Evaluating the Efficacy of Different Platelet-Rich Plasma Regimens for Management of Androgenetic Alopecia: A Single-Center, Blinded, Randomized Clinical Trial

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BACKGROUND Studies suggest platelet-rich plasma (PRP) may mitigate androgenetic alopecia (AGA), but each varies in the frequency of and interval between treatments.

OBJECTIVE To compare the efficacy, satisfaction, tolerability, and safety of 2 initial PRP injection protocols over 6 months.

METHODS Prospective, randomized, single-blinded trial among 40 patients with moderate AGA. Participants received subdermal PRP injections according to 1 of 2 treatment protocols: 3 monthly sessions with booster 3 months later (Group 1) or 2 sessions every 3 months (Group 2). Folliscope hair count and shaft caliber, global photography, and patient satisfaction questionnaires were obtained at baseline, 3 months, and 6 months.

RESULTS At 6 months, both groups demonstrated statistically significant increases in hair count ($p < .001$). These improvements occurred more rapidly and more profoundly for Group 1 (mean percent change: Group 1, 29.6 ± 13.6 vs Group 2, 7.2 ± 10.4; $p < .001$). Shaft caliber also increased significantly with no difference between groups. Treatments produced high satisfaction (82% "satisfied" or "highly satisfied") and were safe and well tolerated (mean pain score 2.1).

CONCLUSION Subdermal PRP injections are an efficacious and tolerable therapy among men and women with AGA. The benefits may be greater if first administered monthly. Clinicians should consider these findings when designing treatment plans.

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Androgenetic alopecia (AGA) is the most common form of hair loss, affecting up to 50% of men and 21 million women in the United States alone.[1] Irrespective of sex, balding is known to influence social interactions and cause substantial emotional distress.[2] Topical minoxidil and oral finasteride, a selective 5 alpha-reductase inhibitor, are currently the only Food and Drug Administration (FDA)-approved therapies for AGA in men, and oral antiandrogens are used off-label in women.[3–5] Both demonstrate the greatest reduction in hair loss and smaller percentage of hair growth after 4 plus months of daily use.[6–8] However, response to minoxidil varies from 20% to 40% with most of the patients on monotherapy progressing despite ongoing therapy, and these beneficial effects do not last on discontinuation.[9] Clinical trial data and off-label experience suggest that dutasteride, a nonselective 5 alpha-reductase inhibitor FDA-approved for benign prostatic hypertrophy, may be as or more efficacious than conventional therapies.[10] Hair restoration surgery is a more permanent option,[11,12] yet for many, the cost is prohibitive. According to *The Washington Post*, in total, nearly 3.5 billion dollars is spent annually on hair loss therapies with varying degrees of success;[13] so, there is a need for effective, scientifically sound alternatives.

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The injection of platelet-rich plasma (PRP) as an autologous therapy has garnered increasing interest across a wide variety of medical specialties. Originally indicated to enhance connective tissue regeneration in orthopedics, PRP has been applied more recently to dermatologic conditions because of its ability to stimulate fibroblast proliferation, induce collagen and elastin production, and improve the quantity and quality of the extracellular matrix.[14,15] Studies in mice suggest that these matrix changes result from increased pro-stimulatory cytokines, such as vascular endothelial growth factor (VEGF), platelet-derived growth factor (PDGF), and transforming growth factor beta (TGFβ).[16] Considering the fact that these signaling cascades are also critical in regulating the hair growth cycle, PRP has emerged as a potential management of AGA.[17,18] Several recently published articles demonstrate efficacy in treating patterned baldness,[19–21] but each varies in the treatment protocol, and there are no evidence-based data guiding frequency of and/or interval between injection sessions. The authors conducted a prospective, randomized, single-blinded pilot trial to investigate the most beneficial number and timing ("protocol") of PRP treatments among men and women with AGA. The objectives of this study were to compare the efficacy, satisfaction, tolerability, and safety of 2 different initial PRP protocols over 6 months and determine which was superior when starting therapy.

