HHS Public Access

Author manuscript

J Eur Acad Dermatol Venereol. Author manuscript; available in PMC 2020 February 01.

Published in final edited form as:

J Eur Acad Dermatol Venereol. 2019 February ; 33(2): 439–446. doi:10.1111/jdv.15257.

Accuracy of tele-consultation on management decisions of lesions suspect for melanoma using reflectance confocal microscopy as a stand-alone diagnostic tool

Dr Scope A[1,2], **Dusza SW**[2], **Pellacani G**[3], **Gill M**[4], **Gonzalez S**[2,5], **Marchetti MA**[2], **Rabinovitz HS**[6], **Marghoob AA**[2], **Alessi-Fox C**[7], and **Halpern AC**[2]

[1]Medical Screening Institute, Sheba Medical Center and Sackler School of Medicine, Tel Aviv University, Tel Aviv, Israel

[2]Dermatology Service, Department of Medicine, Memorial Sloan Kettering Cancer Center, New York, USA

[3]Department of Dermatology, University of Modena and Reggio Emilia, Italy

[4]SkinMedical Research and Diagnostics, Dobbs Ferry, NY, USA and SUNY Downstate, Brooklyn, NY, USA

[5]Department of Medicine and Medical Specialties, University de Alcalà, Madrid, Spain

[6]Skin and Cancer Associates, Plantation, Florida, USA

[7]Caliber Imaging and Diagnostics, Rochester, NY, USA

Abstract

Background: Diagnostic accuracy of reflectance confocal microscopy (RCM) as a stand-alone diagnostic tool for suspect skin lesions has not been extensively studied.

Objective: Primary aim was to measure experts' accuracy in RCM-based management decisions. Secondary aim was to identify melanoma-specific RCM features.

Methods: The study enrolled patients ≥18 years that underwent biopsy of skin lesions clinically suspected to be melanoma. One hundred lesions imaged by RCM were randomly-selected from 439 lesions prospectively collected at four pigmented lesion clinics. The study dataset included 23 melanomas, 3 basal cell and 2 squamous cell carcinomas, 11 indeterminate melanocytic lesions, and 61 benign lesions including 50 nevi. Three expert RCM evaluators were blinded to clinical or dermoscopic images, and to the final histopathologic diagnosis. Evaluators independently issued a binary RCM-based management decision, 'biopsy' vs. 'observation'; these decisions were scored against histopathological diagnosis, with 'biopsy' as the correct management decision for malignant and indeterminate lesions. A subset analysis of 23 melanomas and 50 nevi with unequivocal histopathological diagnosis was performed to identify melanoma-specific RCM features.

Corresponding Author: Allan C. Halpern, MD, Dermatology Service, Memorial Sloan Kettering Cancer Center, 16 East 60th Street, New York, NY 10022, Tel: (646) 888-6012, halperna@mskcc.org.

Results: Sensitivity, specificity and diagnostic accuracy were 74%, 67%, and 70% for reader 1, 46%, 84% and 69% for reader 2, and 72%, 46% and 56% for reader 3, respectively. The overall kappa for management decisions was 0.34. Readers had unanimous agreement on management for 50 of the 100 lesions. Nonspecific architecture, non-visible papillae, streaming of nuclei, coarse collagen fibers and abnormal vasculature showed a significant association with melanoma in the evaluation of at least two readers.

Conclusions: RCM tele-consultation of especially challenging lesions, based on image review without benefit of clinical or dermoscopy images, may be associated with limited diagnostic accuracy and interobserver agreement. Architectural and stromal criteria may emerge as potentially useful and reproducible criteria for melanoma diagnosis.

INTRODUCTION

Reflectance confocal microscopy (RCM) permits non-invasive acquisition of "optical sections" of skin with cellular detail and is particularly applicable to examination of melanocytic neoplasms.[1] A meta-analysis[2] of diagnostic accuracy of RCM for melanoma examined five studies composed of 909 lesions[3–7] and identified an overall sensitivity of 93% and specificity of 76%. Several studies[3,5,6] reported on achieving high diagnostic accuracy, based on RCM reading that was blinded to anamnestic information (other than patients' age and lesions' anatomic location), as well as to dermoscopic and clinical images.

Since RCM diagnostic proficiency has a learning curve, beginners using RCM in their practice may seek experts' guidance when encountering difficult lesions. As RCM is a digital imaging technology, experts may consult novices via cloud-based "tele-RCM".[8] Further, since RCM produces histopathology-like optical sections of tissue, dermatopathologists may be enrolled as "tele-RCM" diagnosticians. Notably, dermatopathologists routinely render diagnoses based on "stand-alone" tissue slides, with clinical description that is lacking or insufficient, and in the absence of supplemental clinical images. To this end, the goal of this prospective, multicenter study was to simulate and test tele-RCM diagnosis, with RCM as a stand-alone diagnostic tool. Experts remotely evaluated standardized RCM images of lesions that were suspicious for melanoma, yet clinically- and dermoscopically-equivocal, and issued management decisions. These readers were provided limited clinical data (age, sex, melanoma history and lesion size and location) and no clinical or dermoscopic lesion images. The principle study aim was to measure experts' accuracy in stand-alone RCM-based management decisions. As secondary aims, we sought to identify attributes that are helpful in reaching the correct diagnosis and to measure readers' interobserver reliability.

