COMMENTARY

neoeptopes. In addition, this study demonstrates that the 15th collagenous domain is the major location of LABD neoeptopes, which could provide an explanation for the discrepant results regarding locations of LABD neoeptopes in previous studies (Nie et al., 2000; Yamauchi et al., 2014; Zillikens et al., 1999).

The finding that the conformation of the 15th collagenous domain is influenced by the C-terminal domain of BP180 that is located several hundred amino acids away is very intriguing. The triple helical structure of the neoeptope region may be loosened by loss of the C-terminal domain. More extensive studies should be performed to clarify how the N- and C-terminal domains of BP180 hide the LABD neoeptopes.

It is still difficult to explain why some conformations in the collagenous region of BP180 remained after extensive denaturation during the immunoblotting procedure, although Toyonaga et al. speculated about this as indicated above. The mechanisms responsible for this phenomenon require additional studies. Finally, it will be interesting to investigate whether different antibodies reactive with LAD-1 and LABD97 possess different pathogenic activities. If this is the case, there may be different clinical and histopathological features associated with patients with LABD whose IgA antibodies react predominantly with LAD-1 or LABD97.

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Conflicts of interest

The authors state no conflict of interest.

References


Marinkovich MP, Taylor TB, Keene DR, Burgeson RE, Zone JJ. LAD-1, the linear IgA bullous dermatosis autoantigen, is a novel 120-kDa anchoring filament protein synthesized by epidermal cells. J Invest Dermatol 1996;106:734–8.


Yamauchi T, Matushita S, Hashimoto T, Hirako Y. Major cleavage-dependent epitopes for linear IgA bullous dermatosis are formed at the boundary between the non-collagenous 16A and collagenous 15 domains of BP180.

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Negative Regulation of Skin Pigmentation in Three-Dimensional Reconstructions by Adipose-Derived Mesenchymal Cells

Xunwei Wu and David E. Fisher

The regulation of site-specific pigmentation in reconstructions used for skin grafting is incompletely understood. Using an engineered skin equivalent model, Klar et al. incorporated adipose-derived mesenchymal cells into skin substitutes and found that adipose-derived mesenchymal cells secreted high levels of transforming growth factor-β1, which down-regulates melanogenic enzymes such as tyrosinase to decrease melanin synthesis and prevent normal pigmentation of resulting skin grafts.

Clinical Implications

- Restoration of patient skin pigmentation in damaged areas is challenging.
- Autologous bioengineered skin grafts should ideally match native skin color.
- Incorporation of adipose derived mesenchymal cells (ASCs) inhibits normal pigmentation of skin grafts.

because of their abundance and accessibility without ethical concerns (Tosyekani et al., 2015; Klar et al., 2017a). In the present issue, Klar et al. (2017b) report that substituting ASCs for dermal fibroblasts alters pigmentation of engineered skin grafts.

Interest in engineered skin substitutes is predicated on longstanding evidence that autologous cutaneous cell populations can be harvested, propagated, differentiated, and reimplanted within individual patients as a means of skin replacement therapy. Although the earliest data focused on the efficacy of this approach for epithelial/keratinocyte populations, the ability to incorporate melanocytes has added the pigmented component to this strategy, with significant success. Therefore, it is important to know whether incorporation of ASCs could influence pigmentation of skin substitutes for clinical application in vivo.

Klar et al. (2017b) studied the influence of ASCs, derived from the subcutis, on melanocytes within engineered skin equivalent structures. The approach requires both epidermal and dermal contributions, and the ability of adipose mesenchymal cells to influence the melanocytic component is assessed. Klar et al. (2017b) show that substitution of adipose mesenchymal cells for dermal fibroblasts results in significant suppression of melanocytes and consequently diminished pigmentation. Their study documents decreased numbers of MITF+ melanocytes in dermal equivalents when adipose mesenchymal cells are included (in a dose-dependent fashion, even when mixed with dermal fibroblasts). They also find diminished expression of the differentiated melanocytic factors tyrosinase, TRP1, and Sox9. Conditioned media from cultured adipose mesenchymal cells also suppressed expression of melanocytic genes, and an examination of several known melanocyte antagonists showed strong expression of transforming growth factor (TGF)-β1 by these cells. Other known melanocytic antagonists were not produced at elevated levels by adipose mesenchymal cells compared with dermal fibroblasts, including DKK1 (the Wnt pathway inhibitor that has been implicated in palmar/plantar control of depigmentation).