Materials and Methods

Study Design

After receiving institutional review board/ethics committee approval, the authors enrolled 40 male and female participants from a private practice in Los Angeles, CA. Twenty were randomly assigned to 1 of 2 different treatment groups: 3 monthly sessions with a booster 3 months later (total 4 treatments; Group 1) versus 2 sessions every 3 months (total 2 treatments; Group 2).

Participants

Forty healthy men and women aged 18 to 60 years with AGA, disease stages Norwood-Hamilton II–V and Ludwig I2–II1, respectively, were recruited between November 2016 and January 2017.

Exclusion criteria included: diagnosis of non-AGA hair loss; active skin disease, infection, cuts, or abrasions on the scalp; history of surgical hair restoration; current or recent malignancy, excluding non-scalp nonmelanoma or melanoma skin cancers, as well as current or recent chemotherapy or radiation treatments; history of thyroid dysfunction, autoimmune disorder that might interfere or increase the risks associated with the treatment, or blood-borne infection (i.e., human immunodeficiency virus [HIV], hepatitis B virus, and hepatitis C virus); tendency to develop keloids; anticoagulant therapy, except for aspirin, nonsteroidal anti-inflammatory drugs, or vitamin E if discontinued 7 to 14 days before each session; hematologic or coagulation disorder (i.e., platelet dysfunction syndrome, thrombocytopenia <150,000 platelets/μL, hypofibrinogenemia); or current or anticipated pregnancy and/or breastfeeding. Subjects whose hair had been clinically stable on FDA-approved AGA therapies—topical minoxidil and/or oral finasteride—for 12 months were allowed to participate without changing their regimens because in practice, PRP is often used in conjunction with other available therapies, as these agents have different mechanisms of action so may have synergistic effects, and because discontinuing such medications could result in telogen effluvium several months into the study, thereby altering results. Use of other products, devices, or medications intended to promote hair growth was prohibited. There was a 90-day washout period for antiandrogen therapies.

To ensure adherence to the above, participants underwent thorough screening history and physical examination. Hair pull test detected abnormal shedding, as seen in telogen effluvium. Laboratory testing included factors that may affect and/or explain hair loss (thyroid stimulating hormone, ferritin level); serologies for important blood-borne infections; risk factors for increased bleeding (platelet count); and pregnancy testing.

Interventions/Preparation of Intervention

The authors randomized eligible participants in a 1:1 ratio to 2 treatment protocols: Group 1 received 4 total injections, the first 3 at monthly intervals and the last 3

TABLE 1. Baseline Characteristics of Patients Treated With Platelet-Rich Plasma

Demographics	*All (n = 39)*	*Group 1 (n = 20)*	*Group 2 (n = 19)*
Sex, n (%)			
Male	29 (74.4)	17 (85.0)	12 (63.2)
Female	10 (25.6)	3 (15.0)	7 (36.8)
Mean age (yr)	43.75	40.1	46.85
Race/ethnicity, n (%)			
Non-Hispanic White/Caucasian	29 (74.4)	16 (80.0)	13 (68.4)
Hispanic	3 (7.6)	1 (5.0)	2 (10.5)
Black	1 (2.6)	0	1 (5.3)
Asian/Pacific Islander	1 (2.6)	1 (5.0)	0
Middle Eastern/Persian	4 (10.2)	2 (10.0)	2 (10.5)
Other	1 (2.6)	0	1 (5.3)
Mean duration of hair loss (yr)	6.45	6.15	6.75
Less than 6 years, n (%)	17 (43.6)	9 (45.0)	8 (42.1)
More than 6 years, n (%)	22 (56.4)	11 (55.0)	11 (57.9)
Hair loss therapy, n (%)			
Previous finasteride	15 (38.5)	10 (50.0)	5 (26.2)
Current finasteride	8 (20.5)	4 (20.0)	4 (21.1)
Previous minoxidil	17 (43.6)	9 (45.0)	8 (42.1)
Current minoxidil	10 (25.6)	6 (30.0)	4 (21.1)
Other	11 (28.2)	3 (15.0)	8 (42.1)
Pattern, n (%)			
Temporal	9 (23.0)	5 (25.0)	4 (21.1)
Vertex	7 (18.0)	4 (20.0)	3 (15.8)
Temporal and vertex	13 (33.3)	8 (40.0)	5 (26.2)
Central part	7 (18.0)	3 (15.0)	4 (21.1)
Central part and temples	3 (7.7)	0	3 (15.8)

months later, whereas Group 2 received 2 total injections, one at baseline and one at 3 months.