MATERIALS AND METHODS

Subjects and lesions

The Institutional Review Boards at Memorial Sloan Kettering Cancer Center, Loma Linda University Medical Center, University of Rochester Medical Center and Western IRB approved the study protocol. Study participants were recruited from patients undergoing skin cancer screening at the aforementioned tertiary medical centers and at a private practice.

Inclusion criteria were: (1) age ≥18 years, (2) consent for biopsy of lesions clinically- and dermoscopically-suspicious for melanoma, and (3) lesions located on anatomical sites amenable to wide-probe RCM imaging. Exclusion criteria were: (1) lesions on acral volar surfaces, (2) lesion treatment with topical therapies within the past six weeks, (3) peri-lesional skin afflicted with another condition, and (4) hypersensitivity to RCM adhesive rings. Informed consent was obtained from each participant. Participants were enrolled from August 21st, 2008 through January 5th, 2010.

Lesion imaging and biopsy protocol

Dermoscopic images were acquired per standard-of-care at each contributing center. Study imaging was performed with VivaScope1500 (CaliberI.D., Rochester, NY). Following oil application, a metal ring attached to polycarbonate window was centered on the lesion. A macroscopic image was captured, allowing dermoscopy-guided lesion navigation. Water-based gel was applied over the adhesive window as immersion medium for the RCM probe. Individual RCM images (500×500μm) were sequentially collected in the horizontal X-Y-plane to create 6×6mm mosaic images.

To ensure standardized RCM imaging across study sites, a software prompted RCM imaging in specific sequence. RCM imaging consisted of appropriately-sized mosaics that encompassed the entire lesion, up to 6×6 mm. If lesion diameter was >6 mm, the ring was centered on the most suspicious area; if heterogeneously colored, the ring was placed so that images sampled disparate areas of the multi-colored lesion.

Standard RCM image set included horizontal mosaics captured at the granular-spinous layers, dermal-epidermal junction (DEJ), and superficial dermis. Predefined image stacks – 30 images spaced 5μm apart from stratum corneum to superficial dermis – were obtained from four lesion margins. Three additional image stacks could be captured at the investigator's discretion.

Lesions were biopsied per standard-of-care for each contributing institution. Biopsy specimens were processed at the contributing institution's pathology laboratory and a board-certified dermatopathologist issued a diagnostic report and categorized the lesions as benign, indeterminate, or malignant. Indeterminate lesions were those with a differential diagnosis between nevus and melanoma (e.g., "atypical melanocytic proliferation") or an equivocal diagnosis of nevus (e.g., "early evolving melanoma in situ cannot be ruled out", "dysplastic nevus with high-grade atypia and regression").

Evaluation of images

Upon completion of participant enrollment, three readers independently reviewed 100 RCM cases randomly-selected from the dataset. All included lesions were equivocal for clinical and dermoscopic diagnosis by pigmented lesion experts. RCM readers were experts in diagnostic interpretation of RCM images and had varied professional backgrounds (two clinical dermatologists and one dermatopathologist). For each case, readers received clinical data, including patient's age, sex, and melanoma history, as well as lesion size and anatomic location. Readers were blinded to lesion's clinical and dermoscopic photographs, as well as histopathological diagnosis. Readers accessed images via secure RCM image server

(VivaNet, CaliberI.D.) and evaluations were entered into standardized study-specific web interface. Each reader was supplied with an imaging workstation and calibrated monitor to assure image display uniformity. Readers received the complete set of RCM images acquired for each case without pre-selection of images. Readers scored the presence of RCM descriptors (eTable 1) and management recommendation (biopsy *vs.* observation). The list of RCM descriptors included terms commonly used in the diagnosis of melanoma and nevi; additional descriptors were included to encompass the possible inclusion of non-melanocytic neoplasms that may clinically mimic melanoma. RCM reader reviews were complete 09/21/2011.

Statistical analysis

Descriptive statistics were used for study participants, lesions and results of RCM assessments. To measure sensitivity, specificity and diagnostic accuracy, we considered a binary test of management decisions based on RCM analysis; malignant if readers recommended biopsy, or benign if readers recommended observation. Accuracy of RCM-based management decisions was judged against histopathological diagnosis; biopsy was scored as correct for histopathologically-malignant or -indeterminate lesions, and observation as correct for histopathologically-benign lesions.

Given the lack of reference standard for presence or absence of RCM features, overall and reviewer pairwise assessments of RCM feature prevalence, overall percent agreement, kappa and prevalence-adjusted-bias-adjusted-kappa (PABAK) statistics were used to evaluate interobserver agreement. Percent agreement was calculated as sum of concordant positive and negative evaluations divided by sample size. PABAK estimates were used due to the low prevalence of RCM features. PABAK assumes a 50% prevalence and was interpreted as standard kappa statistics as described by Landis and Koch with values <0.4 representing poor agreement, 0.4–0.75 fair-to-good agreement, and >0.75 excellent agreement.

