International Journal of
Molecular Sciences

MDPI

Editorial

Looking into the Skin in Health and Disease: From Microscopy Imaging Techniques to Molecular Analysis

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The skin is a complex organ that includes a wide variety of tissue types with different embryological origins. It is structured in different layers that are functionally interdependent. On the one hand, it plays a protective role against potentially aggressive environmental factors; on the other hand, it is also a communication interface between the body and the environment [1,2]. The skin undergoes a constant process of adaptation to various physiological and pathological conditions. Processes such as the maintenance of the integrity of the skin barrier, cutaneous regeneration, skin aging, skin inflammation, and carcinogenesis are currently topics of major interest for the research and medical communities [2,3]. The aim of our Special Issue was to expand our understanding and to emphasize new research directions related to the investigation of various aspects of the physiology and pathology of the skin, with a special focus on cellular and molecular mechanisms.

Skin cancer is the most common type of malignancy worldwide, and comprises two major types of cancer—melanoma, which is derived from the pigmentary cells of the skin, and non-melanoma skin cancer, developed from keratinocytes or their precursors—together with other, less common types of tumors [4–10].

Actinic keratosis (AK) is a very common premalignant skin lesion, which has the potential to progress to keratinocyte carcinoma [11]. Its high incidence and prevalence, as well as the high recurrence rate after treatment, makes AK a major problem for the public health systems [12,13]. However, the clinical or pathological markers for the progressive character of the lesion are still missing.

In our Special Issue, the research of Dubois-Pot-Schneider et al. [14] evaluates a transcriptomic approach to identify biological features, allowing them to objectively differentiate distinct AK subclasses. The study, which has the advantage of being performed on a large number of lesions, proposed a risk stratification of AKs based on their specific transcriptomic profile for the first time. The authors have described two different AK signatures. One resembles normal skin, and was defined as the lower risk, non-lesional type. The other, with a molecular profile similar to malignant lesions, is defined as the lesional type of AK, and carries a higher risk of evolving into cancer. The results indicate that, in the high-risk Aks, the upregulated genes are connected with inflammation, and the downregulated ones are related to the process of keratinization. These data suggest a very similar pattern to skin squamous cell carcinoma [15–17]. Moreover, they identified the VEGF pathway as being involved in the high-risk lesions, suggesting it as a potential therapeutic target [18,19].

Citation: Caruntu, C.; Ilie, M.A.; Neagu, M. Looking into the Skin in Health and Disease: From Microscopy Imaging Techniques to Molecular Analysis. *Int. J. Mol. Sci.* **2023**, *24*, 13737. https://doi.org/10.3390/ijms241813737

Received: 1 August 2023
Accepted: 23 August 2023
Published: 6 September 2023

Melanoma is an aggressive skin malignancy with a rapidly increasing incidence and high mortality rate, inducing an important impact on healthcare systems [20–22]. The complex melanoma pathogenesis involves various risk factors, including genetic susceptibility to UV exposure, chronic inflammation, and impaired immune responses [23–28]. If diagnosed early, melanoma is curable; however, in advanced stages, it involves complex and costly therapeutic strategies [20]. Thus, more and more studies have focused on the development of new strategies for detection of melanomas in the early stages. In recent years, the development of artificial intelligence and machine learning capabilities have opened new areas of research in the diagnosis of melanomas [29]. In our Special Issue, Foahom Gouabou et al. show the design of a new framework for automated melanoma diagnosis [30], which is easier to decrypt as regards the decision process; this automated diagnosis shows an increased performance as compared to previous systems.

Continuing the topic of our Special Issue, Dobre et al. emphasized the importance of early diagnosis in skin cancer once again, reviewing the most recent and relevant discoveries in the field of imaging techniques used for the diagnosis and therapeutic monitoring of skin cancer [31]. The implementation of non-invasive anatomical imaging techniques, such as confocal laser scanning microscopy (CLSM), optical coherence tomography (OCT), multiphoton microscopy (MPM), high-frequency ultrasound (HFUS), terahertz pulsed imaging (TPI), and magnetic resonance imaging (MRI), has provided better sensitivity and specificity, increasing the diagnostic reliability in skin cancer and premalignant lesions. For example, CLSM is one of the most promising techniques for micromorphological investigation in dermato-oncology, allowing examination of skin structures with a resolution comparable to that of conventional histopathology. It has been shown that CLSM is able to identify key features in different types of skin malignancies or premalignant lesions, being a performant, non-invasive diagnostic tool [32–40] with increased accuracy as compared to previous non-invasive approaches [41–46]. Moreover, it is helpful for identification of distinct tumor subtypes with specific malignant behavior [47–49]; CLSM can also be used for the evaluation of tumor edges and surgical margins [50,51], and offers the advantage of non-invasive monitoring of the therapeutic response [52–60]. Furthermore, the major advances in molecular imaging techniques, such as single photon emission computed tomography (SPECT/CT) and positron emission tomography (PET), and the recent burst in artificial intelligence research, have expanded the boundaries for the investigation of skin cancer, and are proven to be valuable tools for its detection and monitoring [61–63].

In the non-invasive imaging techniques domain highlighted by our Special Issue, the study by Tianxin Gao et al. [64] also focused on non-invasive imaging techniques, proposing a segmentation algorithm based on a deep learning network architecture for the segmentation of OCT images of laser-induced skin damage. The authors used an experimental model on adult BALB/c-mu mice, in which damaged skin areas with various degrees of injury were generated using different radiation doses emitted by a laser source. The skin injuries were investigated using an OCT system, and a deep neural network method was used to achieve accurate segmentation of the OCT images. The evaluation has produced good results, with a high overlap rate and short edge distance between the segmentation of OCT images and the manually labeled areas. These results suggest the possibility of using automated processing methods in the rapid detection and monitoring of skin lesions and the healing process.