The use of skin equivalents as a bioengineered approach to skin replacement represents a unique opportunity for patients suffering from skin conditions—an opportunity that is not routinely possible for diseases involving other organ systems. The ability to generate autologous grafts from genetically identical (self) donor tissue that can repair anatomic defects is uncommon in the modern medical armamentarium, although it certainly represents a holy grail of modern medical technology. This approach can restore skin barrier function and be lifesaving. The ability to restore additional features—including less life threatening ones such as melanocyte-derived pigmentation—provides significant benefits as well, and in other organ systems may be essential for restoring distinct functionalities. For this reason, it is important to understand the potential limitations of various approaches, ideally with some understanding of the underlying mechanistic details. The Klar et al. (2017b) study highlights the negative influence of adipose mesenchymal cells on the pigmentation compartment within skin equivalents, even if their support of epithelial regeneration may suffice. It is likely that limitations like this will be intrinsic to numerous bioengineered organ systems, as attempts are made to incorporate increasingly complex and valuable cellular subpopulations.

TGF-β has a well-documented role in the regulation of melanocytic homeostasis (Javelaud et al., 2008). Although adipose populations are unlikely to modulate such effects under normal in vivo conditions, TGF-β signaling has been shown to play a vital role in controlling melanocyte stem cell return to quiescence in the early anagen phase of the hair follicle cycle (Nishimura et al., 2010). The quiescent state includes suppression of MITF and its melanogenic targets. In the hair follicle niche, Bcl2 is required to prevent TGF-β signaling from inducing cell death—a known consequence of TGF-β in other cellular contexts. Thus, the role of adipose mesenchymal cell-derived TGF-β as an antagonist to melanocytic differentiation within three-dimensional reconstructions is a plausible explanation for the hypopigmentary effects.

An important implication of this work, as highlighted by the authors, is the need to reassess the contributions of dermal cells to epidermal function—in this case, pigmentation. Pigmentation patterns clearly vary across anatomic sites within individuals—much of this variation is currently unexplained. To what extent might dermal cells contribute? Could additional secreted factors such as DKK1, agouti signaling protein, and others participate in contexts beyond their documented roles at specific sites? This study (Klar et al., 2017b) focuses on the artificial setting of engineered tissue reconstructs, but the authors correctly point out the temptation to assume that donor cells from distinct anatomic locations will behave similarly—a prospect that will require additional study. Furthermore, in the context of melanoma, it is not uncommon for invasive or metastatic lesions to abut adipose tissue, and recent data have suggested that there may be cell-cell crosstalk that could modulate melanoma cell behavior. In instances like this, the mediators and phenotypic effects may vary, but the principal of microenvironmental contexts playing major roles in modulating (non-cell-autonomous) biological behaviors is well established and certain to be important in many biomedical settings.

CONFLICT OF INTEREST
The authors state no conflict of interest.

REFERENCES


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Lymphatic versus Hematogenous Melanoma Metastases: Support for Biological Heterogeneity without Clear Clinical Application

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Melanoma demonstrates considerable biological heterogeneity and is associated with several routes of dissemination including lymphatic and hematogenous. Locoregional control via surgery may improve outcomes for patients with limited lymphatic metastases. Once stage IV disease is diagnosed, clinical outcomes are determined by molecular and/or immunologic factors and identification of tumor/microenvironmental features correlating with distant metastases is critical for future prognostic stratification.


Characterizing patterns of metastatic dissemination and identifying molecular features correlating with site-specific metastases is crucial for optimal disease management and predicting responses to therapy in melanoma and other tumor types. It is clear from clinical experience that there are both hematogenous and lymphatic routes of metastasis, which may be of clinical relevance particularly in the setting of thick melanomas. Gassenmaier et al. (2017) perform a retrospective analysis of a large, prospectively maintained cohort of patients spanning nearly four decades (1976–2015) who presented initially with early stage (IA–IIC) primary cutaneous melanoma and subsequently progressed to either stage III or IV disease. The authors note three distinct subsets of disease progression: (i) isolated lymphatic spread without subsequent distant metastases, (ii) combined lymphatic and hematogenous distant metastases, and (iii) exclusive distant metastases. Their analysis excludes patients with isolated locoregional disease and focuses exclusively on patients with stage IV disease (Gassenmaier et al., 2017), demonstrating no difference in overall or metastasis-free survival in patients with stage IV disease regardless of the presence of previous or concurrent lymphatic spread. Although the authors describe a large clinical subset of patients with stage IV disease, it is critical to interpret these results with extreme caution for clinical use because the dataset spans an era of heterogeneous clinical management including the pre-sentinel lymph node biopsy (SLNB) era (1976–1996) in combination with the current era in which lymphatic staging has become the standard of care. Of note, patients with a positive sentinel lymph node (in the post-SLNB era) were excluded from analysis. Therefore, although descriptively interesting, these data cannot safely be applied to current clinical scenarios or patient management decisions.

The surgical approach to lymphatic management in melanoma has evolved over the last few decades. For the surgical oncologist, the goals of SLNB are to provide accurate staging and critical prognostic information. The modern era of the SLNB technique began in the early 1990s and has subsequently been adopted as an alternative to elective lymph node dissection (Morton et al., 1992). The Multicenter Selective Lymphadenectomy Trial (MSLT-I) randomized patients to either wide local excision and SLNB with immediate lymphadenectomy for a positive sentinel node or wide local excision and postoperative observation with