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Hair regeneration using microneedle-assisted human basic fibroblast growth factor

Abstract

Hair loss has afflicted numerous patients for several decades. However, the current drug treatment is only effective for specific types of alopecia and their efficacy is limited. Additionally, the cost of hair transplant surgery is relatively high, making it unaffordable for some patients. In this study, we propose a new treatment method using microneedles and growth factors to alleviate alopecia. The results of our study demonstrate that patients receiving human basic fibroblast growth factor (hbFGF) therapy experienced significant hair regeneration. Both male and female patients responded positively to the growth factor treatment, with males showing better results. Importantly, microneedle-assisted hbFGF therapy can be universally applied to androgenetic alopecia, alopecia areata, and telogen effluvium. In summary, this study presents a novel and highly effective method that shows great potential for treating alopecia, with high levels of safety and efficacy.

Keywords: alopecia, hair loss, microneedle, human basic fibroblast growth factor, hair regeneration

Volume 12 Issue 2 - 2023

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Received: July 15, 2023 | **Published:** August 02, 2023

Abbreviations: hbFGF, human basic fibroblast growth factor; DHT, dihydrotestosterone; FDA, Food and Drug Administration; PRP, platelet-rich plasma; bFGF, basic fibroblast growth factor; SEM, standard error of the mean

Introduction

Alopecia, also known as hair loss, is a common dermatological disorder with high prevalence rates in recent years.[1,2] Alopecia can be classified into cicatricial alopecia and non-cicatricial alopecia based on the presence or absence of scarring in the affected areas. Cicatricial alopecia, also known as scarring alopecia, involves the destruction and replacement of hair follicles by scar tissue, leading to irreversible hair loss.[3] Non-cicatricial alopecia refers to hair loss where the hair follicles are still intact, and there is a possibility of hair regeneration.[4] Common types of non-cicatricial alopecia include androgenetic alopecia, alopecia areata, and telogen effluvium.[5] Androgenetic alopecia and alopecia areata have been extensively investigated in scientific research and the clinical field. The prevalence rates of androgenetic alopecia in Chinese men are 19.9%,[6] and 2% among global alopecia areata patients.[7]

The pathogenesis of non-scarring alopecia is dependent on the specific type of hair loss. Androgenetic alopecia is the most common type of hair loss which is characterized by a progressive thinning of hair on the crown in men, and diffuse thinning all over the scalp in women.[8,9] Genetic and hormonal factors contribute to the development of androgenetic alopecia. Men are more prone to suffer from hair loss if their fathers have been diagnosed with androgenetic alopecia.[10] The main hormone involved in androgenetic alopecia is dihydrotestosterone (DHT) which is a potent derivative of testosterone. For patients with a genetic predisposition, the number of androgen receptors is increased in the affected scalp, and hair follicles in the scalp are more sensitive to DHT which leads to the miniaturization of hair follicles. This miniaturization process results in thinner, shorter, and fewer hairs.[11,12]

Alopecia areata is an autoimmune disease in which the immune system attacks the hair follicles erroneously and leads to round or oval-shaped patches of hair loss.[13] T lymphocytes infiltrate the hair follicles, causing inflammation and destruction of the hair follicle structure. Affected hair follicles enter the telogen phase and lead to hair shaft breaking.[14] Telogen effluvium is a common type of hair loss that is often triggered by physical or emotional stress, hormonal changes, medication, or nutritional deficiencies. Hair follicles prematurely enter the resting phase (telogen) and shed excessive hair.[15,16] Besides, other factors such as inflammatory processes, oxidative stress, and mechanical factors may also contribute to alopecia.[17,18]

