BMB
Reports

BMB Rep. 2023; 56(1): 2-9
www.bmbreports.org

Invited Mini Review

Aging of hair follicle stem cells and their niches

Hansaem Jang[1,#], *Yemin Jo*[1,#], *Jung Hyun Lee*[2,3] *& Sekyu Choi*[1,4,5,6,*]

[1]Department of Life Sciences, Pohang University of Science and Technology (POSTECH), Pohang 37673, Korea, [2]Division of Dermatology, Department of Medicine, University of Washington, Seattle, WA 98109, [3]Institute for Stem Cell and Regenerative Medicine, University of Washington, Seattle, WA 98109, USA, [4]School of Medical Science and Engineering, Pohang University of Science and Technology (POSTECH), Pohang 37673, [5]School of Interdisciplinary Bioscience and Bioengineering, Pohang University of Science and Technology (POSTECH), Pohang 37673, [6]Institute for Convergence Research and Education in Advanced Technology (I_CREATE), Yonsei University, Incheon 21983, Korea

Hair follicles in the skin undergo cyclic rounds of regeneration, degeneration, and rest throughout life. Stem cells residing in hair follicles play a pivotal role in maintaining tissue homeostasis and hair growth cycles. Research on hair follicle aging and age-related hair loss has demonstrated that a decline in hair follicle stem cell (HFSC) activity with aging can decrease the regeneration capacity of hair follicles. This review summarizes our understanding of how age-associated HFSC intrinsic and extrinsic mechanisms can induce HFSC aging and hair loss. In addition, we discuss approaches developed to attenuate age-associated changes in HFSCs and their niches, thereby promoting hair regrowth. [BMB Reports 2023; 56(1): 2-9]

INTRODUCTION

Stem cells have self-renewal capacity and ability to generate functionally differentiated cell types. Major roles of adult stem cells are to maintain tissue homeostasis and repair damaged tissues (1). Indeed, various adult tissues contain adult stem cells, such as hematopoietic stem cells (HSCs), mesenchymal stem cells, neural stem cells (NSCs), muscle satellite cells, intestinal stem cells (ISCs), dental pulp stem cells, and skin stem cells (2-8). Different types of stem cells have been found in the skin, including those associated with hair, *i.e.*, hair follicle stem cells (HFSCs) known to regulate hair growth and melanocyte stem cells (MeSCs) associated with hair pigmentation (9, 10). Quiescent adult stem cells can be activated in response to physiological cues (11). For this transition to occur via intrinsic mechanisms of adult stem cells, these stem cells must communicate with their surrounding microenvironment, *i.e.*, the stem cell niche (12), which offers external cues that can maintain and control the activity of stem cells.

The concept of the stem cell niche was first suggested in 1978 by Schofield (13), who hypothesized that the niche was an environment provided by neighboring non-HSCs, in which HSCs would retain their self-renewal ability and adult stem cell identity. This hypothesis is now supported by many studies. The concept of the stem cell niche has been extended (14-18). Including cells directly adjacent to stem cells, the niche consists of stem cells' own differentiated progenies and other cellular components, *e.g.*, blood vessels, lymphatic capillaries, nerves, stromal cells, adipocytes, and tissue-resident immune cells such as regulatory T cells and macrophages (19, 20). In addition, secreted factors, the extracellular matrix (ECM), and various environmental signals such as those related to physical parameters (*e.g.*, stiffness and shear stress), hypoxia, and cellular metabolism are closely associated with the stem cell niche (19-21). Although stem cell niches share some common characteristics, niches around different adult stem cells (*e.g.*, HSCs, ISCs, NSCs, and HFSCs) have been defined and their specific functions have been determined (22, 23).

Several studies have shown that age-related changes occur in adult tissues. For example, in aged mice, the small intestine exhibits a decreased regenerative capacity and altered structure (24, 25). The regenerative capacity of muscles is also decreased in aged mice and the fate of their satellite cells is altered (26, 27). Moreover, gray hairs appear in aged mice owing to depletion of MeSCs (28). Thus, adult stem cells are clearly affected by aging, which in turn can affect the maintenance of tissue homeostasis and reduce the restorative capacity of an aged organism.

Adult stem cells residing in hair follicles (HFs), namely HFSCs, are also affected by aging (29, 30). During the aging process, hair becomes thinner, hair production decreases, and graying and hair loss (senescent baldness) occur. Thus, hair aging is an obvious symptom of aging in long-lived mammals (29, 31). Aged adult stem cells of HFs, especially HFSCs or MeSCs, have been associated with this hair aging phenotype (29, 32).

*Corresponding author. Tel: +82-54-279-2359; Fax: +82-54-279-2199; E-mail: sekyuchoi@postech.ac.kr
#These authors contributed equally to this work.

https://doi.org/10.5483/BMBRep.2022-0183

Received 25 October 2022, Revised 14 November 2022, Accepted 14 November 2022

Keywords: Adult stem cell, Aging, Hair follicle, Skin stem cell, Stem cell niche

ISSN: 1976-670X (electronic edition)

In this review, we focus on HFSCs and their aging. Moreover, we discuss various HFSC intrinsic and extrinsic mechanisms involved in the hair aging process and how they actively communicate. Some researchers are attempting to rejuvenate aged HFSCs to treat aging hair symptoms, which could help advance regeneration therapy. Therefore, we will also summarize studies on hair regeneration therapies.

