OPEN ACCESS Freely available online

Hair Therapy & Transplantation

ISSN: 2167-0951

Short Communication

Recent Updates on Development of Wnt/β-catenin Pathway Activators for Alopecia Treatment

Soung-Hoon Lee[1], Kang-Yell Choi[1,2*]

[1]CK Regeon Inc., B137 Engineering Research Park, Seoul, South Korea; [2]Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University, Seoul, South Korea

ABSTRACT

Wnt/β-catenin signaling pathway plays central roles in regulating homeostasis and regeneration in the hair follicular stem cell niche. Thus, this pathway should be used as a therapeutic target for alopecia, but there are still limitations. Valproic Acid (VPA), an activator of canonical Wnt signaling, induced hair re-growth and regeneration in mice, but had limited clinical efficacy. This might be due to the induction of CXXC-type zinc finger protein 5 (CXXC5), a negative regulatory factor of this regenerative pathway and hair follicle regeneration, by VPA. To overcome the limitations caused by this negative regulatory factor, a competing peptide and small molecular compounds that inhibit CXXC5 function were developed, and they were found to restore the canonical Wnt pathway and hair follicle regeneration suppressed by CXXC5 in bald scalps. Collectively, the development of agents that inhibit CXXC5 function might provide an efficient and safe way to treat alopecia.

Keywords: Hair follicle regeneration; Wnt signaling activator; Valproic acid; Alopecia treatment; CXXC-type zinc finger protein 5

DESCRIPTION

Wnt/β-catenin signaling pathway is requisite for the self-renewal, differentiation of hair follicular stem cells and plays pivotal roles in regulating homeostasis and regeneration in the hair follicular stem cell niche [1-3]. Therefore, this pathway could be exploited as a therapeutic target for alopecia, but it is still limited. Based on the significance of this regenerative pathway in hair follicle regeneration, there has been a long-standing effort to develop hair growth-promoting agents targeting this pathway.

Efficacy and limitation of VPA in hair follicle regeneration

Valproic acid (VPA), which activates the canonical Wnt signaling by functioning glycogen synthase kinase 3β (GSK3β), has been widely used for the treatment of psychiatric disorders over the past decades and has proven to be safe [4]. Given the close association between the canonical Wnt pathway and hair follicle regeneration, VPA was tested in mice and found to induce hair re-growth and regeneration in mouse skins [3]. Especially, VPA increased the expression of hair induction markers and hair follicular stem cell markers with the activation of this regenerative pathway [3]. However, VPA has shown limited clinical efficacy, such as a minor effect on hair diameter, which can be considered as a measure of hair follicle regeneration [5], and this might be attributed to negative regulators of the Wnt/β-catenin signaling.

CXXC5: A Negative Regulator of Wnt/β-Catenin Pathway and Hair Follicle Regeneration

CXXC-type zinc finger protein 5 (CXXC5) was first characterized as a negative regulatory factor of the Wnt signaling and bone function through interaction with Dishevelled (Dvl) [6]. Consistently, we found the role of this protein as a negative feedback regulator of the canonical Wnt pathway in unwounded and wounded skins [7-9]. Specifically, the expression of CXXC5 was triggered in human dermal papilla or mouse skins by Wnt3a or VPA as a negative feedback mechanism [9]. Subsequently, this inhibitory protein was found to significantly suppress the canonical Wnt/β-catenin pathway activation, alkaline phosphatase (ALP) activity, and hair follicle regeneration by binding to Dvl both *in-vitro* and *in-vivo* [9]. The inhibitory role of

Correspondence to: Kang-Yell Choi, Department of Biotechnology, Yonsei University, Seoul, South Korea, Tel:; E-mail: kychoi@yonsei.ac.kr

Received: 02-May-2023, Manuscript No. HTT-23-23924; **Editor assigned:** 04-May-2023, PreQC No: HTT-23-23924 (PQ); **Reviewed:** 19-May-2023, QC No. HTT-23-23924; **Revised:** 26-May-2023, Manuscript No: HTT-23-23924 (R); **Published:** 02-Jun-2023; DOI: 10.35248/2167-0951.23.13.212