The skin regeneration process was another topic explored by Yaotao Guo et al. [65]. They reviewed the fundamental mechanisms associated with skin soft tissue expansion and the involvement of the mechanical stretch process. Skin soft tissue expansion is a common technique in reconstructive specialties, such as plastic surgery and oral and maxillofacial surgery, being widely used in various conditions [66,67].

The action of the mechanical stretch on the skin activates multiple signaling pathways, inducing the activation of cell proliferation, differentiation, and migration. These activation processes involve all layers of the skin, inducing a shift in the behavior of keratinocytes, fibroblasts, and mesenchymal stem cells. Moreover, changes also occur in the hypodermis,

blood vessels, and skin annexes [68,69]. By modulation of these pathways through manipulation of signaling molecules, and increased local supply of growth factors or active cells, the process of skin regeneration associated with soft tissue expansion can be improved.

Skin is constantly exposed to different environmental factors, which may activate potentially harmful processes. In our Special Issue, the effects of tris (1-chloro-2-propyl) phosphate (TCPP)—one of the most used organophosphorus flame retardants [70]—on human skin cells was investigated by Liu at al. [71]. In an in vitro research model on human skin keratinocytes (HaCaT), the authors have shown that TCPP exposure generates intracellular reactive oxygen species, triggers DNA damage, and disturbs the cell cycle control. Moreover, it increases the level of proinflammatory cytokines IL-1beta and IL-6. The keratinocytes' viability is reduced in a concentration-dependent manner, and activation of the pathways involved in cellular senescence suggests that TCPP exposure can be a precipitating factor for skin aging.

Various intrinsic and environmental factors may be involved in skin aging, which is a process of fascinating complexity [72,73]. On the other hand, in recent years, the interest in regenerative medicine has been increasing, and numerous efforts have been made for the development of various anti-aging strategies [74,75].

Adipose-derived stem cells (ASCs) are multipotent cells with a high proliferation ability, which also have important regulatory functions [76]. Their role in rejuvenation and wound healing has been investigated, and the results are promising. ASCs are able to liberate various growth factors, to stimulate secretion of collagen and elastin, and to promote angiogenesis [76–80]. Therefore, the experimental study by Oh et al. published in this Special Issue investigated the impact of high-intensity focused ultrasound (HIFU) on ASCs and adipogenesis [81]. Their results indicate that HIFU modulates the functioning of ASCs by increasing the expression of the heat shock proteins 70 and reducing proinflammatory cytokines, such as NF-κB, IL-6, and TNF-α. Moreover, HIFU intensifies the expression of adipogenesis markers, induces a higher number of adipocytes, and increases the thickness of subcutaneous adipose tissue, suggesting its possible role in rejuvenation procedures.

The skin barrier, along with the other epithelial linings, protects our body against aggressive factors from the environment, and also prevents the loss of essential molecules [82–86]. Its integrity is essential for maintaining the cutaneous and whole-body homeostasis. Stratum corneum plays a fundamental role in the barrier function of the skin, which depends critically on its molecular architecture [1,87–90]. For example, significant alterations in the structure of the stratum corneum and in its lipid composition were identified in skin conditions with an impaired barrier function, such as atopic dermatitis [91–99].

In the study published herein, Sjövall et al. [100] have demonstrated the possibility of determining the molecular composition of superficial layers in stratum corneum, and the spatial distribution of specific lipids using three-dimensional time-of-flight secondary ion mass spectrometry. Thus, this technique could be used to expand the knowledge regarding the skin barrier, and to evaluate the effects of different active pharmaceutical ingredients, cosmetic molecules, or penetration enhancers in relation to this cutaneous function.

Summing up the presented Special Issue, prodigious amounts of research have been carried out in the field of skin imaging using cellular and molecular biology techniques. This technological armamentarium has recently led to massive progress in the diagnosis, targeted treatment, and investigation of pathophysiology of skin cancer.

Nevertheless, new discoveries are in the research pipeline, and many more are needed, as there are still patients that develop skin diseases with no clear pathogenesis, nor an effective treatment.