HFSC AND HF REGENERATION

The HF is a unique organ that undergoes regeneration and regression within a lifetime in periodic cycles. The hair cycle consists of telogen (rest), anagen (growth), and catagen (regression) (Fig. 1) (33). According to a single cell transcriptome atlas of the skin during the hair growth cycle, several types of cells are found in the skin around HFs, including fibroblasts, keratinocytes, immune cells (macrophages, T cells, dendritic cells, and Langerhans cells), and neural crest-derived cells (melanocytes and Schwann cells) (Fig. 2) (34). In addition, dermal papillae (DPs), bulge cells, hair germ (HG), and sebaceous glands are present in HFs (34). Regeneration of HFs is regulated by HFSCs located in the bulge and HG, a distinct cell population that contacts DPs (9, 35-37). HFSC markers are known to include K15, Lgr5, Sox9, and Lhx2 (38-42). However, CD34 and Nfatc1 are only expressed in bulge HFSCs, whereas P-cadherin and Lef1 are expressed in HG. Thus, they are distinct expression markers that can distinguish functionally separated cells in the two regions (37, 43-45).

HFSCs are activated in early anagen for hair growth. In pulse-chase experiments, label-retaining cells were found in the bulge region and to a lesser extent in HG (35). BrdU-labeled cells indicating cell proliferation appeared earlier in HG than in bulge in the late telogen phase (37). Another proliferation marker, Ki67, is expressed in HG in the late telogen phase (37). Bulge HFSCs can promote new hair growth by proliferating during early anagen. They can preserve the quiescent state except during the early-to-mid anagen phase (46, 47). Indeed, HFSCs are considered to be comprised of two stem cell populations: quiescent stem cells (bulge) and primed stem cells (HG) (46). MeSCs located in the HF bulge region can produce hair pigmentation by providing melanocytes to the hair matrix during the hair growth cycle (48, 49). Similar to HFSCs, MeSCs are activated in early anagen to supply pigmentation to growing HFs (50).

Activation-quiescence transition of HFSCs is controlled by their local environment, *i.e.*, the HFSC niche. Indeed, DP-promoting signals such as those associated with fibroblast growth factors and bone morphogenetic protein (BMP) inhibitors can activate HFSCs and initiate hair regeneration (37, 51, 52). Immune cells such as macrophages and regulatory T cells contribute to HFSC quiescence and hair growth (53-56). Rather than simply being passive responders, HFSCs can also build their own niche and HFSCs can actively communicate with their niche to achieve tissue homeostasis and/or wound healing, similar to other adult stem cells (20). For instance, K6-positive inner bulge cells, which can maintain HFSC quiescence during telogen through elevated BMP6 and FGF18 signaling, are derived from the HFSC progenies of the previous anagen phase (57).

HFSC activity associated with HF regeneration is coordinated with remodeling of vasculature and the lymphatic capillary network. Thus, each process can affect the other through molecular cross-talk (58-60). Similarly, thickness of dermal adipose tissue is associated with hair growth cycle. Immature or mature adipocytes can affect HFSC activity (61-63). In addition, HFSCs

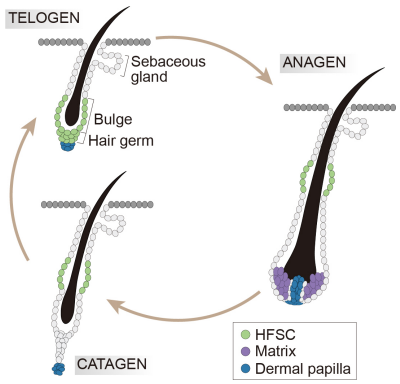

Fig. 1. Scheme of the hair follicle structures during the hair cycle. The hair follicle goes through cycles of telogen (resting), anagen (growth), catagen (regression). HFSCs (light green) are found in the bulge and hair germ in telogen hair follicle. The sebaceous gland is located above the bulge and the dermal papilla (blue) is below the hair germ. In anagen hair follicle, the matrix is produced by the proliferation of the hair germ.

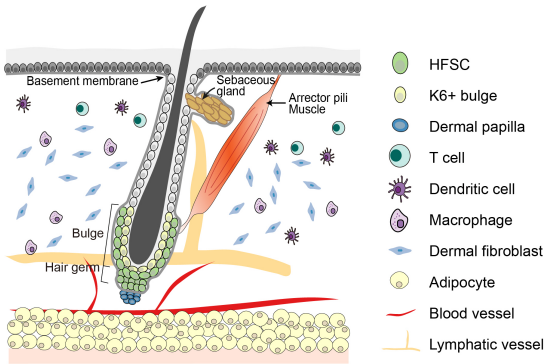

Fig. 2. HFSC and components of the HFSC niche. HFSCs (light green) are quiescent during the telogen phase and are found in the outer layer of the bulge and hair germ. K6+ inner bulge cells maintain the quiescence of HFSC in telogen. HFSC activity related to HF regeneration is coordinated with a number of HFSC niches. Around HFs, the skin contains several HFSC niches, including dermal papilla, dermal fibroblasts, immune cells (T cells, dendritic cells, and macrophages), dermal adipocytes, the arrector pili muscle, blood vessels and lymphatic vessels, which regulate HFSC activity.

construct the niche structure consisting of sympathetic nerves known to regulate HFSC activation via norepinephrine and the arrector pili muscle known to maintain innervation of sympathetic nerves (64, 65).