Platelet-rich plasma preparation involved EclipsePRP kits (Eclipse Aesthetics LLC, Dallas, TX), collecting 22 mL of peripheral blood, which was centrifuged at 3,500 revolutions per minute for 10 minutes. This process separates blood products according to their specific densities. The proprietary gel plug removes 99.9% of erythrocytes and 92% of leukocytes.[22,23] Approximately 50% of resulting supernatant is removed (platelet-poor plasma), and the tube is gently agitated to resuspend all platelets in the remaining smaller volume of plasma. The final yield generally ranges from 4 to 6 mL of PRP at roughly 4 to 6 times the platelet concentration of whole blood. Platelet-rich plasma was injected in 0.2 to 0.5 mL aliquots subdermally using a 32-gauge, half-inch needle every 2 to 3 cm at balding areas. At each appointment, participants had the option to use topical lidocaine 23%/tetracaine 7% ointment for anesthesia at the region of injections.

Assessment Criteria and Post-treatment Data Collection

During screening, the authors obtained baseline magnified (Folliscope 2.8; Anagen Corp., Seoul, Korea) and global (Hair Metrix^{SM}/Canon Rebel T6i; Canfield Scientific Inc., Parsippany, NJ) photographs from which mean hair count (hairs/cm^2), shaft caliber (microns, μm), and Norwood–Hamilton or Ludwig scale were determined. A single representative point at the leading edge of an actively balding region was selected and used for all folliscope images throughout the duration of the trial. Precise measurements obtained between this point and the glabella and bilateral canthi (triangulation, specifically: lines measured obliquely from the lateral canthi to the evaluation point and one line measured from the midway

TABLE 2. Hair Count Parameters for 2 Treatment Protocols at Baseline, 3 Months, and 6 Months

	Count (hairs/cm²), Mean ± SD					*Change Between Groups, Mean ± SD*	
Group	*Baseline*	*3 mo*	p	*6 mo*	p	*Absolute (hairs/cm²)*	*Percent (%)*
1	160.4 ± 36.9	183.5 ± 40.9	<.001	207.1 ± 49.5	<.001	46.7 ± 23.5	29.6 ± 13.6
2	177.6 ± 62.0	181.1 ± 65.1	.23	190.6 ± 66.9	.009	13.1 ± 19.3	7.2 ± 10.4
						$p < .001$	$p < .001$

point on a line drawn between the medial canthi to the evaluation point) were recorded in the study documents and allowed for consistent, reproducible identification of the imaging site at each evaluation appointment. The authors did not tattoo this location because permanent, invasive markings might discourage enrollment and thus, decrease the study's generalizability. Folliscope and global photography were repeated at 3- and 6-month status after first PRP treatment. All data collection, including determination of hair counts and caliber as well as overall severity ratings, were performed in batches without consulting previous images or numbers to ensure better blinding and more objective results.

Subjects also completed satisfaction and outcome questionnaires at 3 and 6 months, rating satisfaction on a 4-point scale (3 = highly satisfied, 2 = satisfied, 1 = dissatisfied, and 0 = highly dissatisfied). Safety and tolerability data were collected during each appointment. After all treatments, participants assigned a pain score on the verified 0-point visual analogue scale.

Statistical Assessment

Statistical analyses were performed in Python using libraries scipy (version 0.19.0), statsmodels (v0.8.0), and seaborn (v0.7.1). To assess changes in mean hair count and caliber over time, the authors applied Student *t*-test or Wilcoxon signed-rank test, if data were not normally distributed. The authors evaluated the relationship between mean percent change in hair count and duration, age, and sex using linear and logistic regression modeling. Other analyses included descriptive statistics (percent, absolute, and relative frequencies) for categorical and ordinal variables such as satisfaction and safety data as well as analysis of variance (mixed model for repeated measures) testing for quality of life metrics.