Author Contributions: Conceptualization, C.C., M.A.I. and M.N.; writing—original draft preparation, C.C., M.A.I. and M.N.; writing—review and editing, C.C., M.A.I. and M.N.; All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Haftek, M.; Roy, D.C.; Liao, I.-C. Evolution of Skin Barrier Science for Healthy and Compromised Skin. *J. Drugs Dermatol.* **2021**, *20*, s3–s9. [CrossRef] [PubMed]
2. Yousef, H.; Alhajj, M.; Sharma, S. Anatomy, Skin (Integument), Epidermis. In *StatPearls*; StatPearls Publishing: Treasure Island, FL, USA, 2023.
3. Nguyen, A.V.; Soulika, A.M. The Dynamics of the Skin's Immune System. *Int. J. Mol. Sci.* **2019**, *20*, 1811. [CrossRef]
4. Aggarwal, P.; Knabel, P.; Fleischer, A.B. United States Burden of Melanoma and Non-Melanoma Skin Cancer from 1990 to 2019. *J. Am. Acad. Dermatol.* **2021**, *85*, 388–395. [CrossRef] [PubMed]
5. Georgescu, S.R.; Tampa, M.; Mitran, C.I.; Mitran, M.I.; Caruntu, C.; Caruntu, A.; Lupu, M.; Matei, C.; Constantin, C.; Neagu, M. Tumor Microenvironment in Skin Carcinogenesis. *Adv. Exp. Med. Biol.* **2020**, *1226*, 123–142. [PubMed]
6. Nehal, K.S.; Bichakjian, C.K. Update on Keratinocyte Carcinomas. *N. Engl. J. Med.* **2018**, *379*, 363–374. [CrossRef]
7. Ion, A.; Popa, I.M.; Papagheorghe, L.M.L.; Lisievici, C.; Lupu, M.; Voiculescu, V.; Caruntu, C.; Boda, D. Proteomic Approaches to Biomarker Discovery in Cutaneous T-Cell Lymphoma. *Dis. Markers* **2016**, *2016*, 1–8. [CrossRef]
8. Souto, E.B.; da Ana, R.; Vieira, V.; Fangueiro, J.F.; Dias-Ferreira, J.; Cano, A.; Zielińska, A.; Silva, A.M.; Staszewski, R.; Karczewski, J. Non-Melanoma Skin Cancers: Physio-Pathology and Role of Lipid Delivery Systems in New Chemotherapeutic Treatments. *Neoplasia* **2022**, *30*, 100810. [CrossRef]
9. Lupu, M.; Caruntu, C.; Ghita, M.A.; Voiculescu, V.; Voiculescu, S.; Rosca, A.E.; Caruntu, A.; Moraru, L.; Popa, I.M.; Calenic, B.; et al. Gene Expression and Proteome Analysis as Sources of Biomarkers in Basal Cell Carcinoma. *Dis. Markers* **2016**, *2016*, 9831237. [CrossRef]
10. Tampa, M.; Georgescu, S.R.; Mitran, M.I.; Mitran, C.I.; Matei, C.; Caruntu, A.; Scheau, C.; Nicolae, I.; Matei, A.; Caruntu, C.; et al. Current Perspectives on the Role of Matrix Metalloproteinases in the Pathogenesis of Basal Cell Carcinoma. *Biomolecules* **2021**, *11*, 903. [CrossRef]
11. Jansen, M.H.E.; Kessels, J.P.H.M.; Nelemans, P.J.; Kouloubis, N.; Arits, A.H.M.M.; van Pelt, H.P.A.; Quaedvlieg, P.J.F.; Essers, B.A.B.; Steijlen, P.M.; Kelleners-Smeets, N.W.J.; et al. Randomized Trial of Four Treatment Approaches for Actinic Keratosis. *N. Engl. J. Med.* **2019**, *380*, 935–946. [CrossRef]
12. Navsaria, L.J.; Li, Y.; Nowakowska, M.K.; Hinkston, C.L.; Wheless, L.; Giordano, S.H.; Wehner, M.R. Incidence and Treatment of Actinic Keratosis in Older Adults With Medicare Coverage. *JAMA Dermatol.* **2022**, *158*, 1076. [CrossRef] [PubMed]
13. Yeung, H.; Baranowski, M.L.; Swerlick, R.A.; Chen, S.C.; Hemingway, J.; Hughes, D.R.; Duszak, R. Use and Cost of Actinic Keratosis Destruction in the Medicare Part B Fee-for-Service Population, 2007 to 2015. *JAMA Dermatol.* **2018**, *154*, 1281. [CrossRef] [PubMed]
14. Dubois-Pot-Schneider, H.; Khairallah, G.; Brzenczek, C.; Plénat, F.; Marchal, F.; Amouroux, M. Transcriptomic Study on Human Skin Samples: Identification of Two Subclasses of Actinic Keratoses. *Int. J. Mol. Sci.* **2023**, *24*, 5937. [CrossRef]
15. Neagu, M.; Constantin, C.; Caruntu, C.; Dumitru, C.; Surcel, M.; Zurac, S. Inflammation: A Key Process in Skin Tumorigenesis (Review). *Oncol. Lett.* **2019**, *17*, 4068–4084. [CrossRef] [PubMed]
16. Voiculescu, V.; Calenic, B.; Ghita, M.; Lupu, M.; Caruntu, A.; Moraru, L.; Voiculescu, S.; Ion, A.; Greabu, M.; Ishkitiev, N.; et al. From Normal Skin to Squamous Cell Carcinoma: A Quest for Novel Biomarkers. *Dis. Markers* **2016**, *2016*, 4517492. [CrossRef]
17. Voiculescu, V.M.; Lisievici, C.V.; Lupu, M.; Vajaitu, C.; Draghici, C.C.; Popa, A.V.; Solomon, I.; Sebe, T.I.; Constantin, M.M.; Caruntu, C. Mediators of Inflammation in Topical Therapy of Skin Cancers. *Mediat. Inflamm.* **2019**, *2019*, 8369690. [CrossRef]
18. Daneluzzi, C.; Seyed Jafari, S.M.; Hunger, R.; Bossart, S. The Immunohistochemical Assessment of Neoangiogenesis Factors in Squamous Cell Carcinomas and Their Precursors in the Skin. *J. Clin. Med.* **2022**, *11*, 4494. [CrossRef]
19. Lu, K.; Bhat, M.; Peters, S.; Mitra, R.; Oberyszyn, T.; Basu, S. Suppression of Beta 2 Adrenergic Receptor Actions Prevent UVB Mediated Cutaneous Squamous Cell Tumorigenesis through Inhibition of VEGF—A Induced Angiogenesis. *Mol. Carcinog.* **2021**, *60*, 172–178. [CrossRef]
20. Forsea, A.-M. Melanoma Epidemiology and Early Detection in Europe: Diversity and Disparities. *Dermatol. Pract. Concept.* **2020**, *10*, e2020033. [CrossRef]
21. Garbe, C.; Keim, U.; Gandini, S.; Amaral, T.; Katalinic, A.; Hollezcek, B.; Martus, P.; Flatz, L.; Leiter, U.; Whiteman, D. Epidemiology of Cutaneous Melanoma and Keratinocyte Cancer in White Populations 1943–2036. *Eur. J. Cancer* **2021**, *152*, 18–25. [CrossRef]
22. Bray, F.; Ferlay, J.; Soerjomataram, I.; Siegel, R.L.; Torre, L.A.; Jemal, A. Global Cancer Statistics 2018: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. CA. *Cancer J. Clin.* **2018**, *68*, 394–424. [CrossRef] [PubMed]
23. Guo, W.; Wang, H.; Li, C. Signal Pathways of Melanoma and Targeted Therapy. *Signal Transduct. Target. Ther.* **2021**, *6*, 424. [CrossRef]
24. Strashilov, S.; Yordanov, A. Aetiology and Pathogenesis of Cutaneous Melanoma: Current Concepts and Advances. *Int. J. Mol. Sci.* **2021**, *22*, 6395. [CrossRef] [PubMed]
25. Mihulecea, C.-R.; Rotaru, M. Review: The Key Factors to Melanomagenesis. *Life* **2023**, *13*, 181. [CrossRef]
26. Surcel, M.; Constantin, C.; Caruntu, C.; Zurac, S.; Neagu, M. Inflammatory Cytokine Pattern Is Sex-Dependent in Mouse Cutaneous Melanoma Experimental Model. *J. Immunol. Res.* **2017**, *2017*, 1–10. [CrossRef]
27. Caruntu, C. Catecholamines Increase in Vitro Proliferation of Murine B16F10 Melanoma Cells. *Acta Endocrinol.* **2014**, *10*, 545–558. [CrossRef]