Currently, only a limited number of drugs have been approved by the FDA for the treatment of alopecia, including Finasteride, Minoxidil, and Olumiant.[19,20] These drugs can only be applied to specific types of alopecia, and the effectiveness of the therapy may vary among individuals. Hair transplant surgery is an effective strategy for hair regeneration that involves removing hair follicles from the back or sides of the scalp and transplanting them into the balding areas while the cost may not be affordable for some alopecia patients.[21] Similarly, platelet-rich plasma (PRP) therapy, which entails extracting a patient's blood, processing it to concentrate the platelets, and subsequently injecting the platelet-rich plasma into the affected areas of the scalp, shows promise in providing a curative effect for patients with androgenetic alopecia.[22] However, it is important to note that the cost of this therapy can amount to ten thousand dollars.[23] In recent years stem cell therapy has emerged as a promising approach to the treatment of alopecia.[24] By harvesting stem cells from the patient's own body or healthy donor, such as from the scalp or adipose tissue, and then processing and injecting them into the affected areas, hair regeneration is initiated.[25] Stem cell therapy is considered a safe and minimally invasive option with the potential for long-lasting results, yet only tens of clinical trials have been approved by FDA which indicates the clinically immature approach. Thus, safe, effective, and universally applied therapies are highly desired for alopecia treatment.

Basic fibroblast growth factor (bFGF) plays a crucial role in embryonic development,[26] cell proliferation,[27] angiogenesis,[28] and tissue repair.[29] Previous studies have elaborated on the significance of bFGF in hair regeneration, both in vitro and in animals. Topically applied bFGF increased hair follicle number and hair regeneration in mice dependent on the β-catenin pathway.[30] Medium conditioned by adipose-derived stem cells was enriched in bFGF which promoted the human dermal papilla cell proliferation in vitro and hair growth in mice.[31]

However, limited research has been conducted on the influence of bFGF on human alopecia participants. Early-stage male androgenetic alopecia patients receiving bFGF combined with minoxidil treatment showed better therapeutic effects than minoxidil treatment alone groups.[32] It remains elusive whether bFGF can be adopted as a novel therapy for different types of alopecia patients.

In this study, we utilized a micro-needling device to create tiny wounds and then applied human basic fibroblast growth factor enriched serum to the scalps of alopecia patients for treatment. After 4-10 sessions of growth factor therapy, the alopecia regions showed a dramatic decrease among all the participants. Interestingly, male participants showed a better response to the treatment compared to female participants. Divergent types of alopecia patients, including androgenetic alopecia, alopecia areata, and telogen effluvium, experienced hair shaft regeneration after the growth factor-based therapy which implies the efficacy of hbFGF treatment in restoring hair growth. Notably, the growth factor therapy was efficient in hair restoration and relieving hair loss for patients of different ages and various alopecia types which may provide a promising and universal applied therapy.

Material and methods

Patients characteristics

12 alopecia patients (5 female and 7 male) aged 25 to 55 were enrolled in this study. Age, gender, genetics, medical history, treatment scheme (number of therapies and time duration), and treatment outcome were recorded.

Growth factor treatment

Alopecia patients were subjected to growth factor treatment. The scalp was cleaned and sanitized thoroughly to prevent any infection or contamination. The micro-needling device was sterilized before use. Then the micro-needling device with tiny needles was used to create small puncture wounds on the scalp. The device was moved in a circular motion across each section of the scalp, creating tiny channels in the skin. After the micro-needling process was completed, a serum containing human basic fibroblast growth factor was applied to the scalp. The hbFGF serum was manufactured by DreamTec Research Limited and the major ingredient consisted of human basic fibroblast growth factor, biotinoyl tripeptide-1, myristoyl pentapeptide-17, and Copper peptide GHK-Cu. Patients received 4-10 sessions of growth-factor treatment for 2-4 months.

Quantification of alopecia area

Images of 12 patients were taken before and after growth factor treatment. The photographs were captured by the identical operator under the same conditions as the patients' position. ImageJ software was utilized to quantify the alopecia area. In brief, the contour of the hair loss area was depicted with "Freehand selections" in ImageJ and then the area was quantified and recorded. The decrease of alopecia lesions was determined by (Alopecia area before treatment-Alopecia area after treatment)/ Alopecia area before treatment.

