Dermatology Practical & Conceptual

Diagnostic Accuracy of Magnified Dermoscopy and Reflectance Confocal Microscopy in Assessing Melanocytic Lesions

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ABSTRACT **Introduction:** Magnified dermoscopy (MD), or optical super-high magnification dermoscopy, is an emerging technique in dermatology.
Objectives: The study aimed to evaluate the distribution of conventional dermoscopy, MD, and reflectance confocal microscopy (RCM) features in dermoscopically equivocal pigmented lesions and to estimate their diagnostic accuracy.
Methods: A retrospective analysis of conventional dermoscopic (20x), MD (400x), and RCM images of dermoscopically equivocal pigmented lesions, diagnosed as either nevi or melanoma, was performed. Distribution of features, sensitivity, and specificity for dermoscopy, MD, RCM, and a combination of these last two with conventional dermoscopy was estimated.
Results: A total of 74 nevi and 20 melanomas were included in the analysis. A positive correlation was observed between seven-point checklist in conventional dermoscopy and the diagnosis of melanoma. With MD, a significant correlation between dots, non-edged papillae, and melanoma was observed, but the technique did not have a significant impact on diagnostic accuracy as compared to traditional dermoscopy. On the other hand, RCM, alone or in combination with traditional dermoscopy, proved to increase diagnostic accuracy, in particular, specificity for melanoma diagnosis.
Conclusions: RCM has a defined role in increasing diagnostic accuracy of doubtful dermoscopic lesions, while the role of MD in clinical practice has yet to be defined, and methodologic standardization as well as a revision of terminology is encouraged to improve the recognition of features.

Introduction

Magnified dermoscopy (MD), or optical super-high magnification dermoscopy, is an imaging technique that offers up to 400x magnification, allowing for the visualization of cellular and structural details not visible with conventional 10-20x dermoscopy [1,2] MD has been applied to various skin lesions, particularly for differentiating between nevi and melanomas, with criteria such as irregular cell shape and size being identified as significantly associated with malignant lesions [1,2]. Despite growing interest in its potential applications in routine skin cancer diagnosis, data on MD remain limited.

Noteworthy, reflectance confocal microscopy (RCM) is a non-invasive diagnostic technique enabling the visualization of cells and architecture on different skin layers; as an adjunctive tool, it has been proven to reduce unnecessary excisions and significantly improve melanoma recognition, as compared to dermoscopy, in a recent randomized control trial [3]. However, the impact of MD in diagnostic accuracy of melanocytic lesions has not yet been provided, and a direct comparison of the adjunctive role of MD and RCM as compared to conventional dermoscopy is currently lacking.

This retrospective study aimed to share our experience with dermoscopically equivocal pigmented lesions, analyzing their characteristics and evaluating the diagnostic accuracy of melanoma detection using conventional dermoscopy, 400x MD, and RCM.

Materials and Methods

Study Population

Conventional dermoscopy and MD (400x) images of patients presenting to the Dermatology Unit at Policlinico of Modena, Italy, with flat, dermoscopically equivocal pigmented lesions—diagnosed as either nevi or melanoma—were retrospectively analyzed. Diagnoses were confirmed through histopathological examination or a two-year clinical follow-up. Data on patient age, sex, anatomical lesion location, and histopathological results were also collected.

Dermoscopy and Magnified Dermoscopy

Dermoscopy images were captured by a technician with over 10 years of experience in both dermoscopy and reflectance confocal microscopy (RCM) using a DermLite Photo device (3Gen, San Juan Capistrano, CA, USA) and evaluated based on the revised 7-point checklist criteria [4]. MD images were obtained using a commercially available Fotofinder Medicam 1000 system (Fotofinder System, Bad Birnbach, Germany) and analyzed following the criteria established by Cinotti et al [2]. MD was performed after a quick exploration of the lesion at lower magnification (usually 70x magnification) to highlight the areas of interest. Then, as previously described [2], at least four images at 400x were taken from the most diagnostically suggestive parts, since MD enables the visualization of small areas at a time of the entire lesion (field of view of 1 x 0.5625 mm).

Reflectance Confocal Microscopy

RCM images were collected with Vivascope 1500® (MAVIG GmbH). RCM criteria included previously described features at the epidermis: atypical cells (presence, shape [dendritic or roundish] and distribution [focal or widespread]); at the dermal-epidermal junction (DEJ): atypical junctional nests, medusa head-like structures, sheet-like structures; at the dermis: heterogeneous dermal nests, and inflammation [5-7].