28. Scheau, C.; Draghici, C.; Ilie, M.A.; Lupu, M.; Solomon, I.; Tampa, M.; Georgescu, S.R.; Caruntu, A.; Constantin, C.; Neagu, M.; et al. Neuroendocrine Factors in Melanoma Pathogenesis. *Cancers* **2021**, *13*, 2277. [CrossRef] [PubMed]
29. Rajkomar, A.; Dean, J.; Kohane, I. Machine Learning in Medicine. *N. Engl. J. Med.* **2019**, *380*, 1347–1358. [CrossRef]
30. Foahom Gouabou, A.C.; Collenne, J.; Monnier, J.; Iguernaissi, R.; Damoiseaux, J.-L.; Moudafi, A.; Merad, D. Computer Aided Diagnosis of Melanoma Using Deep Neural Networks and Game Theory: Application on Dermoscopic Images of Skin Lesions. *Int. J. Mol. Sci.* **2022**, *23*, 13838. [CrossRef]
31. Dobre, E.-G.; Surcel, M.; Constantin, C.; Ilie, M.A.; Caruntu, A.; Caruntu, C.; Neagu, M. Skin Cancer Pathobiology at a Glance: A Focus on Imaging Techniques and Their Potential for Improved Diagnosis and Surveillance in Clinical Cohorts. *Int. J. Mol. Sci.* **2023**, *24*, 1079. [CrossRef]
32. Lupu, M.; Popa, I.M.; Voiculescu, V.M.; Caruntu, A.; Caruntu, C. A Systematic Review and Meta-Analysis of the Accuracy of in VivoReflectance Confocal Microscopy for the Diagnosis of Primary Basal Cell Carcinoma. *J. Clin. Med.* **2019**, *8*, 1462. [CrossRef] [PubMed]
33. Lupu, M.; Caruntu, C.; Popa, M.; Voiculescu, V.; Zurac, S.; Boda, D. Vascular Patterns in Basal Cell Carcinoma: Dermoscopic, Confocal and Histopathological Perspectives (Review). *Oncol. Lett.* **2019**, *17*, 4112–4125. [CrossRef]
34. Moscarella, E.; Rabinovitz, H.; Zalaudek, I.; Piana, S.; Stanganelli, I.; Oliviero, M.C.; Lallas, A.; Ardigo, M.; Cota, C.; Catricalà, C.; et al. Dermoscopy and Reflectance Confocal Microscopy of Pigmented Actinic Keratoses: A Morphological Study. *J. Eur. Acad. Dermatol. Venereol.* **2015**, *29*, 307–314. [CrossRef]
35. Peppelman, M.; Nguyen, K.P.; Hoogedoorn, L.; Van Erp, P.E.J.; Gerritsen, M.J.P. Reflectance Confocal Microscopy: Non-Invasive Distinction between Actinic Keratosis and Squamous Cell Carcinoma. *J. Eur. Acad. Dermatol. Venereol.* **2015**, *29*, 1302–1309. [CrossRef]
36. Lupu, M.; Caruntu, A.; Boda, D.; Caruntu, C. In Vivo Reflectance Confocal Microscopy-Diagnostic Criteria for Actinic Cheilitis and Squamous Cell Carcinoma of the Lip. *J. Clin. Med.* **2020**, *9*, 1987. [CrossRef]
37. Ferreira, P.S.; Rocha, L.; Bezerra, A.P.C.; Nico, M.M.S.; Lourenço, S.V. Reflectance Confocal Microscopy (RCM)-Based Criteria for Progression of Lower-Lip Squamous Cell Carcinoma: A Prospective Study. *Oral Oncol.* **2022**, *125*, 105674. [CrossRef]
38. Ianoși, S.; Batani, A.; Ilie, M.; Tampa, M.; Georgescu, S.-R.; Zurac, S.; Boda, D.; Ianosi, N.; Neagoe, D.; Calina, D.; et al. Non-Invasive Imaging Techniques for the In vivo Diagnosis of Bowen's Disease: Three Case Reports. *Oncol. Lett.* **2019**, *17*, 4094–4101. [CrossRef]
39. Diaconeasa, A.; Boda, D.; Neagu, M.; Constantin, C.; Căruntu, C.; Vlădău, L.; Guţu, D. The Role of Confocal Microscopy in the Dermato-Oncology Practice. *J. Med. Life* **2011**, *4*, 63–74. [PubMed]
40. Ianoși, S.; Forsea, A.; Lupu, M.; Ilie, M.; Zurac, S.; Boda, D.; Ianosi, G.; Neagoe, D.; Tutunaru, C.; Popa, C.; et al. Role of Modern Imaging Techniques for the in Vivo Diagnosis of Lichen Planus (Review). *Exp. Ther. Med.* **2018**, *17*, 1052–1060. [CrossRef]
41. Kadouch, D.J.; Schram, M.E.; Leeflang, M.M.; Limpens, J.; Spuls, P.I.; de Rie, M.A. In Vivo Confocal Microscopy of Basal Cell Carcinoma: A Systematic Review of Diagnostic Accuracy. *J. Eur. Acad. Dermatol. Venereol.* **2015**, *29*, 1890–1897. [CrossRef] [PubMed]
42. Nguyen, K.P.; Peppelman, M.; Hoogedoorn, L.; Van Erp, P.E.J.; Gerritsen, M.-J.P. The Current Role of in Vivo Reflectance Confocal Microscopy within the Continuum of Actinic Keratosis and Squamous Cell Carcinoma: A Systematic Review. *Eur. J. Dermatol.* **2016**, *26*, 549–565. [CrossRef] [PubMed]
43. Stevenson, A.D.; 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. [CrossRef] [PubMed]
44. Pezzini, C.; Kaleci, S.; Chester, J.; Farnetani, F.; Longo, C.; Pellacani, G. Reflectance Confocal Microscopy Diagnostic Accuracy for Malignant Melanoma in Different Clinical Settings: Systematic Review and Meta-Analysis. *J. Eur. Acad. Dermatol. Venereol.* **2020**, *34*, 2268–2279. [CrossRef]
45. Pellacani, G.; Pepe, P.; Casari, A.; Longo, C. Reflectance Confocal Microscopy as a Second-Level Examination in Skin Oncology Improves Diagnostic Accuracy and Saves Unnecessary Excisions: A Longitudinal Prospective Study. *Br. J. Dermatol.* **2014**, *171*, 1044–1051. [CrossRef] [PubMed]
46. Stanganelli, I.; Longo, C.; Mazzoni, L.; Magi, S.; Medri, M.; Lanzanova, G.; Farnetani, F.; Pellacani, G. Integration of Reflectance Confocal Microscopy in Sequential Dermoscopy Follow-up Improves Melanoma Detection Accuracy. *Br. J. Dermatol.* **2015**, *172*, 365–371. [CrossRef]
47. Longo, C.; Lallas, A.; Kyrgidis, A.; Rabinovitz, H.; Moscarella, E.; Ciardo, S.; Zalaudek, I.; Oliviero, M.; Losi, A.; Gonzalez, S.; et al. Classifying Distinct Basal Cell Carcinoma Subtype by Means of Dermatoscopy and Reflectance Confocal Microscopy. *J. Am. Acad. Dermatol.* **2014**, *71*, 716–724. [CrossRef]
48. Lupu, M.; Popa, I.M.; Voiculescu, V.M.; Boda, D.; Caruntu, C.; Zurac, S.; Giurcaneanu, C. A Retrospective Study of the Diagnostic Accuracy of In Vivo Reflectance Confocal Microscopy for Basal Cell Carcinoma Diagnosis and Subtyping. *J. Clin. Med.* **2019**, *8*, 449. [CrossRef]
49. Marconi, A.; Quadri, M.; Farnetani, F.; Ciardo, S.; Palazzo, E.; Lotti, R.; Cesinaro, A.M.; Fabbiani, L.; Vaschieri, C.; Puviani, M.; et al. In Vivo Melanoma Cell Morphology Reflects Molecular Signature and Tumor Aggressiveness. *J. Investig. Dermatol.* **2022**, *142*, 2205–2216. [CrossRef]