Citation: Lee SH (2023) Recent Updates on Development of Wnt/β-catenin Pathway Activators for Alopecia Treatment Hair Ther Trasplant. 13:212.

his protein in hair follicle regeneration was further addressed by studies using *Cxxc5*$^{-/-}$ mice, which displayed promotion of both hair re-growth and wound-induced hair follicle regeneration [9]. Moreover, analysis using human alopecia patient samples revealed that CXXC5 was overexpressed in the miniaturized follicles and arrector pili muscles of alopecia patients, which are known to be the structures prominently observed in alopecia [9]. Taken together, there is a need to use agents inhibiting CXXC5 function in combination with Wnt signaling activators to improve clinical outcomes in the treatment of alopecia.

A competitor peptide, the protein transduction domain fusion Dvl binding motif (PTD-DBM), was designed to inhibit the interaction of CXXC5 with Dvl in various damaged tissues of the patients where CXXC5 is overexpressed. This peptide consists of a PTD for efficient delivery and a DBM to specifically compete for binding of CXXC5 to Dvl [6-9]. PTD-DBM induced the canonical Wnt signaling activation and ALP activity by disrupting the binding of CXXC5 to Dvl in human dermal papilla cells. The promoting effect of this peptide on hair regeneration was further validated by using the mouse wound-induced hair follicle regeneration model [9]. Importantly, co-treatment with PTD-DBM and VPA synergistically induced this regenerative pathway and ALP activity both *in-vitro* and *in-vivo* [9]. In addition, our recent study also demonstrated that PTD-DBM restored the canonical Wnt pathway and hair follicle regeneration which were suppressed by prostaglandin D2 (PGD2), a major factor causing alopecia [10]. Considering that PGD2 exacerbates symptoms such as sebaceous hyperplasia in alopecia [10], the CXXC5-Dvl interaction could be a potential therapeutic target for the development of agents to treat alopecia.

Small Compounds for Hair Follicle Regeneration by Targeting CXXC5-Dvl Interaction

To overcome the limitations of use of the peptide as a therapeutic agent, an *in-vitro* screening assay was developed to monitor the interaction between CXXC5 and Dvl, and effective antagonists of this interaction were screened from small molecule libraries through development and usage of a methodology monitoring the CXXC5-Dvl Protein-Protein Interaction (PPI) [11]. KY19382, a small molecular compound obtained using this screening system, increased the canonical Wnt signaling and ALP activity in human dermal papilla cells by disrupting the binding of CXXC5 to Dvl [12]. Furthermore, KY19382 alone was sufficient to significantly induce hair re-growth and wound-induced hair follicle regeneration due to its effective dual-targeting ability to inhibit both GSK-3β activity and CXXC5–Dvl interaction [12]. Notably, KY19382 exhibited regenerative therapeutic effects in various types of alopecia in which CXXC5 is highly expressed [10,13]. For instance, dihydrotestosterone (DHT)-induced androgenetic alopecia phenotype was significantly alleviated by KY19382 treatment through inhibition of the CXXC5-Dvl interaction and restoration of the lowered Wnt signaling pathway [10]. The diabetes-induced alopecia phenotype was also markedly mitigated by KY19382 treatment through a similar mechanism [13]. These combined findings suggest that compounds that inhibit CXXC5 function as a Wnt/β-catenin signaling regulator could be used for the treatment of various types of alopecia.

CONCLUSION

The Wnt/β-catenin signaling activator induces regeneration of hair follicles as well as re-growth of hair by increasing the expression of its multiple target genes involving hair growth through a mechanism different from minoxidil, which is commonly used to treat alopecia. However, differently to expectations, topical VPA treatment in alopecia patients showed mild adverse effects including ventricular tachycardia and showed weak improvements in hair diameter and linear hair growth rate compared to the placebo group.

After efforts to overcome the limitations of VPA in clinical practice, CXXC5, the negative regulator of the canonical Wnt signaling pathway and hair follicle regeneration, was discovered, and small molecular compounds that inhibit the function of CXXC5 were finally developed. Since these compounds restore the canonical Wnt signaling pathway, which is lowered by CXXC5, rather than directly activating this pathway, they appear to be safer than direct Wnt signaling activators. Their safety was thoroughly verified through preclinical studies, including studies using Cxxc5$^{-/-}$ mice. Overall, agents that inhibit CXXC5 function could potentially be used to treat alopecia without side effects.