Statistical Analysis

All data were presented as the mean ± standard error of the mean (SEM) and the statistics were analyzed by GraphPad Prism 7 (San Diego, CA, USA). Paired student t-test was performed to determine the significant difference in the alopecia area before and after growth factor therapy. One-way ANOVA with Tukey's post hoc test was utilized to evaluate the significant difference in the decrease of alopecia lesions among different groups. It would be considered a significant difference when the p-value was smaller than 0.05. *, $p < 0.05$, * *, $p < 0.01$, * * *, $p < 0.001$.

Results

The characteristics of 12 patients (5 female and 7 male) were summarized in Table 1. The average age of all patients was 34.7 years old, and the average age of female and male patients was 36.4 and 33.4 years old, respectively, with no significant difference. The alopecia patients received 4-10 sessions of growth-factor treatment for 2-4 months. For female and male alopecia patients, the area of alopecia lesions declined drastically after several sessions of growth factor treatment (Figure 1a&1b). The overall result indicated that hairs regenerated rapidly in all patients and the area of alopecia lesions was reduced by 75.6%. The decrease in alopecia lesions was 66.2% in female patients and 82.3% in male patients, respectively. Compared with female patients who received growth-factor treatment, a significantly greater reduction in alopecia lesions was observed. (Figure 1c). For patients aged 20-29 years old, 71.7% of alopecia area restored hair growth after growth factor treatment. Similar results were observed in the 30-39 years old group (80.8%) and 40 and above groups (72%). The above findings illustrated that growth factor treatment was potent for hair regeneration across all age groups, and the treatment was more effective for male alopecia patients.

Alopecia can be classified into two types, scaring and non-scaring alopecia. Scarring alopecia refers to a type of hair loss condition in which hair follicles are destroyed and replaced by scar tissue, leading to permanent hair loss while non-scarring alopecia refers to hair loss that does not result in permanent damage or scarring to the scalp. Androgenetic alopecia, alopecia areata, telogen effluvium, and traction alopecia are common types of non-scaring alopecia. Androgenetic alopecia is the most common type of hair loss. 8 alopecia patients (3 female patients and 5 male patients) were classified as androgenetic alopecia. Before treatment, the patients displayed a typical androgenetic alopecia pattern. Women with this condition experienced an increase in hair shedding, a widening of the part line, and a reduction in hair density while male patients were characterized by gradual hair thinning and loss on the top of the scalp (Figure 2a). After growth factor treatment, the hair loss was mitigated, hair growth was restored, and the alopecia area declined by 79.2% among all androgenetic alopecia patients (Figure 2b and 3e).

Telogen effluvium is a type of hair loss characterized by excessive shedding of hair from the scalp, which is usually triggered by a physical or emotional stressor such as illness, surgery, childbirth, or psychological stress. The two female alopecia patients who experienced childbirth and insomnia were classified as telogen effluvium. Before treatment, both patients exhibited a diffuse thinning of hair on the scalp (Figure 3a). After receiving growth factor-based therapy, the hair regeneration process occurred with elevated higher hair density and declined alopecia lesions (Figure 3b&3e).

Alopecia areata is an autoimmune disorder in which the body's immune system attacks hair follicles, resulting in hair loss in patches on the scalp or other parts of the body. Alopecia areata was observed

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on the right forehead of the male patients (Figure 3c). Following growth factor treatment, the symptoms were much relieved and numerous hairs regenerated on the alopecia area (Figure 3d&3e). In summary, growth factor treatment was efficient in hair restoration and relieving hair loss for various types of non-scaring alopecia which may provide a promising therapy for alopecia patients.