50. Lupu, M.; Voiculescu, V.M.; Caruntu, A.; Tebeica, T.; Caruntu, C. Preoperative Evaluation through Dermoscopy and Reflectance Confocal Microscopy of the Lateral Excision Margins for Primary Basal Cell Carcinoma. *Diagnostics* **2021**, *11*, 120. [CrossRef]
51. Durkin, J.R.; Tchanque-Fossuo, C.N.; Rose, A.N.; Elwood, H.R.; Stepenaskie, S.; Barbosa, N.S. Surgical Margin Mapping of Melanoma In Situ Using In Vivo Reflectance Confocal Microscopy Mosaics. *Dermatol. Surg.* **2021**, *47*, 605–608. [CrossRef]
52. Longo, C.; Casari, A.; Pepe, P.; Moscarella, E.; Zalaudek, I.; Argenziano, G.; Pellacani, G. Confocal Microscopy Insights into the Treatment and Cellular Immune Response of Basal Cell Carcinoma to Photodynamic Therapy. *Dermatology* **2012**, *225*, 264–270. [CrossRef] [PubMed]
53. Navarrete-Dechent, C.; Cordova, M.; Liopyris, K.; Aleissa, S.; Rajadhyaksha, M.; Cohen, G.; Marghoob, A.A.; Rossi, A.M.; Barker, C.A. In Vivo Imaging Characterization of Basal Cell Carcinoma and Cutaneous Response to High-Dose Ionizing Radiation Therapy: A Prospective Study of Reflectance Confocal Microscopy, Dermoscopy, and Ultrasonography. *J. Am. Acad. Dermatol.* **2021**, *84*, 1575–1584. [CrossRef] [PubMed]
54. Sierra, H.; Yélamos, O.; Cordova, M.; Chen, C.-S.J.; Rajadhyaksha, M. Reflectance Confocal Microscopy-Guided Laser Ablation of Basal Cell Carcinomas: Initial Clinical Experience. *J. Biomed. Opt.* **2017**, *22*, 1–13. [CrossRef]
55. Ahlgrimm-Siess, V.; Horn, M.; Koller, S.; Ludwig, R.; Gerger, A.; Hofmann-Wellenhof, R. Monitoring Efficacy of Cryotherapy for Superficial Basal Cell Carcinomas with In Vivo Reflectance Confocal Microscopy: A Preliminary Study. *J. Dermatol. Sci.* **2009**, *53*, 60–64. [CrossRef]
56. Villani, A.; Fabbrocini, G.; Costa, C.; Scalvenzi, M. Reflectance Confocal Microscopy Identification of Subclinical Basal Cell Carcinoma after Vismodegib Treatment: Report of a Case. *Dermatol. Ther.* **2021**, *11*, 1071–1074. [CrossRef] [PubMed]
57. Curiel-Lewandrowski, C.; Myrdal, C.N.; Saboda, K.; Hu, C.; Arzberger, E.; Pellacani, G.; Legat, F.J.; Ulrich, M.; Hochfellner, P.; Oliviero, M.C.; et al. In Vivo Reflectance Confocal Microscopy as a Response Monitoring Tool for Actinic Keratoses Undergoing Cryotherapy and Photodynamic Therapy. *Cancers* **2021**, *13*, 5488. [CrossRef]
58. Ribero, S.; Marra, E.; Tomasini, C.F.; Fierro, M.T.; Bombonato, C.; Longo, C. Confocal Microscopy and Dermoscopy for the Monitoring of BRAF Inhibitor Therapy of Melanoma Skin Metastases. *Br. J. Dermatol.* **2017**, *176*, 1101–1102. [CrossRef]
59. Hibler, B.P.; Yélamos, O.; Cordova, M.; Sierra, H.; Rajadhyaksha, M.; Nehal, K.S.; Rossi, A.M. Handheld Reflectance Confocal Microscopy to Aid in the Management of Complex Facial Lentigo Maligna. *Cutis* **2017**, *99*, 346–352.
60. Cinotti, E.; Labeille, B.; Debarbieux, S.; Carrera, C.; Lacarrubba, F.; Witkowski, A.M.; Moscarella, E.; Arzberger, E.; Kittler, H.; Bahadoran, P.; et al. Dermoscopy vs. Reflectance Confocal Microscopy for the Diagnosis of Lentigo Maligna. *J. Eur. Acad. Dermatol. Venereol.* **2018**, *32*, 1284–1291. [CrossRef]