As described above, the HFSC niche is highly heterogeneous and complex. Therefore, several classifications have been used to explain the HFSC niche effectively. Cells in the niche have been distinguished as three functional modules (*i.e.*, signaling, sensory, and message-relaying modules), which are assembled into a multifunctional HFSC niche (66). Furthermore, because HFSCs reside in HFs and participate in HF cycling, extrinsic factors of HFSCs extend outside of HFs. Thus, the surrounding environment affecting HFSCs can be divided into the intrafollicular niche (*e.g.*, the $K6^+$ inner bulge layer and DP) and the extrafollicular macroenvironment (*e.g.*, the adipose tissue, immune system, or hormone system) (67, 68).

EFFECTS OF AGING ON HF REGENERATION

Changes in HFs occur with aging. In aged mice, telogen is more than doubled, anagen is slightly shortened, hair regeneration is delayed, hair is shorter and more scattered, and hair loss eventually occurs (29, 30, 68, 69). According to changes in hair cycle and hair appearance, HFs of aged mice exhibit extended dormancy (30, 68, 69). The activity of HFSCs and their capacity to form colonies are decreased in aged HFSCs than those in young HFSCs. In addition, aged HFSCs grow slower than young HFSCs *in vitro* (30). However, the transcriptional identity of HFSCs is maintained in aged mice (69). Importantly, changes occur in the activity of HFSCs rather than in the death of HFSCs in aged mice (30). Compared with young mice, aged mice exhibit no reduction in the number of $K15^+$ bulge stem cells (70). However, the number of HFSCs is decreased with aging (71). Considering other adult stem cells, the number of HSCs and their progenitor cells do not decrease sharply in aged mice. Conversely, aged mice exhibit an increase in the number of HSCs (72, 73). In contrast, the number of MeSCs in the skin decreases rapidly during aging. Depletion of MeSCs leads to graying hair (28). Although the number of stem cells does not always change during aging, stem cell populations are known to lose their function with aging (74). Loss of stem cell function can cause changes related to aging in tissues, which are derived from stem cells. Thus, the function rather than the number of HFSCs might be affected by aging. As stated in a previous study, two factors can affect the function of stem cells during aging: intrinsic factors and extrinsic factors (75).

INTRINSIC MECHANISMS RESPONSIBLE FOR HF AGING

BMP signaling is related to HFSC quiescence, whereas Wnt signaling is related to HFSC activation (76). Wnt signaling increases at the telogen-anagen transition, whereas BMP signaling increases during telogen when HFSCs are quiescent (77, 78). Wnt signaling and BMP signaling are known to maintain the activation-quiescence transition in HFSCs via a competitive interaction (79). Aged HFSCs can affect hair regeneration as the signal associated with stem cell activity changes. Wnt signaling can be divided into a canonical pathway related to β-catenin and a non-canonical pathway related to intracellular calcium (80). In aged HFSCs, activity of the non-canonical Wnt pathway, but not that of the canonical pathway, is increased. The expression of Wnt5a is also increased. In addition, polarity of HFSCs is altered by increasing the activity of Cdc42 via Wnt5a (81). Apolarity of aged HFSCs has been associated with Cdc42 activity. It is a condition under which the regenerative ability of HFSCs is decreased (81). Aging and increased Cdc42 activity might be related. Mice deficient in Cdc42GAP (a negative regulator of Cdc42) exhibits genome instability that can lead to premature aging (82). Similar to HFSCs, melanocytes express nuclear β-catenin during the growth of HFs, although it is no longer expressed after the late anagen phase. β-catenin deficiency in melanocytes is known to induce hair graying (48). However, overexpression of Wnt ligands in aged skin can induce differentiation of MeSCs, eventually leading to the production of gray hairs owing to the exhaustion of MeSCs (83). Wnt signaling not only affects HFSCs, but also affects the differentiation of MeSCs responsible for hair pigmentation (83).

Dephosphorylated nuclear factor of activated T cells, cytoplasmic (NFATc) proteins are produced in response to intracellular calcium ions and translocated to the nucleus where they can regulate gene transcription (84). NFATc1 can maintain HFSC quiescence by inhibiting the expression of Cdk4, which is related to G1/S phase progression in telogen (44). In addition, Nfatc1 levels are decreased to a lesser extent in aged HFSCs than in young HFSCs (30). Furthermore, when Nfatc1 is expressed in aged HFs, the number of proliferative cells and colony formation ability are reduced (30). Thus, NFATc1 might play a role in inhibiting the progression of anagen in aged HFSCs.

Foxc1 is known as a transcription factor associated with HFSC quiescence through direct regulation of Nfatc1 and Bmp6 (85). Thus, Foxc1 expression is correlated with BMP signaling as an intrinsic mechanism in HFSCs. In a Foxc1-knockout model, genes related to HFSC quiescence are downregulated, whereas genes related to cell cycle are upregulated (85). Foxc1-conditional knockout mice also show faster progression to the next hair cycle than control mice (86), which might have been due to failure to maintain the duration of telogen. Without Foxc1, HFs cannot produce more than one bulge. Since old bulge plays a role in regulating HFSC quiescence, HFSC expenditure in Foxc1-conditional knockout mice eventually leads to thinning hair as mice age (86).