Logistic regression models were created to assess association between lesion diagnosis and individual RCM features. Analyses were performed as a group, with each RCM reader contributing evaluations and separately, stratified by RCM reader. Standard error estimates for group analyses adjusted for clustered observations within reviewer. Regression models for binary outcomes using general estimating equations approach with a log link and exchangeable correlation structure were utilized. All analyses were performed with Stata v12.1, Stata Corporation, College Station, TX. Primary analyses were performed between January 2012 and December 2014.

RESULTS

One hundred lesions from 92 participants were randomly-selected from 439 lesions collected from participating enrollment sites. Sixty-four (64%) participants were male and mean age was 53.8 (range 18–81) years. The histopathological diagnosis for the 100 lesions consisted of 28 malignant neoplasms (23 melanoma, 3 basal cell carcinoma (BCC), and 2 squamous cell carcinoma (SCC)), 11 indeterminate melanocytic lesions, and 61 benign lesions (50 nevus, 5 seborrheic keratosis/solar lentigo/lichen planus-like keratosis, 3 actinic

keratosis (AK), 1 dermatofibroma, 1 interface dermatitis, 1 post-inflammatory pigment alteration).

There were divergent imaging quality assessments among the RCM readers. Reader 1 indicated that 5% of image sets were limited, 4 (4/100, 4%) by image quality and 2 (2%) by insufficient lesion area imaged; Reader 2 indicated sufficient image quality in all cases; while Reader 3 denoted 27% of imaged lesions as limited, 17 (17%) by insufficient lesion area imaged and 12 (12%) by limited image quality.

For management decisions, the sensitivity, specificity and diagnostic accuracy values were 74%, 67%, and 70% for reader 1, 46%, 84% and 69% for reader 2, and 72%, 46% and 56% for reader 3, respectively (Table 1). The overall kappa for management decision was 0.34. Of 100 lesions, all 3 readers agreed on management decisions (biopsy *vs.* observation) in 50 cases (50%); the pooled sensitivity, specificity and diagnostic accuracy for these RCM unanimously-managed lesions were 75.0%, 72.7% and 74.0%, respectively. Because melanocytic neoplasms with indeterminate diagnosis may introduce a misclassification bias in the interpretation of the results, we also calculated diagnostic accuracy for the subset of 89 lesions for which a clear-cut histopathologic diagnosis was issued; the sensitivity, specificity and diagnostic accuracy values were 82%, 67% and 72% for reader 1, 57%, 84% and 75% for reader 2, and 79%, 46% and 56% for reader 3, respectively.

The prevalence of RCM features as observed by the readers is presented (Table 2). Prevalence of RCM features identified by all 3 readers ranged from 1–37% for the evaluated lesions, with most features being observed in <20% of lesions. Interobserver agreement measures – percent agreement and prevalence-adjusted-bias-adjusted-kappa (PABAK) values – are also presented (Table 2).

The reader-specific association between diagnosis and RCM features based on univariate logistic regression was analyzed. First, since the study set of lesions that were clinically suspected of being melanoma included non-melanocytic neoplasms, we calculated the association between RCM features and malignant (melanoma, indeterminate melanocytic neoplasms, BCC, and SCC) *vs.* benign diagnosis for all 100 study lesions (Table 3); notably, AK was classified as benign in this analysis. Epidermal infiltration by cells in Pagetoid pattern, nonspecific architecture, and abnormal vasculature were associated with malignant diagnosis for two readers. In addition, coarse collagen fibers and streaming of nuclei were associated with malignant diagnosis for all three readers.

To identify melanoma-specific RCM features, we calculated the association between RCM features and diagnosis for the subset of 73 melanocytic neoplasms with unequivocal histopathological diagnosis, including 23 melanomas and 50 nevi (Table 4); none of the features were associated with melanoma in the evaluation of all three readers. However, nonspecific architecture, non-visible papillae, streaming of nuclei, coarse collagen fibers and abnormal vasculature showed a significant association with melanoma in the evaluation of two of three readers. Notably, infiltration in Pagetoid pattern showed nearly-significant association with melanoma in the evaluation of all 3 readers.

DISCUSSION

In this study, three experts, simulating tele-RCM consultants, independently evaluated RCM images of equivocal skin lesions. As the primary aim, we measured accuracy based on experts' management recommendations, judged against the reference standard of histopathologic diagnosis. The accuracy ranged between 56%−70%, with a sensitivity of 46%−74% and a specificity of 46%−84%. Our findings suggest that at the current level of RCM diagnostic expertise, the procedure has limitations as a stand-alone diagnostic tool in the absence of clinical or dermoscopic images.

The diagnostic accuracy measured in our study is notably lower than in other RCM studies. Rao et al[9] evaluated accuracy of RCM reading of 334 lesions by an onsite and a remote reader. Evaluated sets included both dermoscopic and RCM images. Single-reader sensitivity was >90% and specificity was >60%. Ludzik et al[10] retrospectively evaluated 100 equivocal cases with combined dermoscopy-RCM image sets and found single-reader sensitivity of 89% and specificity of 66%. Witkowski et al[11] had 10 readers retrospectively evaluate 1000 combined dermoscopy-RCM image sets and found single-reader sensitivity of 95% and specificity of 76%.