Table 1 Patients characteristics

Gender	Age	Disease description	Treatment	Outcome
Female	34	Postpartum diffuse alopecia with thinning hair two years ago	6 session growth factor treatment for 3 months	Decreased hair loss with normal hair thickness
Female	25	Seborrheic alopecia for over 3 years	4 session growth factor and deep-care treatment for 2 months	Hair density has returned to normal
Female	28	Low levels of Ferritin induced alopecia	6 session growth factor treatment for 2 months	Facilitated hair growth
Female	55	Atrophy of the hair follicles leads to a reduction in the rate of hair growth	10 session growth factor treatment for 4 months	Elevated hair growth rate
Female	40	Insomnia and cortisol-induced severe hair loss	8 session growth factor treatment for 3 months	Reduced hair loss
Male	33	Hereditary hair loss six years ago	6 session growth factor treatment for 3 months	Elevated hair density
Male	41	Seborrheic alopecia	6 session growth factor and deep-care treatment for 3 months	Improved hair conditions
Male	29	Alopecia areata five years ago	6 session growth factor treatment for 2 months	Relieved hair loss
Male	31	Abnormal oil secretion triggers androgenetic alopecia	6 session growth factor treatment	Restored hair growth
Male	30	Hereditary hair loss	6 session growth factor treatment for 3 months	Increased hair follicle growth rate
Male	40	Long-term use of caffeine-containing irritating shampoo leads to a sensitive scalp, which affects the normal growth of hair follicles	8 session growth factor treatment for 3 months	Facilitated hair growth
Male	30	Genetic factors cause abnormal hair follicle growth cycle	8 session growth factor treatment for 3 months	Restored hair growth

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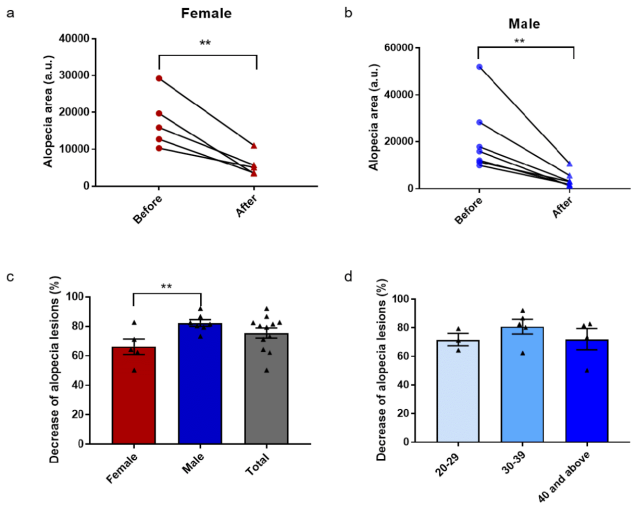

Figure 1 Decrease of alopecia lesions in (a)female (n=5), (b)male (n=7), and (c) all patients (n=12). (d) The decline of alopecia area among all age groups (20-29 years old: n=3; 30-39 years old: n=5; 40 years old and above n=4). Image J was utilized to quantify the alopecia area before and after growth factor treatment. The decrease of alopecia lesions was calculated as (Alopecia area before treatment-Alopecia area after treatment)/ Alopecia area before treatment. Data were presented as the mean ±SEM. Paired t-test was conducted in (a) and (b) and one-way ANOVA tests were performed in (c) and (d). It would be considered significant when the p-value was smaller than 0.05. **, $p < 0.01$.

