Capillary enlargement was rare, being registered in 30 cases (9%). When present, the number of enlarged capillaries varied from one to four per individual. Megacapillaries were not observed in the current series.

The linear density of capillaries varied from 4.9 to 9.2 loops/mm, with a mean of 7.1 loops/mm and standard deviation of 0.8. Deletion areas were detected in seven individuals (2%): five with grade 1 deletion, one with grade 2, and one with grade 3.

The subpapillary venous plexus was not visualized in 13 (4%) of the individuals. Ninety-seven cases (30%) presented high visibility of the plexus (PVS $>$ 20), 164 (50%) presented medium plexus visibility (5 $<$ PVS $\leq$ 20), and 68 children (20%) had low plexus visibility (PVS $\leq$ 5).

The background color was light in 256 cases (78%) and dark in 73 (22%). The general capillary visibility was good in 227 individuals (69%), regular in 79 (24%), and bad in 23 cases (7%). The dominant morphological pattern was open in 257 cases (78%), cuticulitis in two (1%), tortuous in two (1%), mixed in 64 (19%), and undefined in four children (1%). Figure 2 depicts an example of open-loop dominant morphological pattern.

Capillaroscopic Parameters in Relation to Gender, Ethnic Classification, and Age

The periungal color and the presence of micropetechiae were not associated with ethnic classification or gender. There was no statistically significant difference in the number of capillaries per millimeter or in the overall frequency of atypical loops observed between both genders and ethnic groups (Table 1). Conversely, the frequency of bizarre capillaries was greater in girls than in boys, the difference being statistically significant in white subjects and almost significant among nonwhite children. The frequency of enlarged capillaries between genders was not different in both ethnic groups (Table 1). Furthermore, no statistically significant difference was observed in the fre-

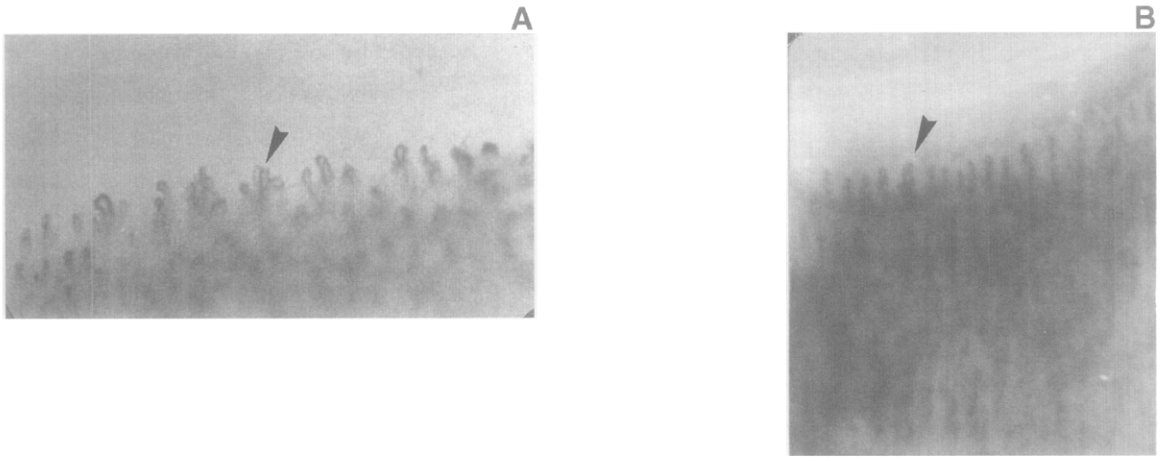

Fig 2. (A) Bushy loop: Several buds are observed sprouting out (arrow); (B) Meandering loop: The limbs are tortuous and convoluted (arrow).

Table 1: Distribution of White and Nonwhite Subjects According to Gender and Capillary Loops/mm, Morphological Anomalies, Bizarre and Enlarged Capillaries, Deletion Score, and PVS

	White		Nonwhite	
	Male	Female	Male	Female
Number of capillary loops/mm (mean and SD)	7.3 ± 0.75	7.3 ± 0.73	6.9 ± 0.74	6.8 ± 0.82
Morphological anomalies (%)	31	42	30	38
Bizarre capillaries (%)	19*	35*	21	34
Enlarged capillaries (%)	6	11	10	10
Deletion areas (%)	3	3	0	0
PVS (mean and SD)	14.3 ± 8.5*	18.6 ± 8.5*	9.4 ± 8.8*	13.8 ± 8.6*

Abbreviations: SD, standard deviation; PVS, plexus visibility score.
*$P < .05$.

quency of deletion areas between genders in both ethnic groups (Table 1). The plexus visibility score presented lower values in boys than in girls and in nonwhite than in the white group, both differences being statistically significant (Table 1).