SIRT7 can activate HFSCs by deacetylating NFATc1. The expression of Sirt7 decreases with age in HFSCs. Indeed, additional expression of Sirt7 in aged mice can induce hair regrowth and pigmentation (87). In summary, factors associated with Wnt and BMP signaling are altered by aging, which in turn can affect the function of HFSCs.

EXTRINSIC MECHANISMS REPONSIBLE FOR HF AGING

As activation of HFSCs is closely associated with their niche, extrinsic mechanisms also play important roles in HFSC aging. Several studies have demonstrated functional recovery of aged HFSCs relative to young HFSCs after their transplantation into the skin of a young mouse both *in vivo* and *in vitro* (88-91). Although the extrinsic mechanisms driving HFSC aging have not been clarified yet, several studies have shown the influence of extrinsic factors on HFSC aging.

Physical niche atrophy can promote HFSC aging. Xie *et al*. (71) have shown that the hair shaft could be a physical niche and that its miniaturization with aging could exert physical pressure on HFSCs, resulting in HFSC depletion. Within this process, mechanosensitive Piezo channels are activated by mechanical compression, which in turn can activate cellular TNF-α signaling and increase HFSC apoptosis during the catagen-telogen transition (71). Moreover, changes in compositions of the ECM also exert mechanical stress that ages HFSCs. Alternative expression of ECM components in an aged HFSC niche, especially those associated with the basement membrane (BM), can elevate BM stiffness. A stiffened BM can silence HFSC bivalent promoters important for HFSC activation (Fig. 3) (91). Therefore, HFSCs can lose their hair cycle activation ability. Thus, HFSC aging can be promoted via an extrinsic mechanism.

HFSC aging can also occur via various secreted factors that regulate key signaling pathways in the HF cycle. In the early anagen phase of an aged mouse skin, expression levels of canonical Wnt signaling inhibitors such as Dkk1 and Sfrp4 are increased, whereas the expression of follistatin, a BMP signaling inhibitor that promotes hair wave propagation, is decreased in the interorgan macroenvironment, especially in intradermal adipose tissues (89). Wnt signaling is known to be important for the regulation of HFSCs and promotion of the HF cycle, whereas BMP signaling has opposite effects (23, 92). Nevertheless, studies have suggested that expression changes in extrafollicular modulators can act on HFSC aging.

Several studies have shown that changes in inflammatory cytokine levels in the extrafollicular macroenvironment can affect HFSC aging. Doles *et al*. (93) have revealed that expression levels of various inflammatory cytokines are increased in an age-dependent manner in the epidermal layer of aged mouse skin. In addition, they found that an age-associated imbalance in epidermal JAK-STAT signaling could inhibit HFSC function (93). In aged mice, levels of inflammatory cytokines such as CXCL1 are also increased substantially in dermal white adipose tissues at telogen (94). Moreover, after treatment with veratric acid, which mitigates inflammation, HF regrowth is promoted in aged mice, suggesting that overactivated inflammation signaling could inhibit HFSC function with aging (94).

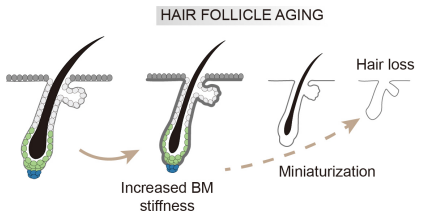

Fig. 3. Scheme for the hair follicle aging. The HFSC is kept in a prolonged quiescent state in the HF bulge during aging. The alternate ECM component expression in the aged HFSC niche result in an increase in basement membrane stiffness. Intrinsic and extrinsic mechanisms that influence HFSC aging cause the HF miniaturization as well as hair thinning and loss.

REJUVENATION OF AGE-ASSOCIATED HAIR LOSS

Aging of HFSCs can lead to hair senescence, which ultimately leads to age-related hair loss (*i.e.*, senile alopecia), one of the most obtrusive symptoms of aging. Several treatments including synthetic drugs such as minoxidil and finasteride have been approved by the FDA. Transplantation therapy can be employed for recovery from general alopecia (95, 96). However, drug treatments have major side effects. In addition, young or rejuvenated stem cell transplantation could not fully regenerate the hair cycle because HFSCs actively interact with their external environment (96-98).

Several approaches have been reported to rejuvenate aged-related hair loss. Age-associated increase of cytokine signaling, especially epidermal JAK-STAT signaling, can impede the function of HFSCs. Thus, pyridine 6, a JAK2 inhibitor, has been applied as a topical treatment to aged mouse skin. After one week of treatment, the number of active hair follicles was increased (93). Although this stimulation ultimately accelerates stem cell exhaustion, pharmacologic inhibitors of JAK-STAT signaling can also induce hair growth. Therefore, with further development, the use of pyridine 6 could help rejuvenate aged HFSCs (93, 97, 99).