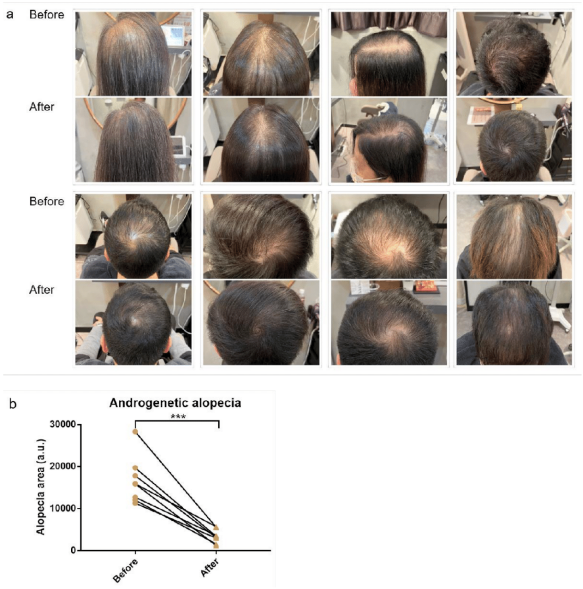

Figure 2 Androgenetic alopecia patients who received growth factor treatment restored hair growth. (a) Images of androgenetic alopecia patients before and after growth factor therapy. (b) Following the growth factor treatment, the alopecia area declined dramatically. Image J was utilized to quantify the alopecia area before and after growth factor treatment. Paired t-test was conducted for data analysis. It would be considered significant when the p-value was smaller than 0.05. ***, $p < 0.01$.

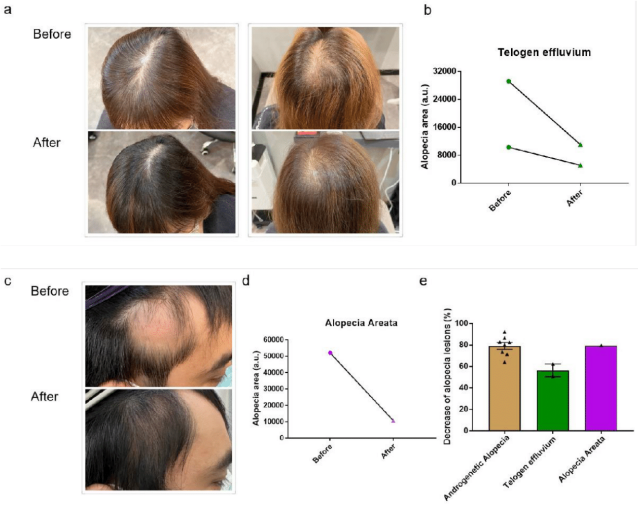

Figure 3 Therapeutic effect of growth factor in telogen effluvium and alopecia areata patients. (a) Images of telogen effluvium patients before and after growth factor therapy. (b) The alopecia area decreased after growth factor treatment. (c) Images of alopecia areata patients before and after growth factor therapy. (d) Reduction in alopecia lesions was observed after growth factor therapy. (e) Summary data on the effect of growth factor treatment among androgenetic alopecia, telogen effluvium, and alopecia areata patients. Image J was utilized to quantify the alopecia area before and after growth factor treatment in (b), (d), and (e). The decrease of alopecia lesions was calculated as (Alopecia area before treatment-Alopecia area after treatment)/ Alopecia area before treatment in (e).

Discussion

Alopecia is a common scalp disorder that disturbs numerous patients. Although hair loss is not a life-threatening disease, it affects the appearance and psychological conditions of the patients or even triggers complications. The worldwide market for alopecia reached $7.6 billion in 2020 and is forecasted to reach $13 billion by 2028.[33] In spite of the tremendous market size, the curative effect of current alopecia treatment is unsatisfactory.

Minoxidil and Finasteride are two conventional FDA-approved medications specifically designed to treat androgenetic alopecia. The main limitations of these drug treatments are their efficacy and potential adverse effects. Previous research has manifested that only 33% of patients with androgenetic alopecia experienced reduced hair breakage following oral administration of Minoxidil,[34] while in our study, a drastic decrease of 79.1% in alopecia lesions was observed among all patients with androgenetic alopecia. Side effects of Finasteride were reported when topically applied on the scalp of alopecia patients, including headaches, sexual dysfunctions, and elevated liver enzyme levels.[35,36] Some androgenetic alopecia patients receiving Minoxidil treatment have reported hypertrichosis, swelling, and dizziness.[34,37] However, in this study, the hbFGF used had the same sequence as the naturally occurring human hbFGF. No adverse effects were reported throughout the entire hbFGF treatment period.