61. Wei, W.; Ehlerding, E.B.; Lan, X.; Luo, Q.; Cai, W. PET and SPECT Imaging of Melanoma: The State of the Art. *Eur. J. Nucl. Med. Mol. Imaging* **2018**, *45*, 132–150. [CrossRef]
62. Beltrami, E.J.; Brown, A.C.; Salmon, P.J.M.; Leffell, D.J.; Ko, J.M.; Grant-Kels, J.M. Artificial Intelligence in the Detection of Skin Cancer. *J. Am. Acad. Dermatol.* **2022**, *87*, 1336–1342. [CrossRef] [PubMed]
63. Jones, O.T.; Matin, R.N.; van der Schaar, M.; Prathivadi Bhayankaram, K.; Ranmuthu, C.K.I.; Islam, M.S.; Behiyat, D.; Boscott, R.; Calanzani, N.; Emery, J.; et al. Artificial Intelligence and Machine Learning Algorithms for Early Detection of Skin Cancer in Community and Primary Care Settings: A Systematic Review. *Lancet Digit. Health* **2022**, *4*, e466–e476. [CrossRef]
64. Gao, T.; Liu, S.; Gao, E.; Wang, A.; Tang, X.; Fan, Y. Automatic Segmentation of Laser-Induced Injury OCT Images Based on a Deep Neural Network Model. *Int. J. Mol. Sci.* **2022**, *23*, 11079. [CrossRef] [PubMed]
65. Guo, Y.; Song, Y.; Xiong, S.; Wang, T.; Liu, W.; Yu, Z.; Ma, X. Mechanical Stretch Induced Skin Regeneration: Molecular and Cellular Mechanism in Skin Soft Tissue Expansion. *Int. J. Mol. Sci.* **2022**, *23*, 9622. [CrossRef]
66. Radwan, A.M.; Zide, M.F. Tissue Expansion in the Head and Neck. *Atlas Oral Maxillofac. Surg. Clin.* **2019**, *27*, 167–173. [CrossRef]
67. Wu, D.; Yan, C.; Gao, H.; Liu, Z.; Sun, Y.; Xu, L.; Xie, F.; Gao, B.; Li, Q.; Zhu, X.; et al. Tissue Expansion Techniques in Reconstructive Surgery: A 10-Year Bibliometric Analysis. *Ann. Transl. Med.* **2023**, *11*, 204. [CrossRef] [PubMed]
68. Yu, Z.; Liu, S.; Cui, J.; Song, Y.; Wang, T.; Song, B.; Peng, P.; Ma, X. Early Histological and Ultrastructural Changes in Expanded Murine Scalp. *Ultrastruct. Pathol.* **2020**, *44*, 141–152. [CrossRef]
69. Liu, S.; Ding, J.; Zhang, Y.; Cheng, X.; Dong, C.; Song, Y.; Yu, Z.; Ma, X. Establishment of a Novel Mouse Model for Soft Tissue Expansion. *J. Surg. Res.* **2020**, *253*, 238–244. [CrossRef]
70. Antonopoulou, M.; Vlastos, D.; Dormousoglou, M.; Bouras, S.; Varela-Athanasatou, M.; Bekakou, I.-E. Genotoxic and Toxic Effects of The Flame Retardant Tris(Chloropropyl) Phosphate (TCPP) in Human Lymphocytes, Microalgae and Bacteria. *Toxics* **2022**, *10*, 736. [CrossRef]
71. Liu, J.-X.; Cui, D.-L.; Yang, D.-L.; Li, J.-Y.; Yang, Z.-Y.; Su, J.-Z.; Ren, C.-X.; Niu, Y.-Y.; Xiang, P. Organophosphorus Flame Retardant TCPP Induces Cellular Senescence in Normal Human Skin Keratinocytes: Implication for Skin Aging. *Int. J. Mol. Sci.* **2022**, *23*, 14306. [CrossRef]
72. Wang, A.S.; Dreesen, O. Biomarkers of Cellular Senescence and Skin Aging. *Front. Genet.* **2018**, *9*, 247. [CrossRef]
73. Ho, C.Y.; Dreesen, O. Faces of Cellular Senescence in Skin Aging. *Mech. Ageing Dev.* **2021**, *198*, 111525. [CrossRef] [PubMed]
74. Ganceviciene, R.; Liakou, A.I.; Theodoridis, A.; Makrantonaki, E.; Zouboulis, C.C. Skin Anti-Aging Strategies. *Dermatoendocrinol.* **2012**, *4*, 308–319. [CrossRef]
75. Csekes, E.; Račková, L. Skin Aging, Cellular Senescence and Natural Polyphenols. *Int. J. Mol. Sci.* **2021**, *22*, 12641. [CrossRef]