Several factors may have contributed to our results. First, most prior studies included dermoscopy images and readers who were proficient in both dermoscopy and RCM. At the bedside, pre-test probability for RCM diagnosis is already narrowed-down by a skilled clinician's differential diagnosis (e.g. BCC vs. intradermal nevus), and in such cases, RCM can greatly enhance diagnostic accuracy. In addition to informing the interpretation of RCM readers, dermoscopy and RCM expertise may have influenced selection of RCM stacks during image acquisition process in prior studies. In practice, to attain high sensitivity in melanoma diagnosis, proficiency in dermoscopy and RCM should be regarded as complementary; RCM testing results need to be reconciled with the overall clinical and dermoscopic judgment.[12,13] For example, Guitera et al have shown that for light-colored lesions, sensitivity for melanoma diagnosis of combined dermoscopy-RCM is higher than RCM alone.[14] Our rationale for blinding the readers was that tele-RCM experts might be dermatopathologists who routinely make stand-alone histopathologic diagnosis, and who may lack clinical and dermoscopic diagnostic expertise.

Second, lesions included in the study may have been more difficult for diagnosis than in prior studies. The dataset consisted of lesions that were suspicious for melanoma and equivocal to clinical and dermoscopic diagnosis in the hands of pigmented lesions experts. Notably, 40% of lesions came from a clinic that largely treats older patients with severely sun-damaged skin. In our experience, evaluators who do not routinely examine skin lesions on severely sun-damaged skin experience a decline in specificity by dermoscopy or RCM in this setting. This observation is reiterated by the fact that 9/11 (82%) of the lesions whose histopathological diagnosis was 'indeterminate' also originated from this clinic.

While if taken at face value, the diagnostic accuracy and interobserver agreement in our study seems concerning, it is important to note that the gold standard of histopathology that we applied has its own limitations. These limitations have led to a recent concerted effort to

create a simplified, clinically-relevant ontology of histopathologic diagnosis of melanocytic tumors termed M-Path.[15,16] However, even with this simplified schema, interobserver agreement for 'type 3' lesions (e.g., melanoma in situ) among experienced pathologists was 40%.[17]

Considering we are relatively early on the learning curve of RCM interpretation, our results can be seen as encouraging and suggest that lessons learned from pathology should be applied. First, the level of pathologists' accuracy and concordance for diagnosis of melanocytic neoplasms significantly increases when pertinent clinical data becomes available (e.g., clinician's diagnosis and dermoscopic images).[18] Second, case-selection can affect diagnostic concordance. In a study of 1249 clinically-equivocal melanocytic neoplasms, interobserver agreement among pathologists was markedly lower for patients with clinically-atypical moles (kappa=0.31) compared to patients without clinically-atypical moles (kappa=0.76).[19] Finally, agreement between experts can increase with time. For example, interobserver agreement on presence of ulceration and mitotic rate in melanomas improved over a 10-year period.[20]

In addition to measuring accuracy, we secondarily aimed at identifying key RCM criteria that help differentiate melanoma from benign neoplasms and to understand whether different RCM readers can recognize these key criteria in a reproducible fashion. For this analysis, we focused on melanocytic neoplasms that were issued an unequivocal diagnosis of melanoma or nevus. We found some criteria that correlated with the diagnosis of melanoma and that had fair interobserver agreement. Nonspecific architecture, nonvisible papillae, coarse collagen fibers, abnormal vasculature and streaming of nuclei showed a significant association with melanoma in evaluation by two of three RCM readers; these criteria also showed fair-to-good interobserver agreement. In addition, epidermal infiltration by cells in a Pagetoid pattern displayed a nearly-significant association with melanoma; however, this RCM criterion showed poor interobserver agreement.

New criteria emerged as potentially useful and reproducible for the diagnosis of melanoma. Nonspecific architecture and non-visible papillae at the DEJ level (eFigure1), and abnormal vasculature (eFigure 2A) and coarse collagen fibers (eFigure 2B) at the superficial dermis level, were significantly-associated with the diagnosis of melanoma in the evaluation of two of the readers and attained fair-to-good inter-observer agreement. Nonspecific architecture seen at 'low-magnification' mosaic view, and non-visible papillae seen at 'high-magnification' optical section view, denote disruption or loss of the undulating DEJ pattern due to flattening of the retes (eFigure1).[21] Coarse collagen fibers likely correlate in some cases with solar elastosis.[22] In other cases, the abnormal collagen seen under RCM is probably related to stromal remodeling in melanoma. These findings support previous dermoscopic observations. [23,24] The presence of abnormal collagen, seen under polarized dermoscopy as white shiny lines, is 10-fold more frequent among melanomas than nevi.[25] The presence of dermoscopically-identified polymorphous blood vessels is also more frequent among melanomas than nevi.[26] Finally, streaming of nuclei has been attributed to the diagnosis of BCC; however, two of the readers identified this feature in melanomas during blinded evaluations, but could not account for this finding in retrospective examination. The utility of this feature requires further study.