Microneedle therapy is commonly used for treating various skin conditions, including alopecia. It involves the use of a handheld device with tiny needles to create channels on the scalp, stimulating collagen production and enhancing the absorption of topical medications.[38] Previous studies have demonstrated the combined effect of microneedle therapy with other hair loss treatments. For example, patients with androgenetic alopecia who received a combination of low-level laser and microneedle therapy showed increased hair density.[39] Additionally, male patients with androgenetic alopecia experienced significant improvement in hair regeneration when minoxidil was combined with microneedle-mediated FGF treatment.[40] Our study further illustrated the effectiveness of microneedle-assisted growth factor therapy in stimulating hair regeneration in patients with

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various types of non-scaring alopecia. Notably, the curative effect was more pronounced in male patients compared to female patients. This difference may be attributed to distinct molecular mechanisms contributing to alopecia in males and females.

The therapeutic effect of microneedle-assisted hbFGF treatment has been well demonstrated in certain non-scarring alopecia; however, some limitations need to be addressed in future studies. Firstly, while the curative effect of growth factor therapy was illustrated in non-scarring alopecia patients, its effect on scarring patients remains elusive. Previous studies have shown that bFGF facilitated hair follicle growth in mice,[41] suggesting that hbFGF may potentially alleviate scarring alopecia. Future studies will focus on examining the clinical outcomes of various types of scarring alopecia patients.

Secondly, the sample size of the current study is limited while conducting large-scale studies provides more robust and statistically significant evidence regarding the effectiveness of the treatment in managing hair loss. Moreover, large-scale studies help identify any variations in treatment response among different subgroups, such as age, gender, or specific types of alopecia. In this study, only two telogen effluvium and one alopecia areata patients receiving hbFGF therapy are evaluated, and recruiting more telogen effluvium and alopecia areata patients will better confirm current findings.

Finally, in future research, a long-term observation will be conducted to evaluate the effects of discontinuing hbFGF treatment. While FDA-approved medications can lead to regrowth, the effects may not be permanent. Hair loss may resume once the treatment is stopped. Therefore, it is necessary to assess the long-term therapeutic effects after terminating microneedle-assisted hbFGF treatment.

Conclusion

In summary, this study demonstrated that microneedle-assisted hbFGF therapy effectively stimulated hair regeneration in patients with non-scarring alopecia. The treatment reduced the affected area in both female and male patients, with males experiencing a better curative effect. Furthermore, hbFGF therapy proved to be suitable for patients across a wide age range and various types of alopecia. Collectively, these findings suggest the potential of a novel microneedle-assisted hbFGF therapy in addressing hair loss.

Acknowledgments

We would like to express our heartfelt appreciation to Kwok Ming Hei from Rare One Limited for the contributions to our research on hair regeneration treatment procedures for alopecia patients. The support and participation have been crucial to the successful completion of this study.

Conflicts of interest

Ying Xin and Kam-Chau Wu have been employed at DreamTec Research Limited. Kwok Ming Hei has been employed at Rare One Limited. Nelson Cheuk-Yin Lai and Keith Wai-Yeung Kwong have been employed at DreamTec Research Limited, Oristry BioTech (HK) Limited, and Theratide BioTech (HK) Limited. Dominic Man-Kit Lam has been employed at DrD Novel Vaccines Ltd.

