論　文　内　容　の　要　旨
Synopsis of Thesis

氏　　名 Name	Wuergezhen Duligengaowa
論文題名 Title	An *eGFP-Col4a2* mouse model reveals basement membrane dynamics underlying hair follicle morphogenesis （*eGFP-Col4a2* マウスモデルが明らかにする毛包形態形成を支える基底膜動態）

論文内容の要旨

〔目　的(Purpose)〕

This study investigates the dynamic role of the basement membrane (BM) in hair follicle morphogenesis, focusing on Collagen IV turnover and its impact on epithelial progenitor behavior and tissue shape. While the BM is known to be essential for morphogenesis, its dynamic characteristics remain poorly understood due to limited visualization techniques. To address this, we aimed to develop knock-in mice expressing fluorescently tagged endogenous Collagen IV (*eGFP-Col4a2*) and studied spatial and temporal changes in BM structure, turnover, and their contributions to BM expansion, cell division angles and organ shaping.

〔方法(Methods)〕

To visualize BM dynamics in live tissues, Knock-in mice expressing eGFP-tagged Collagen IV were developed. Fluorescent Recovery After Photobleaching (FRAP) and 3D confocal imaging of embryonic skin explants were used to measure BM turnover and expansion. *mKikGR-Col4a2* mice, expressing a photoconvertible version of Collagen IV, enabled tracking of pre-existing and newly incorporated Collagen IV proteins. To investigate the role of Matrix metalloproteinases (MMP) activity in BM remodeling, batimastat was used to inhibit MMP activity. Epithelial progenitor proliferation and cell division patterns were assessed with EdU incorporation and live imaging. Hair follicle morphology was quantified under control and MMP-inhibited conditions to understand the roles of BM dynamics in morphogenesis.

〔成績(Results)〕

This study revealed region-specific COL4A2 turnover rates in the BM, with the highest turnover occurring near the tips of developing hair follicles. BM expansion was closely associated with epithelial progenitor proliferation and directional movement. MMP inhibition reduced COL4A2 turnover and BM expansion, and caused abnormal hair follicle widening. These changes were linked to an increased perpendicular divisions of epithelial progenitors. Experiments with photoconvertible *mKikGR-Col4a2* mice demonstrated that the COL4A2 turnover occurrs via protein replacement, rather than diffusion. These findings underscore the critical role of MMP-dependent BM remodeling in normal morphogenesis, highlighting the interplay between BM expansion, epithelial cell behavior, and tissue shape.

〔総　括(Conclusion)〕

By establishing a live imaging system for BM molecules in tissues, this study reveals that BM is not a static structure but exhibits region-specific molecular turnover and expansion rates. These dynamic properties are driven by MMP-mediated extracellular matrix (ECM) cleavage, which plays a critical role in BM expansion, epithelial cell division and movement, ultimately shaping tissue structure. The development of *eGFP-Col4a2* and *mKikGR-Col4a2* mouse models provides powerful tools for future in vivo investigation of BM dynamics, such as wound healing. These findings enhance our understanding of how ECM remodeling orchestrate organ development, underscoring the important role of BM in integrating molecular turnover to tissue-level morphogenesis.

論文審査の結果の要旨及び担当者

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論文審査の結果の要旨

細胞接着の足場となる基底膜は、細胞外の非細胞性成分であり、ほぼ全ての生体組織の発生や恒常性の維持に不可欠である。しかし、これまでは、建物の床のような静的で受動的な細胞の支持構造と見なされ、基底膜に動的で積極的な役割があるとは考えられてこなかった。本研究においてWuergezhen氏は、全身の基底膜が蛍光で光る遺伝子組換えマウスの作製に成功し、それを用いた基底膜のライブイメージング技術を新たに開発した。この技術を活用し、皮膚の毛包の形態形成における基底膜の動態と機能を解析した結果、基底膜は、動き、伸びる性質を持つことが明らかとなった。さらに、その動的特性が毛包の領域ごとに異なることで、基底膜を足場とする上皮前駆細胞の移動や分裂様式、さらには上皮組織全体の形態が制御されていることが分かった。本研究成果は、形態形成における基底膜の動的な役割を示す重要な成果で、器官の発生や再生のメカニズムの解明に広く貢献すると期待される。以上の研究成果は、学位に値するものと認める。