Our study has limitations. First, the study is based on a limited number of lesions. Second, our methodology risks RCM misclassification of lesions>6mm in diameter due to subjective selection of the focus of interest within the lesion. Third, the study uses histopathological diagnosis, and not biological outcome, as reference standard; subsequently, 11% of the dataset was histopathologically-classified as indeterminate. Fourth, there was variability in perception of image quality – one reader marked a fourth of images as limited in quality. This may be due to a learning curve in the training of RCM technicians or due to a decreased confidence level of this reader in the blinded setting, leading to a higher expected image quality standard. Fifth, a substantial fraction of melanomas consisted of melanomas arising on sun-damaged skin. Pellacani et al have reported variability in RCM criteria between different subtypes of melanoma.[27] Thus, our results may not be generalizable to populations composed of different proportions of melanoma subtypes. Finally, despite utilizing a uniform feature-set, readers appeared to be giving different diagnostic weight to features. Readers may have also uniquely recognized additional RCM findings, not listed in the reportable set. These interpreter variations were likely diagnostically advantageous in certain cases and distracting in others.

In conclusion, our data suggest that RCM tele-consultation of especially challenging lesions, based on image review without benefit of clinical or dermoscopy images, may be associated with limited diagnostic accuracy and interobserver agreement. We suggest that supplementation of the standardized image acquisition with stacks from sites deemed concerning by the technician, along with inclusion of clinical and dermoscopy images to benefit interpretation by reviewers, will improve diagnostic accuracy. Moreover, our data support the need for formal RCM courses with integrated dermoscopic and histopathologic training. As with the ongoing efforts in histopathology, a simplified, clinically-relevant ontology of diagnostic categories and shared training in image interpretation should improve both diagnostic accuracy and reproducibility. To this end, our data suggest that upon further study, architectural and stromal RCM criteria may emerge as potentially useful and reproducible criteria for diagnosis of melanoma.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

ACKNOWLEDGEMENTS:

The authors thank Alice Pentland, MD (Chair and Professor of Dermatology, University of Rochester Medical Center, New York, NY) and Abel Torres, MD, JD (Chair and Professor of Dermatology, Loma Linda University Medical Center, Loma Linda, CA) for their contribution in patient accrual and acquisition of study data.

Funding: This research was funded in part through the NIH/NCI Cancer Center Support Grant P30 CA008748 and by the NIH/NCI Grant 5R44CA058054–06. Funding source was not involved in design and conduct of the study, collection, management, analysis and interpretation of data, preparation, review, or approval of the manuscript, or decision to submit the manuscript for publication.

Financial Disclosure:

Dr. Pellacani – honoraria for courses on confocal microscopy from MAVIG GmbH and advisory board member for Caliber ID, manufacturer of a confocal microscope.

Dr. Gill was a funded investigator of the NCI grant. She has no relevant conflicts of interest to report.

Dr. Gonzalez – scientific Advisory Board member of Caliber ID

Dr. Rabinovitz – speaker and clinical investigator for Caliber ID

Ms. Alessi-Fox – employee and a shareholder of CaliberID

Dr. Halpern – scientific Advisory Board member of Caliber ID

Disclosure for all the other authors - None reported.