Treatment with CASIN, a Cdc42 pharmacological inhibitor, can rejuvenate aged HFSCs because Cdc42 is a downstream molecule of noncanonical Wnt signaling, which hinders HFSC activation during the aging process (81). After aged HFSCs were treated with CASIN, the aging phenotype of HFSCs was reduced and the hair regrowth ability of aged mice was increased by the reactivation of Wnt canonical signaling, which facilitated HFSC activation (81).

Platelet-rich plasma therapy and mitochondrial transplantation are possible strategies for hair loss treatment as they both can activate HFSCs (100, 101). Indeed, they are effective treatments for hair aging. In particular, pep-1-mediated mitochondrial transplantation can induce hair regrowth in aged mice, help maintain hair length for longer, and yield an increased number of anagen follicles (102).

Photobiomodulation therapy (PBMT), which is an FDA-approved alopecia therapy, can decrease age-associated HF atrophy via intrinsic and extrinsic cues that regulate HFSCs (95). PBMT can stimulate cellular reactive oxygen species-activated intrinsic

signaling transduction that promotes aged HFSC proliferation and increase the HF-inducing ability of skin keratinocyte precursors and Wnt ligand secretion in the HFSC niche, thereby enhancing aged HFSC activation (95).

CONCLUSION

As discussed in this review, intrinsic and extrinsic mechanisms are known to affect HFSC aging. However, studies on specific treatments for age-related hair loss are still lacking. Although the change in the number of HFSCs with aging remains controversial, *e.g.*, in androgenic alopecia, HFSCs can be preserved in aged HFs with their own lineage identity (69). Therefore, we expect that rejuvenation of aged HFSCs based on mechanisms that cause HFSC aging will be effective in patients who suffer from age-related hair disorders. Future studies on age-associated mechanisms related to HFSCs will help us develop new therapeutics for maintaining proper HF function during aging.

ACKNOWLEDGEMENTS

We thank members of the Choi laboratory for their helpful discussions and comments on the manuscript. This work was supported by the National Research Foundation of Korea (NRF) grants funded by the Korea government (MSIT) (NRF-2022R1 C1C1011895 and NRF-2022M3A9D3016848) and Basic Science Research Institute Fund (NRF-2021R1A6A1A10042944). Y.J. was supported by a Genexine Research Fellowship Award. This work was also supported by a BK21 FOUR Research Fellowship funded by the Ministry of Education, Republic of Korea.

CONFLICTS OF INTEREST

The authors have no conflicting interests.

REFERENCES

1. Rando TA (2006) Stem cells, ageing and the quest for immortality. Nature 441, 1080-1086
2. Van Der Flier LG and Clevers H (2009) Stem cells, self-renewal, and differentiation in the intestinal epithelium. Annu Rev Physiol 71, 241-260
3. Temple S (2001) The development of neural stem cells. Nature 414, 112-117
4. Collins CA, Olsen I, Zammit PS et al (2005) Stem cell function, self-renewal, and behavioral heterogeneity of cells from the adult muscle satellite cell niche. Cell 122, 289-301
5. Pittenger MF, Mackay AM, Beck SC et al (1999) Multilineage potential of adult human mesenchymal stem cells. Science 284, 143-147
6. Weissman IL (2000) Stem cells: units of development, units of regeneration, and units in evolution. Cell 100, 157-168
7. Gronthos S, Mankani M, Brahim J, Robey PG and Shi S (2000) Postnatal human dental pulp stem cells (DPSCs) in vitro and in vivo. Proc Natl Acad Sci U S A 97, 13625-13630
8. Fuchs E (2008) Skin stem cells: rising to the surface. J Cell Biol 180, 273-284
9. Blanpain C, Lowry WE, Geoghegan A, Polak L and Fuchs E (2004) Self-renewal, multipotency, and the existence of two cell populations within an epithelial stem cell niche. Cell 118, 635-648
10. Inomata K, Aoto T, Binh NT et al (2009) Genotoxic stress abrogates renewal of melanocyte stem cells by triggering their differentiation. Cell 137, 1088-1099
11. Scadden DT (2014) Nice neighborhood: emerging concepts of the stem cell niche. Cell 157, 41-50
12. Scadden DT (2006) The stem-cell niche as an entity of action. Nature 441, 1075-1079
13. Schofield R (1978) The relationship between the spleen colony-forming cell and the haemopoietic stem cell. Blood Cells 4, 7-25
14. Kiger AA, White-Cooper H and Fuller MT (2000) Somatic support cells restrict germline stem cell self-renewal and promote differentiation. Nature 407, 750-754
15. Zhang J, Niu C, Ye L et al (2003) Identification of the haematopoietic stem cell niche and control of the niche size. Nature 425, 836-841
16. Tavazoie M, Van der Veken L, Silva-Vargas V et al (2008) A specialized vascular niche for adult neural stem cells. Cell Stem Cell 3, 279-288
17. Shin J, Berg DA, Zhu Y et al (2015) Single-cell RNA-seq with waterfall reveals molecular cascades underlying adult neurogenesis. Cell Stem Cell 17, 360-372
18. Rayagiri SS, Ranaldi D, Raven A et al (2018) Basal lamina remodeling at the skeletal muscle stem cell niche mediates stem cell self-renewal. Nat Commun 9, 1075
19. Lane SW, Williams DA and Watt FM (2014) Modulating the stem cell niche for tissue regeneration. Nat Biotechnol 32, 795-803
20. Fuchs E and Blau HM (2020) Tissue stem cells: architects of their niches. Cell Stem Cell 27, 532-556
21. Mohyeldin A, Garzon-Muvdi T and Quinones-Hinojosa A (2010) Oxygen in stem cell biology: a critical component of the stem cell niche. Cell Stem Cell 7, 150-161
22. Li L and Xie T (2005) Stem cell niche: structure and function. Annu Rev Cell Dev Biol 21, 605-631
23. Moore KA and Lemischka IR (2006) Stem cells and their niches. Science 311, 1880-1885
24. Martin K, Potten CS, Roberts SA and Kirkwood T (1998) Altered stem cell regeneration in irradiated intestinal crypts of senescent mice. J Cell Sci 111, 2297-2303
25. Nalapareddy K, Nattamai KJ, Kumar RS et al (2017) Canonical wnt signaling ameliorates aging of intestinal stem cells. Cell Rep 18, 2608-2621
26. Conboy IM, Conboy MJ, Smythe GM and Rando TA (2003) Notch-mediated restoration of regenerative potential to aged muscle. Science 302, 1575-1577
27. Brack AS, Conboy MJ, Roy S et al (2007) Increased Wnt signaling during aging alters muscle stem cell fate and increases fibrosis. Science 317, 807-810
28. Nishimura EK (2011) Melanocyte stem cells: a melanocyte reservoir in hair follicles for hair and skin pigmentation. Pigment Cell Melanoma Res 24, 401-410