Results

Forty patients, 20 per group, enrolled in this study, including 30 men and 10 women, of which 39 completed the protocol (20 in Group 1 and 19 in Group 2). One man withdrew after his first treatment for unrelated personal reasons. Table 1 summarizes baseline characteristics of all subjects. Mean age at enrollment was 43.75 years and similar between groups ($p = .69$).

Hair Counts

Mean baseline hair counts were similar between the 2 groups ($p = .37$). Only Group 1 demonstrated statistically significant increases at 3 months (Group 1, $p < .001$; Group 2, $p = .23$). By 6 months, both achieved statistical significance relative to baseline ($p < .001$). However, when comparing absolute and percent change between groups, the Group 1 protocol yielded superior results and was statistically

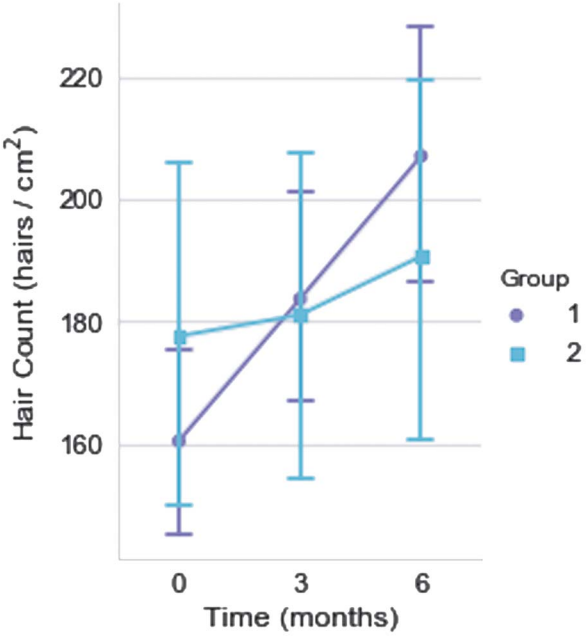

Figure 1. Change in hair counts over 6 months.