In comparing the various parameters in relation to age, bizarre capillaries were significantly more frequent at ages older than 10 years in comparison with ages younger than 10 years in the male group (Table 2). In the female group, there was no such difference (35% younger than 10 years and 32% older than 10 years). Age did not influence significantly the incidence of meandering, enlarged, or bushy capillaries (data not shown). Conversely, the number of capillaries per millimeter in the white and nonwhite groups increased progressively with age (Table 3). This trend was explored by analyzing children 10 years of age or younger against those older than 10 years of age. In both white and nonwhite groups, children older than 10 years of age presented a significantly higher number of capillaries/mm than those younger than or equal to 10 years (Mann-Whitney: $Z = 3.36$ and $Z = 3.37$, respectively; critical $Z = 1.96$).

PVS showed a clear-cut trend for higher values in younger children, as depicted in Table 4. However, when we analyzed children 10 years of age or younger against those older than 10 years of age, statistical significance was detected only for white girls (Mann-Whitney: $Z = 2.82$; critical $Z = 1.96$).

Table 2: Distribution of Male Children (White and Nonwhite) Presenting Bizarre Capillaries According to Age

Age Group	Bizarre Capillaries	
	Yes	No
≤10 years	22 (16%)	112 (84%)
>10 years	10 (37%)	17 (63)
Total	32 (20%)	129 (80%)

NOTE. Chi square = 5.99.

DISCUSSION

During the last 30 years, panoramic periungal capillaroscopy (10× to 16× magnification) has been shown to be extremely useful in diagnosing diseases of the scleroderma spectrum, which share microvascular features characterized by disorganization of the capillary distal palisade, represented mainly by devascularization and capillary enlargement. Analytical and objective methodology has been useful for quantitative and reproducible recording of these parameters in adults. In children, the progressive maturation of the microvascular network can be a complicating factor when attempting

Table 3: The Number of Capillary Loops According to Age and Ethnicity

Age-Group (yr)	White		Nonwhite	
	Male	Female	Male	Female
2 (n = 11)	6.4*	6.4	5.9	6.0
3 (n = 31)	6.8	6.5	6.6	6.3
4 (n = 28)	6.9	7.2	6.7	6.4
5 (n = 19)	8.0	7.6	6.8	5.8
6 (n = 29)	7.0	6.9	6.6	6.8
7 (n = 31)	7.1	7.4	6.9	6.2
8 (n = 35)	7.4	7.8	6.7	7.1
9 (n = 50)	7.7	7.5	7.1	7.2
10 (n = 42)	7.3	7.5	6.9	6.6
>10 (n = 51)	7.9	7.6	7.6	7.3

NOTE. Two children could not have the number of loops/mm determined due to poor visibility.
*The values represent the means for each age-group.

Table 4: The Influence of Age and Ethnicity on Plexus Visibility Score (PVS)

Age-Group (yr)	PVS—White		PVS—Nonwhite	
	Male	Female	Male	Female
2 (n = 11)	10.5*	20.0	17.3	21.0
3 (n = 31)	14.2	16.8	9.2	24.0
4 (n = 28)	18.0	19.9	15.5	22.2
5 (n = 19)	14.0	23.6	5.7	9.0
6 (n = 29)	17.9	20.0	10.0	14.4
7 (n = 31)	12.9	17.7	9.4	9.0
8 (n = 36)	14.1	20.8	6.5	13.6
9 (n = 50)	10.7	19.8	10.2	9.5
10 (n = 42)	15.7	17.2	7.2	11.4
>10 (n = 52)	15.1	11.8	7.3	11.7

*The values represent the means for each age-group.

to extrapolate the findings obtained in normal adults. With this in mind, we sought to report the main capillaroscopic parameters in a representative normal pediatric sample. The derived normal standards should be helpful in the interpretation of capillaroscopic findings in children with scleroderma, dermatomyositis, and other autoimmune rheumatic diseases.