Statistical Analysis

Statistical analysis was carried out with the SPSS software version 24 (SPSS, Armonk, NYC, US). Demographic, clinical, dermoscopic, MD, and RCM variables were included in the analysis. Continuous variables (patients [N], mean, standard deviation [SD]) were compared using an unpaired student's t test (two groups). Categorical variables (frequency [N, %]) were compared using Pearson's chi-squared test.

Sensitivity, specificity, and 95% confidence intervals (CI) were calculated for melanoma diagnosis with either conventional dermoscopy, MD, or a combination of dermoscopy and MD as well as with RCM alone or in combination with dermoscopy.

In addition, the receiver operating characteristic (ROC) curve was constructed with pulled evaluations of the three evaluators and the area under the curve (AUC) obtained. According to the literature, AUC of 0.5 was considered non-discriminant, 0.7 to 0.8 was considered acceptable, 0.8 to 0.9 was considered excellent, and more than 0.9 was considered outstanding [8]. Alpha level was set at less than 0.05.

Results

Study Population and Dermoscopy

An overall number of 104 lesions, one for each patient, were retrieved. Eight lesions located on the face and two acral located on the feet were excluded, leading to the inclusion of 74 nevi and 20 melanomas, with a mean Breslow thickness of 0.17 ± 0.2 (range 0-0.6).

The distribution of demographic data, dermoscopic evaluation, MD, and RCM features are reported in Table 1. Intuitively, we observed a significant correlation between increased age, size, parameters of the seven-point checklist, and melanoma diagnosis (Table 1).

Magnified Dermoscopy Features

We found some significant correlations between MD features and the final diagnosis of melanocytic lesions. Accordingly, about 24% of melanomas versus 2% nevi showed dots (P=0.005), while 38% melanomas versus 16% nevi showed a network without edged papillae (P=0.03) (Figure 1).

Reflectance Confocal Microscopy

A significant correlation between melanoma and RCM features, including atypical cells with either dendritic or roundish shape showing widespread distribution and atypical junctional nests, was observed (Table 1, Figure 2).

Diagnostic Accuracy

Results related to diagnostic accuracy are reported in Table S1 and Figure 2. Sensitivity for melanoma diagnosis with dermoscopy reached 80% (95% CI: 59.5–93.3), while specificity was 65% (95% CI: 54.5–75.3), very similar to results observed with MD combined with dermoscopy, with an acceptable AUC (Table S1, Figure 3).

Notably, RCM alone or in combination with dermoscopy reached the highest sensitivity (85%, 95% CI: 65.6–96.0) and specificity (81%, 95% CI: 71.2–88.9 and 79%, 95% CI: 69.6–87.8, respectively), with excellent AUC (Table S1, Figure 3).

Discussion

In recent decades, dermoscopy has emerged as an important tool in dermatology, particularly in the diagnosis of skin cancers, earning the reputation as being the dermatologist's stethoscope [9]. More recently, interest has grown in magnified dermoscopy (MD), a technique utilizing 400x magnification to visualize structures not detectable with conventional 20x dermoscopy [10]. While MD has been applied to skin cancer diagnostics, and criteria distinguishing nevi from melanoma have been identified [1,2], this paper presents the first report of diagnostic accuracy of MD as compared to and integrated with dermoscopy and compared to RCM with or without dermoscopy.

The current study highlighted that the diagnostic accuracy of dermoscopy and its combination with MD was similar, while with RCM, we observed a slight increase in sensitivity and a substantial increase in specificity, resulting in improved diagnostic accuracy, as indicated by an excellent AUC and a reduction in false negative cases.

Our results are in line with data from recent meta-analyses highlighting a pooled accuracy for melanoma diagnosis of 88% sensitivity for dermoscopy and 90% for RCM and 38–49% specificity for dermoscopy and 42–77% for RCM, revealing a sensitivity and specificity for dermoscopy of 80% and 65% and RCM of 85% and 81%, respectively [11-13]. As a matter of fact, in our study, RCM showed the highest diagnostic accuracy for melanoma, in particular in terms of specificity, with an increase of about 10% in specificity, as compared to dermoscopic approach. This result can be related to the unique possibility of exploring the skin in different layers separately and of identifying qualitative and quantitative signs of atypia due to the good cellular resolution of the instrument [5,7].