REFERENCES

1. Rajadhyaksha M, Grossman M, Esterowitz D, Webb RH, Anderson RR. In vivo confocal scanning laser microscopy of human skin: melanin provides strong contrast. J Invest Dermatol 1995;104:946–952. [PubMed: 7769264]
2. Stevenson AD, Mickan S, Mallett S, Ayya M. Systematic review of diagnostic accuracy of reflectance confocal microscopy for melanoma diagnosis in patients with clinically equivocal skin lesions. Dermatol Pract Concept 2013;3:19–27. [PubMed: 24282659]
3. Pellacani G, Guitera P, Longo C, Avramidis M, Seidenari S, Menzies S. The impact of in vivo reflectance confocal microscopy for the diagnostic accuracy of melanoma and equivocal melanocytic lesions. J Invest Dermatol 2007;127:2759–2765. [PubMed: 17657243]
4. Pellacani G, Cesinaro AM, Seidenari S. Reflectance-mode confocal microscopy of pigmented skin lesions--improvement in melanoma diagnostic specificity. J Am Acad Dermatol 2005;53:979–985. [PubMed: 16310058]
5. Guitera P, Pellacani G, Crotty KA, et al. The impact of in vivo reflectance confocal microscopy on the diagnostic accuracy of lentigo maligna and equivocal pigmented and nonpigmented macules of the face. J Invest Dermatol 2010;130:2080–2091. [PubMed: 20393481]
6. Guitera P, Pellacani G, Longo C, Seidenari S, Avramidis M, Menzies SW. In vivo reflectance confocal microscopy enhances secondary evaluation of melanocytic lesions. J Invest Dermatol 2009;129:131–138. [PubMed: 18633444]
7. Curchin CE, Wurm EM, Lambie D, Longo C, Pellacani G, Soyer HP. First experiences using reflectance confocal microscopy on equivocal skin lesions in Queensland. Australas J Dermatol 2011;52:89–97. [PubMed: 21605091]
8. Witkowski A, Ludzik J, Soyer HP. Telediagnosis with Confocal Microscopy: A Reality or a Dream? Dermatol Clin 2016;34:505–512. [PubMed: 27692456]
9. Rao BK, Mateus R, Wassef C, Pellacani G. In vivo confocal microscopy in clinical practice: comparison of bedside diagnostic accuracy of a trained physician and distant diagnosis of an expert reader. J Am Acad Dermatol 2013;69:e295–300. [PubMed: 24035553]
10. Ludzik J, Witkowski AM, Roterman-Konieczna I. New telemedicine techniques in dermatology - evaluation with reflectance confocal microscopy via cloud-based platform. Folia Med Cracov 2016;56:21–29.
11. Witkowski AM, Ludzik J, Arginelli F et al. Improving diagnostic sensitivity of combined dermoscopy and reflectance confocal microscopy imaging through double reader concordance evaluation in telemedicine settings: A retrospective study of 1000 equivocal cases. PLoS One 2017;12:e0187748. [PubMed: 29121636]
12. Scope A, Marchetti MA. An Evolving Approach to the Detection of Melanoma and Other Skin Cancers Using In Vivo Reflectance Confocal Microscopy. JAMA Dermatol 2016;152:1085–1087. [PubMed: 27580064]
13. Scope A, Longo C. Recognizing the benefits and pitfalls of reflectance confocal microscopy in melanoma diagnosis. Dermatol Pract Concept 2014;4:67–71.
14. Guitera P, Menzies SW, Argenziano G, et al. Dermoscopy and in vivo confocal microscopy are complementary techniques for diagnosis of difficult amelanotic and light-coloured skin lesions. Br J Dermatol 2016;175:1311–1319. [PubMed: 27177158]
15. Piepkorn MW, Barnhill RL, Elder DE, et al.The MPATH-Dx reporting schema for melanocytic proliferations and melanoma. J Am Acad Dermatol 2014;70:131–41. [PubMed: 24176521]

16. Lott JP, Elmore JG, Zhao GA, et al. Evaluation of the Melanocytic Pathology Assessment Tool and Hierarchy for Diagnosis (MPATH-Dx) classification scheme for diagnosis of cutaneous melanocytic neoplasms: Results from the International Melanoma Pathology Study Group. J Am Acad Dermatol 2016;75:356–63. [PubMed: 27189823]
17. Elmore JG, Barnhill RL, Elder DE, et al. Pathologists' diagnosis of invasive melanoma and melanocytic proliferations: observer accuracy and reproducibility study. BMJ 2017;357:j2813. [PubMed: 28659278]
18. Ferrara G, Argenyi Z, Argenziano G, et al. The influence of clinical information in the histopathologic diagnosis of melanocytic skin neoplasms. PLoS One 2009;4:e5375. [PubMed: 19404399]
19. Braun RP, Gutkowicz-Krusin D, Rabinovitz H, et al. Agreement of dermatopathologists in the evaluation of clinically difficult melanocytic lesions: how golden is the 'gold standard'? Dermatology 2012;224:51–58. [PubMed: 22433231]
20. Niebling MG, Haydu LE, Karim RZ, Thompson JF, Scolyer RA. Reproducibility of AJCC staging parameters in primary cutaneous melanoma: an analysis of 4,924 cases. Ann Surg Oncol 2013;20:3969–3975. [PubMed: 23851608]
21. Scope A, Zalaudek I, Ferrara G, Argenziano G, Braun RP, Marghoob AA. Remodeling of the dermoepidermal junction in superficial spreading melanoma: insights gained from correlation of dermoscopy, reflectance confocal microscopy, and histopathologic analysis. Arch Dermatol 2008;144:1644–9. [PubMed: 19075152]
22. Bassoli S, Rabinovitz HS, Pellacani G, et al. Reflectance confocal microscopy criteria of lichen planus-like keratosis. J Eur Acad Dermatol Venereol 2012;26:578–590. [PubMed: 21605173]
23. Zalaudek I, Kreusch J, Giacomel J, Ferrara G, Catricala C, Argenziano G. How to diagnose nonpigmented skin tumors: a review of vascular structures seen with dermoscopy: part II. Nonmelanocytic skin tumors. J Am Acad Dermatol 2010;63:377–386; quiz 387–378. [PubMed: 20708470]
24. Marghoob AA, Cowell L, Kopf AW, Scope A. Observation of chrysalis structures with polarized dermoscopy. Arch Dermatol 2009;145:618. [PubMed: 19451524]
25. Balagula Y, Braun RP, Rabinovitz HS, et al. The significance of crystalline/chrysalis structures in the diagnosis of melanocytic and nonmelanocytic lesions. J Am Acad Dermatol 2012;67:194.e191–198. [PubMed: 22030020]
26. Menzies SW, Kreusch J, Byth K, et al. Dermoscopic evaluation of amelanotic and hypomelanotic melanoma. Arch Dermatol 2008;144:1120–1127. [PubMed: 18794455]
27. Pellacani G, De Pace B, Reggiani C, et al. Distinct melanoma types based on reflectance confocal microscopy. Exp Dermatol 2014;23:414–418. [PubMed: 24750486]