Previous studies on the standardization of nail fold capillaroscopy in normal children and adolescents have dealt mostly with the subpapillary PVS (2, 4-6, 9). In this report, we have focused on five microvascular phenomena, namely, microhemorrhage, subpapillary venous plexus visibility, capillary enlargement, morphological abnormalities, and devascularization. Objective and reproducible parameters were sought to describe these phenomena and thus minimize the intrinsic subjectivity of the method. Normal individual variation and the influence of variables, such as gender, age, ethnicity, and local conditions of the periungal region, were explored in relation to each capillaroscopic parameter. To evaluate the possible influence of age on capillaroscopic parameters, we divided the children into approximate age-groups, stratified on an annual basis from 2 to 10 years and one group older than 10 years. Given the considerable racial miscegenation of our population, we simply classified children as white and nonwhite.

The frequency of micropetechiae (20%) was lower than that found in normal adults (60%) (10). The focal distribution of micropetechiae observed in almost all cases in the current series confirmed previous findings of Andrade et al (18), who found only rare cases of diffuse microhemorrhage in 800 normal subjects. Together with the previous observation of diffuse microhemorrhage in some systemic diseases, these findings suggest that the micropetechiae distribution pattern might be of greater relevance than the mere quantification of the individual lesions. As previously suggested, diffuse microhemorrhage is compatible with endogenous endothelial lesions, whereas focal micropetechiae probably originate from daily microtrauma (18).

The PVS is the most frequently described parameter in the pediatric literature regarding nail fold microscopy (21). The visibility of the subpapillary venous plexus was, for many decades, taken to be a sign of capillary immaturity, as was atypical loop morphology (22). There are conflicting opinions as to the age at which capillary maturity is reached. Some studies indicate that the subpapillary vascular network is exuberantly visible in the newborn and in children a few weeks or months old, in whom the periungal capillary loops are not individually discernible (2, 15, 23). In adults, however, the periungal capillaries are well defined, and the subpapillary venous plexus tends to be rudimentary (24, 25). According to Leader (2), the standard adult nail fold capillary network, in which only typical loops are seen, appears between 6 months and 1 year of age. Mayer (15) reports that the standard capillary panorama in the periungal region develops rapidly into its final form during the first 3 months and that no major change takes place thereafter. Plexus visibility in the intermediate years, that is, throughout infancy and adolescence, has not been appropriately defined in the literature. Rondelli and De Matteis (26) found the subpapillary venous plexus still present in older children, but much less frequently as age increased. Although we could not show a statistically significant correlation between PVS and age, there was a definite tendency for higher PVS in younger ages.

The number of capillaries per millimeter appears to be an objective index for evaluating the frequency of capillary loops and of possible devascularization. Within the current sample, children presented lower capillary density than adults. This is important information because devascularization is one of the major objective signs of scleroderma microangiopathy. Only in children older than 10 years of age did we begin to find figures similar to

those observed in normal adults (7 to 12 loops/mm). Two percent of the sample examined presented some evidence of focal devascularization, characterized by the presence of deletion areas, comparable to the 7% frequency in normal adults (18). Also similar to the findings in adults, the few affected children showed a low quantitative expression of devascularization, that is, a deletion index not greater than 0.3. To our knowledge, this is the first study focusing on nail fold microvascular devascularization in normal children.

The interpretation of capillary loop morphology involves a great deal of subjectivity. Atypical capillary morphology occurred in one third of the individuals and included meandering, enlarged, bushy, and bizarre capillaries. Megacapillaries were not found. It is important to emphasize the high frequency of atypical capillaries, especially the bizarre type, in children and adolescents. Bizarre capillaries were observed in 27% of the sample, the most frequent being the open-ended spanner loop. These bizarre capillary forms represent a distinct feature of children's capillary network, because they are not usually observed in normal adults.

The data presented herein provide a framework for the standardization of normal parameters observed in the periungal microvascular structure of children and adolescents. The proposed method is objective and quantitative and should help the reproducibility and widespread applicability of this procedure in the evaluation of children with Raynaud's phenomenon and diseases related to the scleroderma spectrum.

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