References

1. Lei M, Chuong CM. STEM CELLS. Aging, alopecia, and stem cells. *Science*. 2016;351(6273):559–560.
2. Chen CL, Huang WY, Wang EHC, et al. Functional complexity of hair follicle stem cell niche and therapeutic targeting of niche dysfunction for hair regeneration. *J Biomed Sci*. 2020;27(1):43.
3. Kanti V, Röwert–Huber J, Vogt A, et al. Cicatricial alopecia. JDDG: *Journal der Deutschen Dermatologischen Gesellschaft*. 2018;6(4):435–461.
4. Rudnicka L, Kaczorowska A. Treatment of diseases associated with non–cicatricial alopecia. *Dermatology Review/Przegląd Dermatologiczny*. 2021;108(6):504–516.
5. Gordon KA, Tosti A, Alopecia: evaluation and treatment. *Clin Cosmet Investig Dermatol*. 2011;101–106.
6. Xu F, Sheng YY, Mu ZL, et al. Prevalence and types of androgenetic alopecia in Shanghai, China: a community□based study, Br J Dermatol. 2009;160(3):629–632.
7. Asfour L, De Brito M, Al–Janabi A, et al. Global Guidelines in dermatology mapping project (GUIDEMAP): a systematic review of alopecia areata clinical practice guidelines, *Clin Exp Dermatol*. 2023;48(2):100–107.
8. Lolli F, Pallotti F, Rossi A, et al. Androgenetic alopecia: a review. *Endocrine*. 2017;57(1):9–17.
9. Starace M, Orlando G, Alessandrini A, et al. Female androgenetic alopecia: an update on diagnosis and management. *Am J Clin Dermatol*. 2020;21(1):69–84.
10. Chumlea WC, Rhodes T, Girman CJ, et al. Family history and risk of hair loss. *Dermatology*. 2004;209(1):33–39.
11. Nestor MS, Ablon G, Gade A, et al. Treatment options for androgenetic alopecia: Efficacy, side effects, compliance, financial considerations, and ethics, *J Cosmet Dermatol*. 2021;20(12):3759–3781.
12. Trüeb RM. Molecular mechanisms of androgenetic alopecia, Exp Gerontol. 2002;37(8):981–990.
13. Pratt CH, King LE, Messenger AG, et al. Alopecia areata. *Nat Rev Dis Primers*. 2017;3(1):17011.
14. Zhou C, Li X, Wang C, et al. Alopecia areata: an update on etiopathogenesis, diagnosis, and management. *Clin Rev Allergy Immunol*. 2021;61(3):403–423.
15. Malkud S. Telogen effluvium: A review. *J Clin Diagn Res*. 2015;9(9):WE01–WE03.
16. Asghar F, Shamim N, Farooque U, et al. Telogen effluvium: a review of the literature. *Cureus*. 2020;12(5).
17. Mustafa AI, Khashaba RA, Fawzy E, et al. Cross talk between oxidative stress and inflammation in alopecia areata. *J Cosmet Dermatol*. 2021;20(7):2305–2310.
18. Tellez–Segura R. Involvement of mechanical stress in androgenetic alopecia. *Int J Trichology*. 2015;7(3):95–99.
19. Saceda–Corralo D, Domínguez–Santas M, Vañó–Galván S, et al. What's New in Therapy for Male Androgenetic Alopecia?, *Am J Clin Dermatol*. 2023;24(1):15–24.
20. Fung S, Shirley M. Baricitinib: a review in severe alopecia areata. *Am J Clin Dermatol*. 2023;24(4):661–668.
21. Vañó–Galván S, Villodres E, Pigem R, et al. Hair transplant in frontal fibrosing alopecia: A multicenter review of 51 patients. *J Am Acad Dermatol*. 2019;81(3):865–866.
22. Etulain J, Mena HA, Meiss RP, et al. An optimised protocol for platelet–rich plasma preparation to improve its angiogenic and regenerative properties. *Sci Rep*. 2018;8(1):1513.
23. Klifto KM, Othman S, Kovach SJ. Minoxidil, platelet–rich plasma (PRP), or combined minoxidil and PRP for androgenetic alopecia in men: a cost–effectiveness Markov decision analysis of prospective studies. *Cureus*. 2021;13(12):e20839.