29. Matsumura H, Mohri Y, Binh NT et al (2016) Hair follicle aging is driven by transepidermal elimination of stem cells via COL17A1 proteolysis. Science 351, aad4395
30. Keyes BE, Segal JP, Heller E et al (2013) Nfatc1 orchestrates aging in hair follicle stem cells. Proc Natl Acad Sci U S A 110, E4950-4959
31. Trueb RM (2006) Pharmacologic interventions in aging hair. Clin Interv Aging 1, 121-129
32. Nishimura EK, Granter SR and Fisher DE (2005) Mechanisms of hair graying: incomplete melanocyte stem cell maintenance in the niche. Science 307, 720-724
33. Müller-Röver S, Foitzik K, Paus R et al (2001) A comprehensive guide for the accurate classification of murine hair follicles in distinct hair cycle stages. J Invest Dermatol 117, 3-15
34. Joost S, Annusver K, Jacob T et al (2020) The molecular anatomy of mouse skin during hair growth and rest. Cell Stem Cell 26, 441-457 e7
35. Cotsarelis G, Sun TT and Lavker RM (1990) Label-retaining cells reside in the bulge area of pilosebaceous unit: implications for follicular stem cells, hair cycle, and skin carcinogenesis. Cell 61, 1329-1337
36. Tumbar T, Guasch G, Greco V et al (2004) Defining the epithelial stem cell niche in skin. Science 303, 359-363
37. Greco V, Chen T, Rendl M et al (2009) A two-step mechanism for stem cell activation during hair regeneration. Cell Stem Cell 4, 155-169
38. Morris RJ, Liu Y, Marles L et al (2004) Capturing and profiling adult hair follicle stem cells. Nat Biotechnol 22, 411-417
39. Liu Y, Lyle S, Yang Z and Cotsarelis G (2003) Keratin 15 promoter targets putative epithelial stem cells in the hair follicle bulge. J Invest Dermatol 121, 963-968
40. Jaks V, Barker N, Kasper M et al (2008) Lgr5 marks cycling, yet long-lived, hair follicle stem cells. Nat Genet 40, 1291-1299
41. Vidal VP, Chaboissier M-C, Lützkendorf S et al (2005) Sox9 is essential for outer root sheath differentiation and the formation of the hair stem cell compartment. Current Biology 15, 1340-1351
42. Rhee H, Polak L and Fuchs E (2006) Lhx2 maintains stem cell character in hair follicles. Science 312, 1946-1949
43. Shirokova V, Biggs LC, Jussila M, Ohyama T, Groves AK and Mikkola ML (2016) Foxi3 deficiency compromises hair follicle stem cell specification and activation. Stem Cells 34, 1896-1908
44. Horsley V, Aliprantis AO, Polak L, Glimcher LH and Fuchs E (2008) NFATc1 balances quiescence and proliferation of skin stem cells. Cell 132, 299-310
45. Morris RJ, Bortner CD, Cotsarelis G et al (2003) Enrichment for living murine keratinocytes from the hair follicle bulge with the cell surface marker CD34. J Invest Dermatol 120, 501-511
46. Hsu Y-C, Li L and Fuchs E (2014) Transit-amplifying cells orchestrate stem cell activity and tissue regeneration. Cell 157, 935-949
47. Wilson C, Cotsarelis G, Wei Z-G et al (1994) Cells within the bulge region of mouse hair follicle transiently proliferate during early anagen: heterogeneity and functional differences of various hair cycles. Differentiation 55, 127-136
48. Rabbani P, Takeo M, Chou W et al (2011) Coordinated activation of Wnt in epithelial and melanocyte stem cells initiates pigmented hair regeneration. Cell 145, 941-955
49. Tanimura S, Tadokoro Y, Inomata K et al (2011) Hair follicle stem cells provide a functional niche for melanocyte stem cells. Cell Stem Cell 8, 177-187
50. Nishimura EK, Jordan SA, Oshima H et al (2002) Dominant role of the niche in melanocyte stem-cell fate determination. Nature 416, 854-860
51. Jahoda CA, Horne KA and Oliver RF (1984) Induction of hair growth by implantation of cultured dermal papilla cells. Nature 311, 560-562
52. Rompolas P, Deschene ER, Zito G et al (2012) Live imaging of stem cell and progeny behaviour in physiological hair-follicle regeneration. Nature 487, 496-499
53. Paus R, Maurer M, Slominski A and Czarnetzki BM (1994) Mast cell involvement in murine hair growth. Dev Biol 163, 230-240
54. Castellana D, Paus R and Perez-Moreno M (2014) Macrophages contribute to the cyclic activation of adult hair follicle stem cells. PLoS Biol 12, e1002002
55. Ali N, Zirak B, Rodriguez RS et al (2017) Regulatory T cells in skin facilitate epithelial stem cell differentiation. Cell 169, 1119-1129 e11
56. Wang ECE, Dai Z, Ferrante AW, Drake CG and Christiano AM (2019) A subset of TREM2(+) dermal macrophages secretes oncostatin M to maintain hair follicle stem cell quiescence and inhibit hair growth. Cell Stem Cell 24, 654-669 e656
57. Hsu YC, Pasolli HA and Fuchs E (2011) Dynamics between stem cells, niche, and progeny in the hair follicle. Cell 144, 92-105
58. Li KN, Jain P, He CH, Eun FC, Kang S and Tumbar T (2019) Skin vasculature and hair follicle cross-talking associated with stem cell activation and tissue homeostasis. Elife 8, e45977
59. Gur-Cohen S, Yang H, Baksh SC et al (2019) Stem cell-driven lymphatic remodeling coordinates tissue regeneration. Science 366, 1218-1225
60. Pena-Jimenez D, Fontenete S, Megias D et al (2019) Lymphatic vessels interact dynamically with the hair follicle stem cell niche during skin regeneration in vivo. EMBO J 38, e101688
61. Plikus MV, Mayer JA, de la Cruz D et al (2008) Cyclic dermal BMP signalling regulates stem cell activation during hair regeneration. Nature 451, 340-344
62. Festa E, Fretz J, Berry R et al (2011) Adipocyte lineage cells contribute to the skin stem cell niche to drive hair cycling. Cell 146, 761-771
63. Shook B, Rivera Gonzalez G, Ebmeier S, Grisotti G, Zwick R and Horsley V (2016) The role of adipocytes in tissue regeneration and stem cell niches. Annu Rev Cell Dev Biol 32, 609-631
64. Fujiwara H, Ferreira M, Donati G et al (2011) The basement membrane of hair follicle stem cells is a muscle cell niche. Cell 144, 577-589
65. Shwartz Y, Gonzalez-Celeiro M, Chen C-L et al (2020) Cell types promoting goosebumps form a niche to