ACKNOWLEDGEMENTS

S-H Lee and K-Y Choi wrote the manuscript

SOURCES OF FUNDING

This work was supported by the National Research Foundation of Korea (NRF) grant funded by the Korean Government (MSIP) (2020M3E5E2040018). SHL was supported by National Research Foundation of Korea (NRF) through Creative Challenge Research Foundation Support Project (2022R1I1A1A01073599).

CONFLICT OF INTEREST

K-Y Choi is the CEO of CK Regeon Inc. (Seoul, Korea), which holds a license to develop and use KY19382 disclosed in the publication. The authors have declared that no competing interests exist.

REFERENCES

1. Ito M, Yang Z, Andl T, Cui C, Kim N, Millar SE, et al. Wnt-dependent *de novo* hair follicle regeneration in adult mouse skin after wounding. Nature. 2007;447(7142):316-320.
2. Lee SH, Zahoor M, Hwang JK, Min do S, Choi KY. Valproic acid induces cutaneous wound healing *in-vivo* and enhances keratinocyte motility. PLoS One. 2012;7(11):e48791.
3. Lee SH, Yoon J, Shin SH, Zahoor M, Kim HJ, Park PJ, et al. Valproic acid induces hair regeneration in murine model and activates

alkaline phosphatase activity in human dermal papilla cells. PLoS One. 2012;7(4):e34152.

4. Jung GA, Yoon JY, Moon BS, Yang DH, Kim HY, Lee SH, et al. Valproic acid induces differentiation and inhibition of proliferation in neural progenitor cells *via* the beta-catenin-Ras-ERK-p21Cip/WAF1 pathway. BMC Cell Biol. 2008;9-66.
5. Jo SJ, Shin H, Park YW, Paik SH, Park WS, Jeong YS, et al. Topical valproic acid increases the hair count in male patients with androgenetic alopecia: a randomized, comparative, clinical feasibility study using phototrichogram analysis. J Dermatol. 2014;41(4): 285-291.
6. Kim HY, Yoon JY, Yun JH, Cho KW, Lee SH, Rhee YM, et al. CXXC5 is a negative-feedback regulator of the Wnt/β-catenin pathway involved in osteoblast differentiation. Cell Death Differ. 2015;22(6): 912-920.
7. Lee SH, Kim MY, Kim HY, Lee YM, Kim H, Nam KA, et al. The Dishevelled-binding protein CXXC5 negatively regulates cutaneous wound healing. J Exp Med. 2015;212(7):1061-1080.
8. Lee SH, An S, Ryu YC, Seo SH, Park S, Lee MJ, et al. Adhesive Hydrogel Patch-Mediated Combination Drug Therapy Induces Regenerative Wound Healing through Reconstruction of Regenerative Microenvironment. Adv Healthc Mater. 2023;e2203094.
9. Lee SH, Seo SH, Lee DH, Pi LQ, Lee WS, Choi KY. Targeting of CXXC5 by a Competing Peptide Stimulates Hair Regrowth and Wound-Induced Hair Neogenesis. J Invest Dermatol. 2017;137(11): 2260-2269.
10. Ryu YC, Park J, Kim YR, Choi S, Kim GU, Kim E, et al. CXXC5 Mediates DHT-Induced Androgenetic Alopecia *via* PGD2. Cells. 2023;12(4):555.
11. Choi S, Kim HY, Cha PH, Seo SH, Lee C, Choi Y, et al. CXXC5 mediates growth plate senescence and is a target for enhancement of longitudinal bone growth. Life Sci Alliance. 2019;2(2):e201800254.
12. Ryu YC, Lee DH, Shim J, Park J, Kim YR, Choi S, et al. KY19382, a novel activator of Wnt/β-catenin signaling, promotes hair regrowth and hair follicle neogenesis. Br J Pharmacol. 2021;178(12): 2533-2546.
13. Ryu YC, Kim YR, Park J, Choi S, Kim GU, Kim E, et al. Wnt/β-catenin signaling activator restores hair regeneration suppressed by diabetes mellitus. BMB Rep. 2022;55(11):559-564.