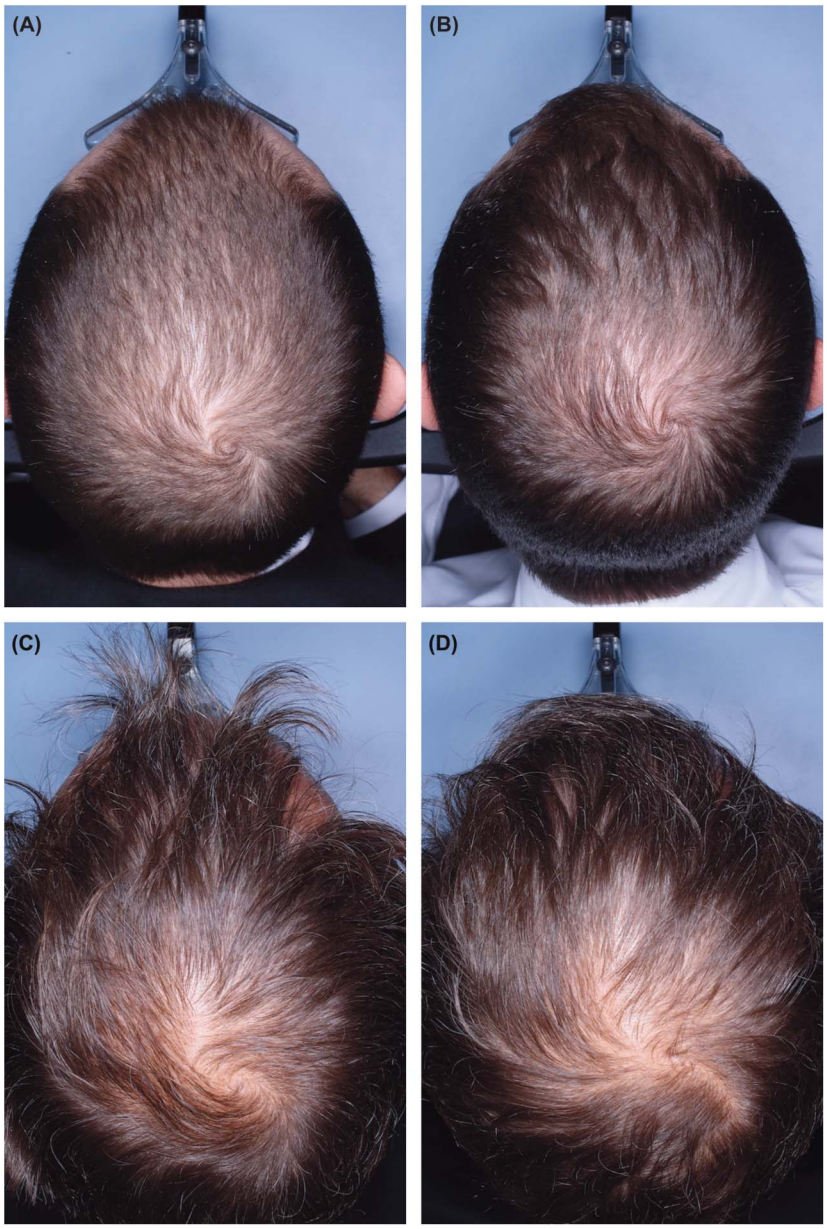

Figure 2. Global photography of representative patients from Group 1 at (A) baseline and (B) 6 months as well as from Group 2 at (C) baseline and (D) 6 months.

significantly better than that of Group 2 (mean percent change: Group 1, 29.6 ± 13.6 vs Group 2, 7.2 ± 10.4; $p < .001$; Table 2, Figures 1 and 2).

Using linear regression to evaluate important variables potentially associated with response to PRP, the authors found no difference in the increases seen among men and women ($p = .78$). Longer duration of AGA correlated with lower mean percent change in hair count, particularly for Group 1 (Figure 3). There was a trend toward smaller change with increasing age, although the confidence intervals were wide. "Being in Group 1" was the only statistically significant variable predicting a mean percent change >20% in a multivariable logistic regression controlling for age, AGA duration, and sex ($p < .001$).

Hair Shaft Caliber

Statistically significantly increased mean hair shaft caliber was evident at 3 and 6 months irrespective of group ($p < .001$). At the study end point, there was no

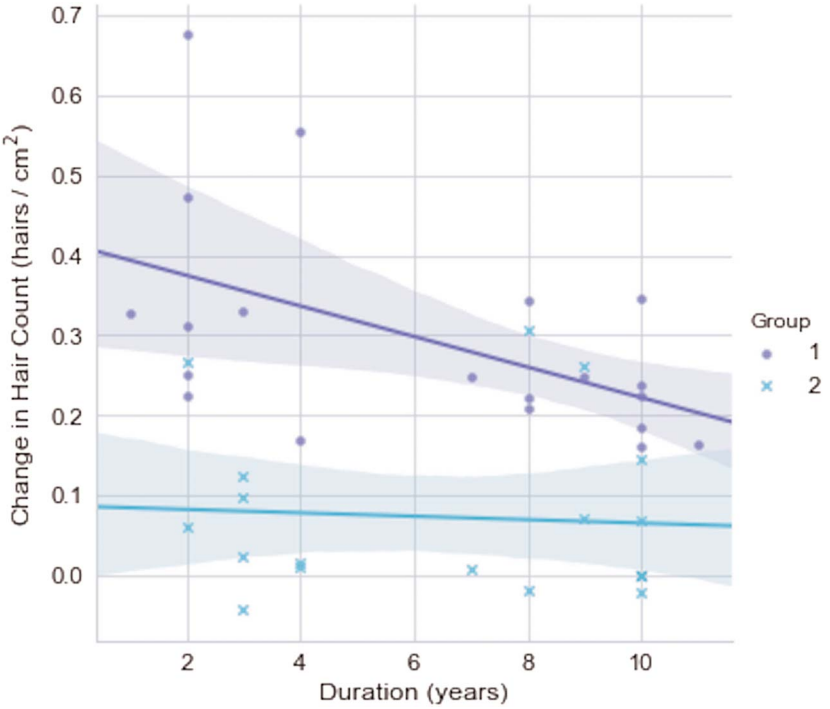

Figure 3. Correlation between change in hair count and duration of androgenetic alopecia.

significant difference in absolute or percent change between the treatment protocols (absolute change, $p = .59$ and percent change, $p = .36$, respectively; Table 3 and Figure 4).