regulate hair follicle stem cells. Cell 182, 578-593 e519
66. Chen CL, Huang WY, Wang EHC, Tai KY and Lin SJ (2020) Functional complexity of hair follicle stem cell niche and therapeutic targeting of niche dysfunction for hair regeneration. J Biomed Sci 27, 43
67. Chen C-C, Plikus MV, Tang P-C, Widelitz RB and Chuong CM (2016) The modulatable stem cell niche: tissue interactions during hair and feather follicle regeneration. J Mol Biol 428, 1423-1440
68. Choi S, Zhang B, Ma S et al (2021) Corticosterone inhibits GAS6 to govern hair follicle stem-cell quiescence. Nature 592, 428-432
69. Ge Y, Miao Y, Gur-Cohen S et al (2020) The aging skin microenvironment dictates stem cell behavior. Proc Natl Acad Sci U S A 117, 5339-5350
70. Giangreco A, Qin M, Pintar JE and Watt FM (2008) Epidermal stem cells are retained in vivo throughout skin aging. Aging Cell 7, 250-259
71. Xie Y, Chen D, Jiang K et al (2022) Hair shaft miniaturization causes stem cell depletion through mechanosensory signals mediated by a Piezo1-calcium-TNF-alpha axis. Cell Stem Cell 29, 70-85 e6
72. Geiger H and Van Zant G (2002) The aging of lympho-hematopoietic stem cells. Nat Immunol 3, 329-333
73. Sudo K, Ema H, Morita Y and Nakauchi H (2000) Age-associated characteristics of murine hematopoietic stem cells. J Exp Med 192, 1273-1280
74. Schultz MB and Sinclair DA (2016) When stem cells grow old: phenotypes and mechanisms of stem cell aging. Development 143, 3-14
75. Liu L and Rando TA (2011) Manifestations and mechanisms of stem cell aging. J Cell Biol 193, 257-266
76. Kobielak K, Stokes N, de la Cruz J, Polak L and Fuchs E (2007) Loss of a quiescent niche but not follicle stem cells in the absence of bone morphogenetic protein signaling. Proc Natl Acad Sci U S A 104, 10063-10068
77. Lien W-H, Polak L, Lin M, Lay K, Zheng D and Fuchs E (2014) In vivo transcriptional governance of hair follicle stem cells by canonical Wnt regulators. Nat Cell Biol 16, 179-190
78. Daszczuk P, Mazurek P, Pieczonka TD, Olczak A, Boryń ŁM and Kobielak K (2020) An intrinsic oscillation of gene networks inside hair follicle stem cells: an additional layer that can modulate hair stem cell activities. Front Cell Dev Biol 8, 595178
79. Kandyba E, Leung Y, Chen YB, Widelitz R, Chuong C-M and Kobielak K (2013) Competitive balance of intrabulge BMP/Wnt signaling reveals a robust gene network ruling stem cell homeostasis and cyclic activation. Proc Natl Acad Sci U S A 110, 1351-1356
80. Veeman MT, Axelrod JD and Moon RT (2003) A second canon: functions and mechanisms of β-catenin-independent Wnt signaling. Dev Cell 5, 367-377
81. Tiwari RL, Mishra P, Martin N et al (2021) A Wnt5a-Cdc42 axis controls aging and rejuvenation of hair-follicle stem cells. Aging (Albany NY) 13, 4778
82. Wang L, Yang L, Debidda M, Witte D and Zheng Y (2007) Cdc42 GTPase-activating protein deficiency promotes genomic instability and premature aging-like phenotypes. Proc Natl Acad Sci U S A 104, 1248-1253
83. Zhang Z, Lei M, Xin H et al (2017) Wnt/β-catenin signaling promotes aging-associated hair graying in mice. Oncotarget 8, 69316