Furthermore, in this study, we observed a correlation between melanoma and the presence of dots and non-edged papillae observed with MD. While the association of non-edged papillae, referred to ill-defined network lines, with melanoma appears intuitive, dots have not previously been associated with a specific diagnosis; this is thus a novel finding. Indeed, as dots have not been correlated to specific anatomic or dermoscopic structures, further investigations should clarify this aspect. According to previous studies, melanoma diagnosis was associated with the presence of

Table 1. Demographic Characteristics and Dermoscopic and MD Features, according to Melanoma or Nevi Diagnosis.

		Melanoma N=20	Nevus N=74	Total N=94	p-value
Demographic characteristics					
Sex, n (%)	female	13 (65)	37 (50)	50 (53.2)	0.233
	male	7 (35)	37 (50)	44 (46.8)	
Age, mean ± SD (range)		59.1 ± 15 (36-93)	44.9 ± 15.4 (8-80)	47.8 ± 16.4 (8-93)	0.001
Skin site, n (%)	neck	1 (5)	4 (5.4)	5 (5.3)	0.307
	trunk	12 (60)	56 (75.7)	68 (72.3)	
	limbs	7 (35)	14 (18.9)	21 (22.3)	
Size, mean ± SD (range)	mm	9.3±4.8 (3-20)	5.6±2.6 (1-12)	6.2±3.4 (1-20)	0.001
Dermoscopy features					
Prevalent pattern, n (%)	reticular	6 (30)	24 (32.4)	30 (31.9)	0.309
	globular	1 (5)	10 (13.5)	11 (11.7)	
	homogeneous	1 (5)	3 (4.1)	4 (4.3)	
	Peripheral globules/starburst	0	4 (5.4)	4 (4.3)	
	Two-component	7 (35)	27 (36.5)	34 (36.2)	
	Multi-component	5 (25)	6 (8.1)	11 (11.7)	
Atypical pigmentation, n (%)		18 (90)	41 (55.4)	59 (62.8)	0.004
Blue-white veil, n (%)		12 (60)	11 (14.9)	23 (24.5)	0.001
Atypical vessels, n (%)		1 (5)	6 (8.1)	7 (7.4)	1
Irregular streaks, n (%)		5 (25)	4 (5.4)	9 (9.6)	0.019
Irregular dots/globules, n (%)		11 (55)	38 (51.4)	49 (52.1)	0.772
Irregular blotches, n (%)		13 (65)	24 (32.4)	37 (39.4)	0.008
Regression, n (%)		14 (70)	39 (52.7)	53 (56.4)	0.166
MD features					
Keratinocytes, n (%)		18 (90)	67 (90.5)	85 (90.4)	1.000
Roundish melanocytes, n (%)		11 (55)	39 (52.7)	50 (53.2)	0.855
Dendritic melanocytes, n (%)		2 (10)	11 (14.9)	13 (13.8)	0.728
Melanophages, n (%)		6 (30)	16 (21.6)	22 (23.4)	0.432
Cell irregularity, n (%)		8 (40)	19 (25.7)	27 (28.7)	0.209
Cell distribution, n (%)		12 (60)	57 (77)	69 (73.4)	0.126
Dots, n (%)		5 (25)	2 (2.7)	7 (7.4)	0.004
Roundish nests, n (%)		1 (5)	12 (16.2)	13 (13.8)	0.287
Structureless, n (%)		15 (75)	47 (63.5)	62 (66)	0.430
Vessels, n (%)		7 (35)	29 (39.2)	36 (38.3)	0.732
Hyperkeratotic concentric areas, n (%)		3 (15)	7 (9.5)	10 (10.6)	0.439
Network with edged papillae, n (%)		9 (45)	46 (62.2)	55 (58.5)	0.167
Network without edged papillae, n (%)		8 (40)	12 (16.2)	19 (21.3)	0.021
RCM features					
Atypical cells, n (%)		16 (80)	6 (8.1)	22 (23.4)	0.001
Widespread atypical cells, n (%)		2 (10)	1 (1.4)	3 (3.2)	0.001

Table 1. Demographic Characteristics and Dermoscopic and MD Features, according to Melanoma or Nevi Diagnosis. ***(continued)***