Satisfaction

Mean satisfaction score across the entire study period was 2.3 (Group 1, 2.5; Group 2, 2.1). At 6 months, 82% of participants reported being "satisfied" or "highly satisfied" (Table 4). The majority would continue therapy or recommend it to others. Larger changes in mean hair count did correlate with higher satisfaction ratings among both groups. This trend appeared during the first 3 months of treatment, and then persisted ($p = .02$). Of note, those in Group 1 were more likely to report the highest scores.

Tolerability and Safety

Although just under a third of subjects reported associated pain or discomfort, 74.4% rated the procedure as "tolerable" with a mean pain score of 2.1 (Table 5). All adverse events were mild as outlined in Table 6.

Discussion

Androgenetic alopecia affects a substantial portion of the population; yet, there are few scientifically proven therapies. This prospective, randomized, controlled trial is the first to directly compare different initial PRP treatment protocols in the hopes of optimizing outcomes. The authors demonstrated that PRP positively affects hair parameters and mitigates changes associated with patterned hair loss, regardless of the 2

TABLE 3. Hair Shaft Caliber Parameters for 2 Treatment Protocols at Baseline, 3 Months, and 6 Months

	Caliber (μm), Mean ± SD					*Change Between Groups, Mean ± SD*	
Group	*Baseline*	*3 mo*	p	*6 mo*	p	*Absolute (μm)*	*Percent (%)*
1	54.5 ± 9.3	64.7 ± 6.8	<.001	70.5 ± 8.5	<.001	16.0 ± 7.3	31.1 ± 17.2
2	57.5 ± 10.7	65.2 ± 10.4	<.001	72.3 ± 10.6	<.001	14.8 ± 5.9	26.7 ± 11.8
						$p = .59$	$p = .36$

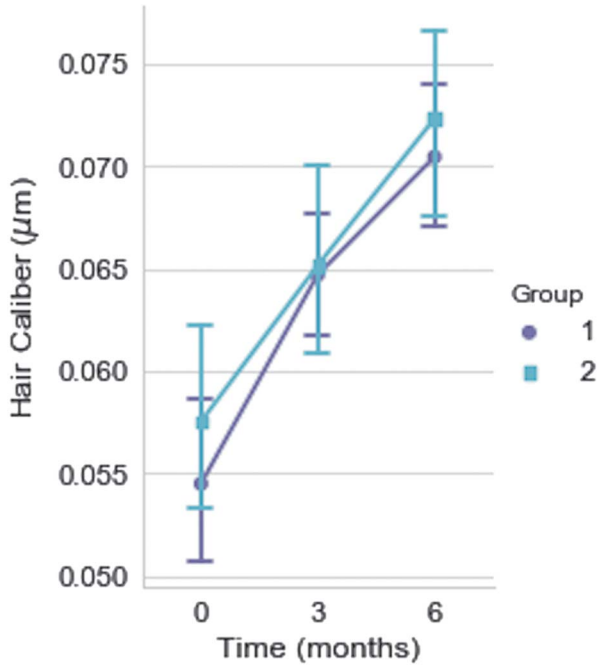

Figure 4. Change in shaft caliber over 6 months.

injection regimens. However, when initiating therapy, monthly treatments transitioning to every 3 months provide greater increases in hair regrowth than sessions every 3 months only. The 2 protocols thickened hair shafts similarly.

Although both groups showed statistically significant improvements in hair count at 6 months, changes were evident earlier and more clinically relevant among Group 1 who had a mean 30% increase compared with Group 2 (7% increase). The human eye cannot reliably detect a 7% overall difference in the number of hairs, a fact that may at least partially explain why subjects in Group 1 were more likely to report the highest levels of satisfaction. Multivariate analysis controlled for sex, age, and disease duration further confirmed that "being in Group 1" predicted a greater response.