84. Crabtree GR and Olson EN (2002) NFAT signaling: choreographing the social lives of cells. Cell 109, 67-79
85. Wang L, Siegenthaler JA, Dowell RD and Yi R (2016) Foxc1 reinforces quiescence in self-renewing hair follicle stem cells. Science 351, 613-617
86. Lay K, Kume T and Fuchs E (2016) FOXC1 maintains the hair follicle stem cell niche and governs stem cell quiescence to preserve long-term tissue-regenerating potential. Proc Natl Acad Sci U S A 113, 1506-1515
87. Li G, Tang X, Zhang S et al (2020) SIRT7 activates quiescent hair follicle stem cells to ensure hair growth in mice. EMBO J 39, e104365
88. Cao W, Li L, Kajiura S et al (2016) Aging hair follicles rejuvenated by transplantation to a young subcutaneous environment. Cell Cycle 15, 1093-1098
89. Chen CC, Murray PJ, Jiang TX et al (2014) Regenerative hair waves in aging mice and extra-follicular modulators follistatin, dkk1, and sfrp4. J Invest Dermatol 134, 2086-2096
90. Ji J, Ho BS, Qian G, Xie XM, Bigliardi PL and Bigliardi-Qi M (2017) Aging in hair follicle stem cells and niche microenvironment. J Dermatol 44, 1097-1104
91. Koester J, Miroshnikova YA, Ghatak S et al (2021) Niche stiffening compromises hair follicle stem cell potential during ageing by reducing bivalent promoter accessibility. Nat Cell Biol 23, 771-781
92. Lei M and Chuong CM (2016) Stem cells. Aging, alopecia, and stem cells. Science 351, 559-560
93. Doles J, Storer M, Cozzuto L, Roma G and Keyes WM (2012) Age-associated inflammation inhibits epidermal stem cell function. Genes Dev 26, 2144-2153
94. Chen J, Fan ZX, Zhu DC et al (2021) Emerging role of dermal white adipose tissue in modulating hair follicle development during aging. Front Cell Dev Biol 9, 728188
95. Jin H, Zou Z, Chang H, Shen Q, Liu L and Xing D (2021) Photobiomodulation therapy for hair regeneration: A synergetic activation of beta-CATENIN in hair follicle stem cells by ROS and paracrine WNTs. Stem Cell Reports 16, 1568-1583
96. Hu XM, Li ZX, Zhang DY et al (2021) A systematic summary of survival and death signalling during the life of hair follicle stem cells. Stem Cell Res Ther 12, 453
97. Neves J, Sousa-Victor P and Jasper H (2017) Rejuvenating strategies for stem cell-based therapies in aging. Cell Stem Cell 20, 161-175
98. Narbonne P (2018) The effect of age on stem cell function and utility for therapy. Cell Med 10, 215517901 8773756
99. Harel S, Higgins CA, Cerise JE et al (2015) Pharmacologic inhibition of JAK-STAT signaling promotes hair growth. Sci Adv 1, e1500973
100. Cervelli V, Garcovich S, Bielli A et al (2014) The effect of autologous activated platelet rich plasma (AA-PRP) injection on pattern hair loss: clinical and histomorphometric evaluation. Biomed Res Int 2014, 760709
101. Son MJ, Jeong JK, Kwon Y et al (2018) A novel and safe

small molecule enhances hair follicle regeneration by facilitating metabolic reprogramming. Exp Mol Med 50, 1-15
102. Wu HC, Fan X, Hu CH et al (2020) Comparison of mitochondrial transplantation by using a stamp-type multineedle injector and platelet-rich plasma therapy for hair aging in naturally aging mice. Biomed Pharmacother 130, 110520