		Melanoma N=20	Nevus N=74	Total N=94	p-value
Dendritic cells, n (%)		13 (65)	6 (8.1)	19 (20.2)	0.001
Roundish cells, n (%)		1 (5)	0	1 (1.1)	0.001
Atypical junctional nests, n (%)		11 (55)	9 (12.2)	20 (21.3)	0.001
Medusa head like structures, n (%)		1 (%)	0	1 (1.1)	0.213
Sheet-like structures, n (%)		0	1 (1.4)	1 (1.1)	1.000
Heterogeneous dermal nests		2 (10)	1 (1.2)	3 (3.2)	0.113
Inflammation		0	5 (6.8)	5 (5.3)	0.581

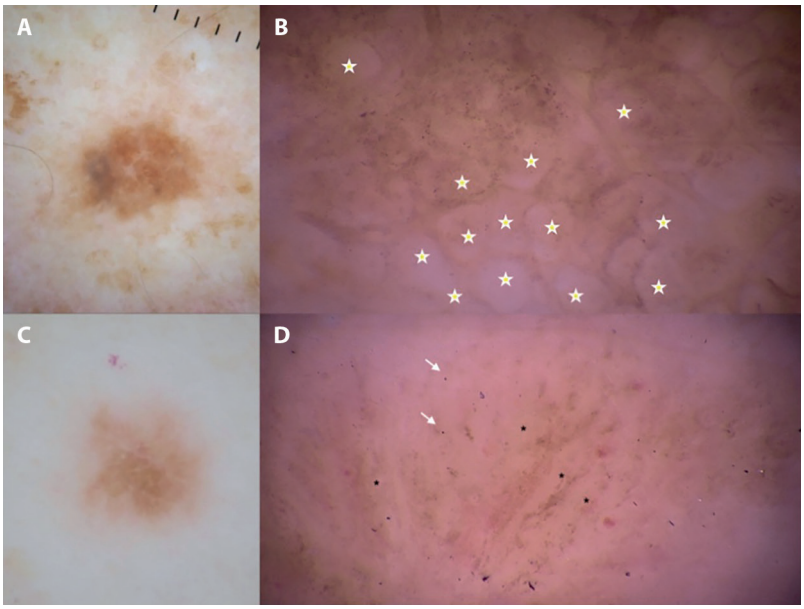


Figure 1. Pictures of conventional dermoscopy (CD) and magnified dermoscopy (MD) in a nevus and a melanoma. A) CD showing a pigmented lesion with different shades of brown color, irregular network and reticular depigmentation, with a histopathological diagnosis of nevus, and B) corresponding MD showing edged papillae/well-defined network lines (stars). C) CD showing a pigmented brown lesion with irregular network and focal reticular depigmentation, with a histopathological diagnosis of melanoma 0.3 mm Breslow, and D) corresponding MD showing non-edged papillae/ill-defined network lines (asterisks) and dots (arrows).

scattered, large, irregular (in shape and size), dendritic or roundish violet/blue pigmented cells in melanoma, while edged papillae (or well-defined network lines) were more commonly associated with nevi in MD [1,2].

This study represents the first attempt to clarify the impact of the emerging MD in melanoma diagnosis, showing that currently, MD diagnostic accuracy is comparable to conventional dermoscopy. One potential explanation for this finding is the recently reported lack of consensus among evaluators regarding the identification of MD features [14]. Accordingly, a notable challenge lies in the terminology used for MD, which shares similarities with features observed in RCM and conventional dermoscopy, potentially causing confusion for evaluators experienced with different imaging modalities. To date, no comprehensive alignment between the features of conventional dermoscopy, histology, RCM, and MD has been established, highlighting the need for standardized terminology to define MD criteria.