Table 1:

RCM-based management decisions by final histopathological diagnosis (n=100 lesions)

Histopathological Diagnosis	RCM-based Management Decision					
	Reader 1		Reader 2		Reader 3	
	Biopsy n (%)	Observe n (%)	Biopsy n (%)	Observe n (%)	Biopsy n (%)	Observe n (%)
Invasive melanoma (n=12)	10 (83.3)	2 (16.7)	7 (58.3)	5 (41.7)	11 (91.7)	1 (8.3)
Melanoma in situ (n=11)	8 (72.7)	3 (27.3)	5 (45.5)	6 (54.6)	6 (54.6)	5 (45.5)
Indeterminate melanocytic neoplasms (n=11)	6 (54.6)	5 (45.5)	2 (18.2)	9 (81.8)	6 (54.6)	5 (45.5)
Basal cell carcinoma (n=3)	3 (100)	0 (0)	3 (100)	0 (0)	3 (100)	0 (0)
Squamous cell carcinoma (n=2)	2 (100)	0 (0)	1 (50)	1 (50)	2 (100)	0 (0)
Nevi (n=50)	17 (35.4)	31 (64.6)	7 (14.3)	42 (85.7)	29 (59.2)	20 (40.8)
Benign non-melanocytic neoplasms/ lesions (n=11)	3 (25)	9 (75)	3 (25)	9 (75)	4 (33.3)	8 (66.7)

Table 2.

Overall inter-observer agreement on RCM characteristics (n=100 lesions)

RCM Characteristic	Feature Concordance (Present)	Feature Concordance (Absent)	Percent Agreement	prevalence-adjusted bias-adjusted kappa (PABAK)
Epidermal Infiltration by cells in Pagetoid pattern	15	45	60	0.2
Erosion	1	89	91	0.81
Streaming of nuclei	6	86	92	0.84
Ringed Architecture	22	40	61	0.23
Meshwork Architecture	17	55	73	0.45
Clods Architecture	12	80	93	0.85
Nonspecific Architecture	17	58	75	0.49
Polycyclic papillary Contours	13	70	83	0.67
Non-Edged Papillae	9	60	69	0.37
Non-Visible Papillae	10	64	75	0.49
Junctional Nests / Clusters	37	37	74	0.48
Dermal Nests / Clusters	27	52	79	0.59
Cord-Like Structures	6	84	89	0.79
Cells in Sheet-Like arrangement	1	91	92	0.84
Presence of Atypical Cells	12	42	55	0.09
Plump Bright Cells	32	30	62	0.24
Small Bright Cells	26	29	55	0.11
Coarse Collagen Fibers	8	71	79	0.57
Abnormal Vasculature	11	69	80	0.6

Table 3.

Association between malignant diagnosis (melanoma, 'indeterminate melanocytic neoplasm', BCC, SCC) and RCM feature based on univariate logistic for all lesions (n=100)

	Rater 1		Rater 2		Rater 3	
Characteristic	**OR (95% CI)**	**p-value**	**OR (95% CI)**	**p-value**	**OR (95% CI)**	**p-value**
Epidermal Infiltration by cells in Pagetoid pattern	3.5 (1.4 – 8.5)	**0.006**	5.8 (1.5 – 23)	**0.012**	2.0 (0.9 – 4.7)	0.099
Erosion	3.4 (0.6 – 19.4)	0.173	1.6 (0.2 – 11.8)	0.648	5.4 (1 – 28.1)	**0.047**
Streaming of nuclei	8.8 (1 – 78.7)	**0.051**	4.2 (1 – 17.5)	**0.047**	4.3 (1.2 – 15)	**0.024**
Ringed Architecture	0.5 (0.2 – 1.4)	0.206	0.4 (0.2 – 0.9)	**0.037**	0.7 (0.3 – 1.7)	0.448
Meshwork Architecture	0.6 (0.2 – 1.4)	0.23	1.6 (0.6 – 4.1)	0.325	1.1 (0.5 – 2.4)	0.867
Clods Architecture	0.8 (0.2 – 2.4)	0.626	0.5 (0.2 – 1.8)	0.294	1.3 (0.5 – 3.7)	0.602
Nonspecific Architecture	8.3 (3.2 – 21.5)	**<0.001**	8.9 (2.7 – 29.6)	**<0.001**	1.8 (0.8 – 4.0)	0.181
Polycyclic Papillary Contours	1.6 (0.6 – 4.1)	0.325	1.1 (0.4 – 3.2)	0.84	1.2 (0.5 – 2.9)	0.759
Non-Edged Papillae	5.7 (2.0 – 16.6)	**0.001**	2.2 (0.5 – 10.5)	0.318	1.2 (0.5 – 2.7)	0.663
Non-Visible Papillae	5.7 (2.0 – 16.6)	**0.001**	7.6 (2 – 29.4)	0.003	1.4 (0.6 – 3.2)	0.452
Junctional Nests / Clusters	0.8 (0.3 – 1.8)	0.567	0.7 (0.3 – 1.6)	0.388	0.9 (0.4 – 2.0)	0.797
Dermal Nests / Clusters	0.8 (0.4 – 1.9)	0.657	0.7 (0.3 – 1.7)	0.416	0.7 (0.3 – 1.6)	0.373
Cord-Like Structures	2 (0.6 – 7.2)	0.27	4.2 (1 – 17.5)	**0.047**	2.5 (0.7 – 8.4)	0.152
Cells in Sheet-Like arrangement	6.9 (0.7 – 63.8)	0.091	-	-	3.5 (0.8 – 15)	0.089
Atypical Cells	1.4 (1.0 – 2.2)	0.079	1.5 (1 – 2.4)	0.07	1.5 (0.9 – 2.3)	0.084
Plump Bright Cells	1.8 (0.8 – 4.1)	0.157	2.4 (1 – 5.5)	**0.039**	1.5 (0.6 – 3.7)	0.329
Small Bright Cells	1.3 (0.6 – 2.9)	0.567	1.3 (0.5 – 3.1)	0.561	1.6 (0.7 – 3.7)	0.253
Coarse Collagen Fibers	2.9 (1.1 – 7.3)	**0.029**	4.4 (1.4 – 13.9)	**0.012**	4.4 (1.4 – 13.9)	**0.012**
Abnormal Vasculature	7.1 (2.1 – 24)	**0.002**	4.1 (1.5 – 11.1)	**0.005**	1.6 (0.6 – 4.1)	0.325