Premature transition from anagen to telogen and follicular miniaturization are hallmarks of androgenetic hair loss resulting from altered expression of growth factors. Hair cycling and growth depends on intricate signaling between mesenchymal-derived dermal papilla cells and multipotent stem cells in the follicular bulge region. Studies indicate that increasing Wnt/beta-catenin, sonic hedgehog (Shh), and signal transducer and activator of transcription 3 (STAT3) pathways while downregulating bone morphogenetic protein signaling is critical for transition of hair follicles from telogen to anagen.[24–26] Circulating androgens, particularly dihydrotestosterone, bind to androgen receptors in the dermal papilla and suppress pro-growth Wnt, Shh, and STAT3 signaling while activating inhibitory cascades.[25] The growth factors found in PRP, including PDGF, TGFβ, epidermal

TABLE 4. Patient-Reported Satisfaction

Metric, n *(%)*	*All* (n = *39)*	*Group 1* (n = *20)*	*Group 2* (n = *19)*
Satisfaction			
Highly satisfied	16 (41.0)	11 (55.0)	5 (26.3)
Satisfied	16 (41.0)	6 (30.0)	10 (52.6)
Dissatisfied	5 (12.8)	2 (10.0)	3 (15.8)
Highly dissatisfied	2 (5.2)	1 (5.0)	1 (5.3)
Results			
Y	28 (71.8)	15 (75.0)	13 (68.4)
N	6 (15.4)	2 (10.0)	4 (21.1)
Unsure/maybe	5 (12.8)	3 (15.0)	2 (10.5)
Recommend to others			
Y	23 (59.0)	14 (70.0)	9 (47.4)
N	1 (2.6)	0	1 (5.3)
Maybe	15 (38.4)	6 (30.0)	9 (47.4)
Motivated to continue			
Y	28 (71.8)	17 (85.0)	11 (57.9)
N	1 (2.6)	0	1 (5.3)
Maybe	10 (25.6)	3 (15.0)	7 (36.8)

TABLE 5. Patient-Reported Tolerability Over 119 Treatment Sessions (n = 39)

Tolerability	
Verbal classification, n (%)	
Tolerable	29 (74.4)
Moderately comfortable	10 (25.6)
Clearly unpleasant	0
Score, 0–10 (mean, range)	2.1 (0.5–7)

growth factor (EGF), insulin-like growth factor-1 (IGF1), and VEGF, reinstate the necessary signaling pathways and gene transcription that result in cell survival, proliferation, and differentiation. They stimulate telogen-to-anagen transition, prolong anagen, and promote neoangiogenesis.[27] Thicker shafts may also occur due to neovascularization providing better nutrient supply to existing follicular cells,[28–31] decreases in perifollicular microinflammation,[32] and upregulation of antiapoptotic proteins in dermal papilla cells.[18,33] The authors' discrepant findings in hair count versus caliber suggest that it may require a lower concentration of PRP-contained growth factors to thicken miniaturized hairs than to transition into anagen phase and promote new growth. Folliscope photographs (Figure 5) show a progressively pinker-colored scalp and support the role of enhanced vascularization in the mechanisms driving PRP-mediated hair restoration.

Although the authors' study was not intended primarily to determine clinical parameters predicting response to PRP therapy, the authors did note some important trends that warrant further investigation. Improvement in hair counts depended on duration of AGA. This relationship supports the authors' clinical experience: those noting hair loss for less than 5 to 6 years tend to respond more rapidly and profoundly.

TABLE 6. Treatment-Related Adverse Events Occurring in >1% of Participants (n = 39)

Adverse Events	n *(%)*
Pain/discomfort	12 (30.7)
Headache	5 (12.8)
Itching	8 (15.4)
Other	0

Change in mean hair count by age was less straightforward, partially because age and duration of AGA are often, but not always, collinear. There were insufficient numbers to examine changes by race/ethnicity, current or past FDA-approved therapy, or pattern of alopecia.