24. Park J, Jun EK, So D, et al. Overexpression of Nanog in amniotic fluid–derived mesenchymal stem cells accelerates dermal papilla cell activity and promotes hair follicle regeneration. *Exp Mol Med*. 2019;51(7):1–15.
25. Tak YJ, Lee SY, Cho AR, et al. A randomized, double–blind, vehicle–controlled clinical study of hair regeneration using adipose–derived stem cell constituent extract in androgenetic alopecia. *Stem Cells Transl Med*. 2020;9(8):839–849.
26. Neira JA, Tainturier D, Peña MA, et al. Effect of the association of IGF–I, IGF–II, bFGF, TGF–β1, GM–CSF, and LIF on the development of bovine embryos produced in vitro. *Theriogenology*. 2010;73(5):595–604.
27. Nguyen TH, Paluck SJ, McGahran AJ, et al. Poly(vinyl sulfonate) Facilitates bFGF–Induced Cell Proliferation. *Biomacromolecules*. 2015;16(9):2684–2692.
28. Arunkumar P, Dougherty JA, Weist J, et al. Sustained release of Basic Fibroblast Growth Factor (bFGF) Encapsulated Polycaprolactone (PCL) microspheres promote angiogenesis *In Vivo*. *Nanomaterials*. 2019;20(9):1037.
29. Yang F, Liu Y, Tu J, et al. Activated astrocytes enhance the dopaminergic differentiation of stem cells and promote brain repair through bFGF. *Nature Communications*. 2014;5(1):5627.
30. Lin WH, Xiang LJ, Shi HX, et al. Fibroblast growth factors stimulate hair growth through β–catenin and Shh expression in C57BL/6 mice. *Biomed Res Int*. 2015;2015:730139.
31. Xiao S, Den Y, Mo X, et al. Promotion of hair growth by conditioned medium from extracellular matrix/stromal vascular fraction gel in C57BL/6 mice. *Stem Cells Int*. 2020;2020:9054514.
32. Liu C, Zhao H, Zhang Y, et al. Clinical observation of basic fibroblast growth factor (bFGF) combined with minoxidil in the treatment of male androgenetic alopecia. *J Cosmet Dermatol*. 2022;21(9):4053–4059.
33. Castro AR, Portinha C, Logarinho E. The booming business of hair loss. *Trends Biotechnol*. 2023;41(6):731–735.
34. Beach RA. Case series of oral minoxidil for androgenetic and traction alopecia: Tolerability & the five C's of oral therapy. *Dermatol Ther*. 2018;31(6):e12707.
35. Caserini M, Radicioni M, Leuratti C, et al. Effects of a novel finasteride 0.25% topical solution on scalp and serum dihydrotestosterone in healthy men with androgenetic alopecia. *Int J Clin Pharmacol Ther*. 2016;54(1):19–27.
36. Giatti S, Diviccaro S, Panzica G, et al. Post–finasteride syndrome and post–SSRI sexual dysfunction: two sides of the same coin?. *Endocrine*. 2018;61(2)180–193.
37. Tanaka Y, Aso T, Ono J, et al. Androgenetic alopecia treatment in Asian men. *J Clin Aesthet Dermatol*. 2018;11(7):32–35.
38. Jia L, Xiong J, Guo R, et al. A comprehensive review of microneedling as a potential treatment option for androgenetic alopecia. *Aesthetic Plast Surg*. 2022;46(6):2979–2994.
39. Gentile P, Garcovich S, Lee SI, et al. Regenerative biotechnologies in plastic surgery: A multicentric, retrospective, case–series study on the use of micro–needling with low–level light/laser therapy as a hair growth boost in patients affected by androgenetic alopecia. *Appl Sci*. 2022;12(1):217.
40. Yu CQ, Zhang H, Guo ME, et al. Combination therapy with topical minoxidil and nano–microneedle–assisted fibroblast growth factor for male androgenetic alopecia: a randomized controlled trial in Chinese patients. *Chin Med J (Engl.)*. 2020;134(7):851–853.
41. Ozeki M, Tabata Y. Promoted growth of murine hair follicles through controlled release of basic fibroblast growth factor. *Tissue Eng*. 2002;8(3):359–366.

Citation: Xin Y, Hei M, Cheuk-Yin NL, et al. Hair regeneration using microneedle-assisted human basic fibroblast growth factor. *J Anal Pharm Res.* 2023;12(2):95–100. DOI: 10.15406/japlr.2023.12.00429