76. Surowiecka, A.; Strużyna, J. Adipose-Derived Stem Cells for Facial Rejuvenation. *J. Pers. Med.* **2022**, *12*, 117. [CrossRef]
77. Chen, S.; He, Z.; Xu, J. Application of Adipose-Derived Stem Cells in Photoaging: Basic Science and Literature Review. *Stem Cell Res. Ther.* **2020**, *11*, 491. [CrossRef] [PubMed]
78. Qin, F.; Zhang, W.; Zhang, M.; Long, X.; Si, L.; Li, Z.; Huang, J.; Wang, X. Adipose-Derived Stem Cells Improve the Aging Skin of Nude Mice by Promoting Angiogenesis and Reducing Local Tissue Water. *Aesthetic Surg. J.* **2021**, *41*, NP905–NP913. [CrossRef] [PubMed]
79. Mazini, L.; Rochette, L.; Admou, B.; Amal, S.; Malka, G. Hopes and Limits of Adipose-Derived Stem Cells (ADSCs) and Mesenchymal Stem Cells (MSCs) in Wound Healing. *Int. J. Mol. Sci.* **2020**, *21*, 1306. [CrossRef] [PubMed]
80. Long, C.; Wang, J.; Gan, W.; Qin, X.; Yang, R.; Chen, X. Therapeutic Potential of Exosomes from Adipose-Derived Stem Cells in Chronic Wound Healing. *Front. Surg.* **2022**, *9*, 1030288. [CrossRef]
81. Oh, S.; Kim, H.M.; Batsukh, S.; Sun, H.J.; Kim, T.; Kang, D.; Son, K.H.; Byun, K. High-Intensity Focused Ultrasound Induces Adipogenesis via Control of Cilia in Adipose-Derived Stem Cells in Subcutaneous Adipose Tissue. *Int. J. Mol. Sci.* **2022**, *23*, 8866. [CrossRef]
82. Maranduca, M.; Hurjui, L.; Branisteanu, D.; Serban, D.; Branisteanu, D.; Dima, N.; Serban, I. Skin—A Vast Organ with Immunological Function (Review). *Exp. Ther. Med.* **2020**, *20*, 18–23. [CrossRef] [PubMed]
83. Scheau, C.; Caruntu, C.; Badarau, I.A.; Scheau, A.-E.; Docea, A.O.; Calina, D.; Caruntu, A. Cannabinoids and Inflammations of the Gut-Lung-Skin Barrier. *J. Pers. Med.* **2021**, *11*, 494. [CrossRef] [PubMed]
84. Celebi Sozener, Z.; Ozdel Ozturk, B.; Cerci, P.; Turk, M.; Gorgulu Akin, B.; Akdis, M.; Altiner, S.; Ozbey, U.; Ogulur, I.; Mitamura, Y.; et al. Epithelial Barrier Hypothesis: Effect of the External Exposome on the Microbiome and Epithelial Barriers in Allergic Disease. *Allergy* **2022**, *77*, 1418–1449. [CrossRef]
85. Celebi Sozener, Z.; Özbey Yücel, Ü.; Altiner, S.; Ozdel Oztürk, B.; Cerci, P.; Türk, M.; Gorgülü Akin, B.; Akdis, M.; Yilmaz, I.; Ozdemir, C.; et al. The External Exposome and Allergies: From the Perspective of the Epithelial Barrier Hypothesis. *Front. Allergy* **2022**, *3*, 887672. [CrossRef]
86. Pat, Y.; Ogulur, I.; Yazici, D.; Mitamura, Y.; Cevhertas, L.; Küçükkase, O.C.; Mesisser, S.S.; Akdis, M.; Nadeau, K.; Akdis, C.A. Effect of Altered Human Exposome on the Skin and Mucosal Epithelial Barrier Integrity. *Tissue Barriers* **2022**, 2133877. [CrossRef] [PubMed]
87. Sparr, E.; Björklund, S.; Pham, Q.D.; Mojumdar, E.H.; Stenqvist, B.; Gunnarsson, M.; Topgaard, D. The Stratum Corneum Barrier—From Molecular Scale to Macroscopic Properties. *Curr. Opin. Colloid Interface Sci.* **2023**, *67*, 101725. [CrossRef]
88. Menon, G.K.; Cleary, G.W.; Lane, M.E. The Structure and Function of the Stratum Corneum. *Int. J. Pharm.* **2012**, *435*, 3–9. [CrossRef]
89. van Smeden, J.; Janssens, M.; Gooris, G.S.; Bouwstra, J.A. The Important Role of Stratum Corneum Lipids for the Cutaneous Barrier Function. *Biochim. Biophys. Acta-Mol. Cell Biol. Lipids* **2014**, *1841*, 295–313. [CrossRef] [PubMed]