Table 4.

Association between melanoma status and RCM features based on univariate logistic regression for the subset of melanocytic neoplasms with unequivocal histopathological diagnosis (n=73 lesions)

	Rater 1		Rater 2		Rater 3	
Characteristic	**OR (95% CI)**	**p-value**	**OR (95% CI)**	**p-value**	**OR (95% CI)**	**p-value**
Epidermal Infiltration by cells in Pagetoid pattern	2.7 (0.9 – 7.7)	0.072	4.3 (0.9 – 19.7)	0.064	2.9 (0.9 – 9.2)	0.064
Erosion	4.6 (0.4 – 53.2)	0.225	-	-	7.2 (0.7 – 73.4)	0.096
Streaming of nuclei	-	-	13.3 (1.5 – 122.1)	**0.022**	8.3 (1.5 – 45.1)	**0.014**
Ringed Architecture	0.3 (0.1 – 1.3)	0.119	0.4 (0.1 – 1.1)	0.063	0.4 (0.1 – 1.4)	0.158
Meshwork Architecture	0.4 (0.1 – 1.3)	0.119	1.1 (0.3 – 3.4)	0.884	1.1 (0.4 – 3)	0.816
Clods Architecture	0.7 (0.2 – 2.7)	0.574	0.4 (0.1 – 1.9)	0.227	1.6 (0.5 – 5.1)	0.454
Nonspecific Architecture	11.3 (3.5 – 36.4)	**<0.001**	9.9 (2.3 – 41.5)	**0.002**	1.6 (0.6 – 4.3)	0.372
Polycyclic Papillary Contours	2.4 (0.6 – 9.5)	0.196	4.3 (0.9 – 19.7)	0.064	2.5 (0.7 – 8.9)	0.15
Non-Edged Papillae	8.7 (2.3 – 32.2)	**0.001**	1.5 (0.2 – 9.4)	0.69	1.4 (0.5 – 3.7)	0.551
Non-Visible Papillae	3.8 (1.1 – 12.8)	**0.03**	16.9 (1.9 – 151.1)	**0.011**	1.3 (0.5 – 3.7)	0.591
Junctional Nests / Clusters	0.7 (0.3 – 2)	0.551	0.5 (0.2 – 1.3)	0.161	0.8 (0.3 – 2.4)	0.724
Dermal Nests / Clusters	1 (0.4 – 2.8)	0.96	0.5 (0.2 – 1.4)	0.175	0.6 (0.2 – 1.6)	0.281
Cord-Like Structures	2.3 (0.4 – 12.4)	0.332	10.1 (1.1 – 96.3)	**0.044**	2.4 (0.5 – 10.5)	0.255
Cells in Sheet-Like arrangement	7.2 (0.7 – 73.4)	0.096	-	-	4.3 (0.9 – 19.7)	0.064
Atypical Cells	1.3 (0.8 – 2.1)	0.339	1.4 (0.8 – 2.3)	0.272	1.3 (0.7 – 2.2)	0.407
Plump Bright Cells	3.5 (1.2 – 10)	**0.018**	2.1 (0.7 – 5.8)	0.16	1.3 (0.5 – 3.6)	0.625
Small Bright Cells	1.7 (0.6 – 4.9)	0.316	1.1 (0.4 – 3.2)	0.871	1.7 (0.6 – 4.6)	0.333
Coarse Collagen Fibers	2 (0.5 – 7.4)	0.302	6.7 (1.5 – 29.1)	**0.011**	5.4 (1.2 – 24.1)	**0.027**
Abnormal Vasculature	15.1 (2.9 – 78.2)	**0.001**	3.2 (1 – 10.3)	**0.052**	1.4 (0.4 – 4.4)	0.59