However, the retrospective design of the study may present certain limitations. Indeed, only images of small areas from each skin lesion can be captured with MD (while RCM Vivascope 1500 enables the visualization of the full lesion), impairing a comprehensive evaluation of the entire lesion. Additionally, the images in the current study were acquired

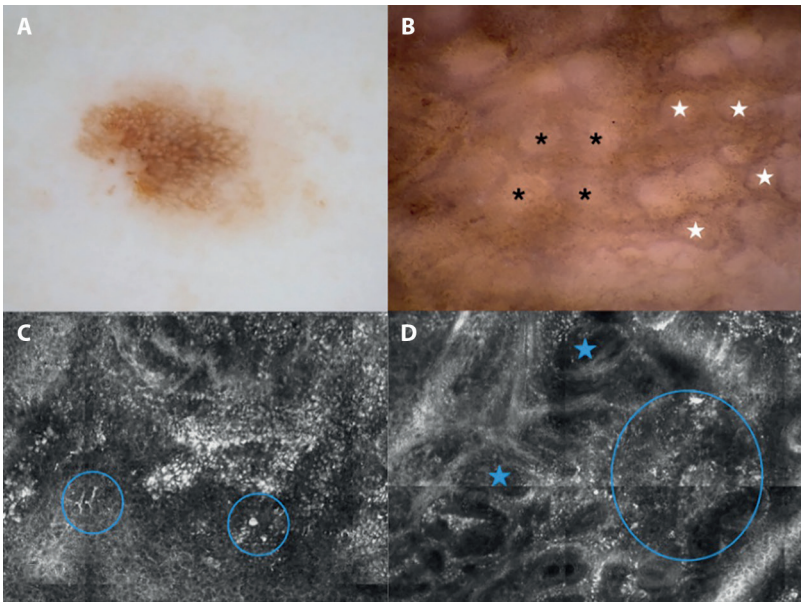
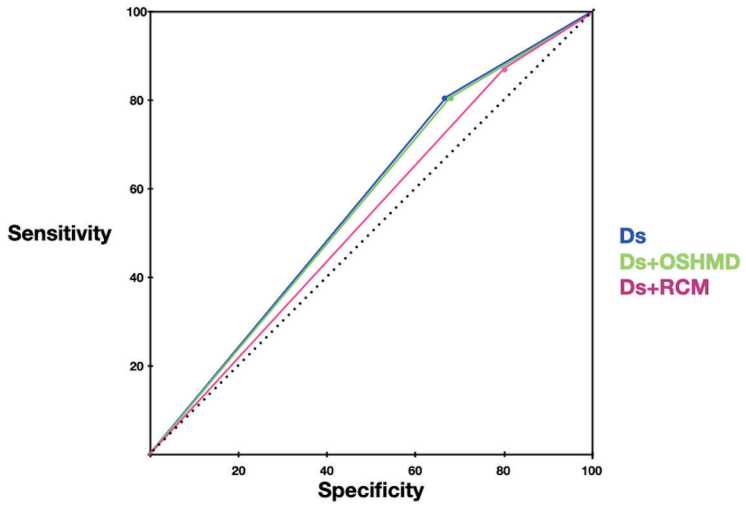


Figure 2. A) Conventional dermoscopy of a melanoma showing an irregular pigmented network; B) corresponding magnified dermoscopy showing non-edged papillae/ill-defined network lines (asterisks) as well as edged papillae/well-defined network lines (stars); C) corresponding reflectance confocal microscopy (RCM) image at epidermal level showing atypical cells (blue circles) and D) RCM of the same lesion at dermo-epidermal level showing atypical cells (blue circle) and non-edged papillae (blue stars).



	AUC	SE	95% CI
Ds	0.718	0.062	0.595-0.840
Ds+OSHMD	0.731	0.062	0.586-0.701
Ds+RCM	0.824	0.054	0.718-0.929

Figure 3. Receiver operating characteristic (ROC) curve for dermoscopy, magnified dermoscopy (MD), reflectance confocal microscopy, and the area under the curve (AUC) obtained.

by an investigator without prior experience in MD, suggesting that diagnostic accuracy could be enhanced through prospective studies conducted by experienced investigators with better area selection for imaging.

Another significant challenge with MD is related to the quality of images necessary for reliable feature identification. Current guidelines for image acquisition are lacking, further complicating the proper application of this technique.

Furthermore, melanocytic lesions in this study included cases with challenging differential diagnoses, such as clinically atypical nevi and early-stage melanomas, as indicated by the low median Breslow index and moderate diagnostic accuracy of conventional dermoscopy, which may explain some differences in terms of MD feature distribution as compared to previous studies also including thicker melanomas [1,2].

Taken together, our results underscore the adjunctive value of RCM in improving melanoma diagnosis and suggest that while MD has the potential to enhance diagnostic sensitivity, advancements in image acquisition techniques and feature recognition are critical for its broader application.

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