90. Jiao, Q.; Yue, L.; Zhi, L.; Qi, Y.; Yang, J.; Zhou, C.; Jia, Y. Studies on Stratum Corneum Metabolism: Function, Molecular Mechanism and Influencing Factors. *J. Cosmet. Dermatol.* **2022**, *21*, 3256–3264. [CrossRef]
91. Danby, S.G.; Andrew, P.V.; Kay, L.J.; Pinnock, A.; Chittock, J.; Brown, K.; Williams, S.F.; Cork, M.J. Enhancement of Stratum Corneum Lipid Structure Improves Skin Barrier Function and Protects against Irritation in Adults with Dry, Eczema-prone Skin*. *Br. J. Dermatol.* **2022**, *186*, 875–886. [CrossRef]
92. Murphy, B.; Grimshaw, S.; Hoptroff, M.; Paterson, S.; Arnold, D.; Cawley, A.; Adams, S.E.; Falciani, F.; Dadd, T.; Eccles, R.; et al. Alteration of Barrier Properties, Stratum Corneum Ceramides and Microbiome Composition in Response to Lotion Application on Cosmetic Dry Skin. *Sci. Rep.* **2022**, *12*, 5223. [CrossRef]
93. Bratu, D.; Boda, D.; Caruntu, C. Genomic, Epigenomic, Transcriptomic, Proteomic and Metabolomic Approaches in Atopic Dermatitis. *Curr. Issues Mol. Biol.* **2023**, *45*, 5215–5231. [CrossRef]
94. Solomon, I.; Ilie, M.; Draghici, C.; Voiculescu, V.; Căruntu, C.; Boda, D.; Zurac, S. The Impact of Lifestyle Factors on Evolution of Atopic Dermatitis: An Alternative Approach (Review). *Exp. Ther. Med.* **2018**, *17*, 1078–1084. [CrossRef] [PubMed]
95. Montero-Vilchez, T.; Cuenca-Barrales, C.; Rodriguez-Pozo, J.-A.; Diaz-Calvillo, P.; Tercedor-Sanchez, J.; Martinez-Lopez, A.; Molina-Leyva, A.; Arias-Santiago, S. Epidermal Barrier Function and Skin Homeostasis in Atopic Dermatitis: The Impact of Age. *Life* **2022**, *12*, 132. [CrossRef] [PubMed]
96. Emmert, H.; Baurecht, H.; Thielking, F.; Stölzl, D.; Rodriguez, E.; Harder, I.; Proksch, E.; Weidinger, S. Stratum Corneum Lipidomics Analysis Reveals Altered Ceramide Profile in Atopic Dermatitis Patients across Body Sites with Correlated Changes in Skin Microbiome. *Exp. Dermatol.* **2021**, *30*, 1398–1408. [CrossRef]
97. van den Bogaard, E.H.; Elias, P.M.; Goleva, E.; Berdyshev, E.; Smits, J.P.H.; Danby, S.G.; Cork, M.J.; Leung, D.Y.M. Targeting Skin Barrier Function in Atopic Dermatitis. *J. Allergy Clin. Immunol. Pract.* **2023**, *11*, 1335–1346. [CrossRef] [PubMed]
98. Blaak, J.; Dähnhardt, D.; Bielfeldt, S.; Theiss, C.; Simon, I.; Wilhelm, K.-P.; Dähnhardt-Pfeiffer, S.; Staib, P. Improvement of Human Epidermal Barrier Structure and Lipid Profile in Xerotic- and Atopic-Prone Skin via Application of a Plant-Oil and Urea Containing PH 4.5 Emulsion. *Cosmetics* **2023**, *10*, 95. [CrossRef]

99. Shin, K.-O.; Ha, D.H.; Kim, J.O.; Crumrine, D.A.; Meyer, J.M.; Wakefield, J.S.; Lee, Y.; Kim, B.; Kim, S.; Kim, H.; et al. Exosomes from Human Adipose Tissue-Derived Mesenchymal Stem Cells Promote Epidermal Barrier Repair by Inducing de Novo Synthesis of Ceramides in Atopic Dermatitis. *Cells* **2020**, *9*, 680. [CrossRef]
100. Sjövall, P.; Gregoire, S.; Wargniez, W.; Skedung, L.; Luengo, G.S. 3D Molecular Imaging of Stratum Corneum by Mass Spectrometry Suggests Distinct Distribution of Cholesteryl Esters Compared to Other Skin Lipids. *Int. J. Mol. Sci.* **2022**, *23*, 13799. [CrossRef]