The authors' results are in line with those from previous studies, reporting increases in hair density and/or number of hairs among patients treated with PRP.[19,21,31,33,34] This body of literature is growing quickly, but most studies are open-label or unblinded using different treatment protocols that are difficult to evaluate head-to-head. The authors sought to compare 2 previously reported protocols in a rigorous prospective, randomized, controlled design using several practical outcome metrics.

Another strength of this study was the novel, subdermal injection technique that has been used clinically and described by Rapaport in a 2017 commentary piece but never tested rigorously to date. This method allows for fewer, more widely spaced injection points than the traditional nappage procedure (0.1 mL injected intradermally on a 1-cm grid) because PRP can diffuse further once in the deeper, subgaleal space. Anecdotally, several patients commented on increased eyebrow growth, presumably from diffusion within this potential space, which is continuous with the frontalis down to the brow. Subdermal injection also was safe and well tolerated, with a mean pain score of approximately 2 on a 0 to 10 scale, although more than 80% declined topical anesthetic. Of the 199 treatment sessions performed for this trial, only during 4 did subjects rate a discomfort score of 5 or higher (3.4%). Half of these occurred in a single patient whose pain dropped to 3 after agreeing to use the optional anesthetic.

In addition, the authors' study included both men and women who responded similarly. This finding is important because there are few data on PRP efficacy among women. Many early reports investigated treatment of male pattern hair loss only. Two recent randomized controlled trials involving women had conflicting results. Alves and Grimalt[21] reported statistically significant increases in hair count irrespective

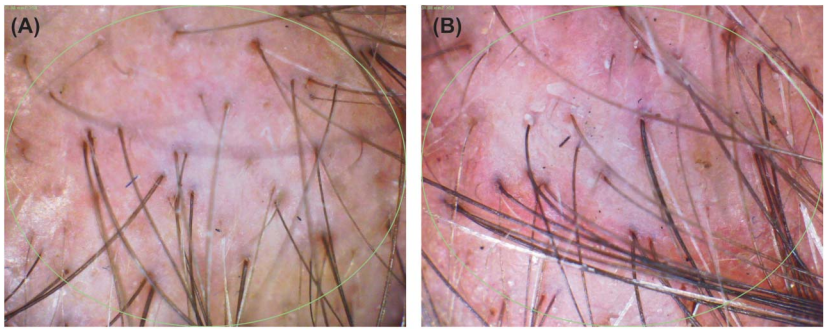

Figure 5. Folliscope images from a patient in Group 1 at (A) baseline and (B) 6 months with 33.1% increase in hair count and 29.4% increase in hair caliber.

of sex, whereas Puig and colleagues[35] found perceived but not actual improvements. The authors suspect that the latter's study protocol (single injection session only) may be more responsible for their statistically null results than the participants' sex. Platelet-rich plasma is an ongoing therapy that requires maintenance when used for a chronic condition such as hair loss.

Despite these strengths, the authors' analysis also had several weaknesses. The sample size was small, albeit one of the largest to date.[33] The 6 month follow-up period was short, given the slow rate of hair growth (1.25 cm/mo), and does not capture long-term effects. The authors continue to follow many of the patients included in this analysis, and several have shown substantial delayed improvements during the 6- to 12-month period; however, those data are outside the scope of this trial. The longest duration study monitored subjects for 2 years with relapse noted at roughly 12 months.[20] Others have seen fall-off after 3 months. The authors' goal was to investigate initial therapy; so, the follow-up period was kept short.

Conclusion

Several studies document the efficacy of PRP in combating AGA, but treatment frequencies and parameters vary widely. The authors' randomized, controlled trial is the first to compare directly the efficacy of 2 starting injection protocols and the first to examine rigorously a well-tolerated, safe subdermal injection technique. Platelet-rich plasma showed biologic activity in both groups. However, those receiving 3 monthly treatments with a 3-month booster had better statistically and clinically significant outcomes with substantial improvements in hair count and shaft thickness. Future studies are necessary to fine-tune preparation methods, determine optimal maintenance schedule(s), and parse out clinical predictors of efficacy.

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