

# Effectiveness of Platelet-Rich Plasma for Androgenetic Alopecia: A Review of the Literature

Jessica Cervantes<sup>a</sup> Marina Perper<sup>a</sup> Lulu L. Wong<sup>a</sup> Ariel E. Eber<sup>a</sup>  
Alexandra C. Villasante Fricke<sup>b</sup> Tongyu Cao Wikramanayake<sup>a</sup>  
Joaquin J. Jimenez<sup>a, c</sup>

<sup>a</sup>Department of Dermatology and Cutaneous Surgery, University of Miami Miller School of Medicine, Miami, FL,

<sup>b</sup>Department of Medicine, Brown University Alpert School of Medicine, Rhode Island Hospital, Providence, RI, and

<sup>c</sup>Department of Biochemistry and Molecular Biology, University of Miami Miller School of Medicine, Miami, FL, USA

## Keywords

Platelet-rich plasma · Androgenetic alopecia · Androgenic alopecia · Hair disorder · Hair follicle · Hair growth · Growth factors

## Abstract

Androgenetic alopecia (AGA) is a hair loss disorder affecting 80% of men and 50% of women throughout their lifetime. Therapies for AGA are limited and there is no cure. There is a high demand for hair restoration. Platelet-rich plasma (PRP), a treatment modality shown to promote wound healing, has also been explored as a treatment for AGA. This literature review was conducted to assess the effectiveness of PRP treatment for AGA. Twelve studies conducted from 2011 to 2017 were evaluated and summarized by study characteristics, mode of preparation, and treatment protocols. A total of 295 subjects were given PRP or control treatment in these studies, and evaluated for terminal hair density, hair quality, anagen/telogen hair ratio, keratinocyte proliferation, blood vessel density, etc. Some studies also provided subject self-assessment reports. Most of the studies reviewed showed effectiveness of PRP in increasing terminal hair density/diameter. Additional investigations are needed to determine the optimal treatment regimen for high efficacy of PRP in AGA.

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## Introduction

### *Androgenetic Alopecia*

Androgenetic alopecia (AGA) is the most common form of alopecia, affecting up to 80% of men and 50% of women during their lifetime [1]. It is a chronic, non-scarring, age-related disorder that is marked by a progressive reduction in the diameter, length, and pigmentation of the hair. This disorder is located primarily on the central scalp with various patterns of loss [1–4]. Pathophysiological features include an alteration in the hair cycle via reduction of the anagen (growth) phase, inflammation, and follicular miniaturization [5]. AGA is determined by genetics and influenced by hormones. The key hormone is dihydrotestosterone (DHT), a metabolite of testosterone, which activates androgen receptors. In men, testosterone is converted to DHT by 5 $\alpha$ -reductase, while dehydroepiandrosterone and other weaker androgens are the precursors of DHT in women. Hair follicles in the scalp vertex and frontotemporal areas have an increased density of androgen receptors; hence, they exhibit a greater response to DHT and experience increased hair loss in AGA [6].

Currently, the Hamilton-Norwood classification is the standard system for classifying different stages of

AGA in males [7]. Hamilton first proposed a detailed system grading severity of hair loss based on frontoparietal and frontal recessions as well as frontal thinning in 1951. The grading system was based on 8 evolutionary types of hair loss and 3 subgroups: types I–III described scalps which were not bald, while types IV–VII classified bald scalps. Later, in 1975, Norwood expanded on Hamilton's system by creating the Hamilton-Norwood classification, which encompassed major patterns of hair loss, but also rarer patterns of male pattern balding. The Hamilton-Norwood system includes 7 types of hair loss, as well as information about a type A variant, based on the notion that thinning begins in the temples and crown/vertex and continues to encompass the entire top of the scalp [8].

The Ludwig classification system is used to describe the severity of AGA in women. Ludwig based the system on 3 grades of hair loss and emphasized the preservation of the frontal fringe despite progressive centrifugal loss over the upper portion of the scalp in females [8, 9]. Nevertheless, he did not account for the accentuation of frontovertical alopecia in his classification – this information was later described by Olsen in her own classification [8, 10].

The current standard of treatment for AGA includes oral finasteride and topical minoxidil solution or foam in males and minoxidil solution or foam in females [11]. Additional therapies including dutasteride, ketoconazole, prostaglandin analogues, and hormonal therapy have also been used in treating AGA [6, 2]. Unfortunately, current therapies are not effective for all subjects with AGA. On the one hand, medication is required for an indefinite period of time, and effectiveness is limited by patient adherence. In addition, they may cause side effects such as hypertrichosis close to the area of minoxidil application, and possible birth defects, decreased libido, and prolonged impotence with finasteride use in males [12]. On the other hand, because of its invasive nature and high price, surgeries such as hair transplantation and scalp reduction are generally reserved for patients who do not achieve success with medical therapy [2]. Surgical options are dependent on each patient's supply of donor hair, and possible scarring in donor sites is a shortcoming although this is becoming less of a problem with follicular unit grafting. Recently, low-level laser therapy has been shown to be effective in promoting hair preservation and regrowth in some AGA patients [13–15]. Due to the varying efficacy and safety profiles of the present treatment modalities for AGA, there remains the need for additional treatments promoting hair regrowth.

### *Platelet-Rich Plasma*

Platelet-rich plasma (PRP), a new biotechnology, is the product of a heightened interest in cell-based therapy and tissue engineering. This therapy is defined as an autologous preparation of plasma with concentrated platelets. PRP contains various growth factors and cytokines that enhance the body's inherent capacity to repair and regenerate [16, 17]. PRP has been traditionally employed in periodontal therapy, maxillofacial surgery, orthopedics, and sports medicine. More recently, it has captured attention in the field of dermatology, particularly for its role in treating acne scars, fat grafting, wound healing, and hair regrowth [18]. Research has demonstrated the beneficial effects of PRP, such as proliferation of adipose precursor cells, wound repair, cellular differentiation, and angiogenesis [16].

Among the cells in a typical blood sample, 93% are red blood cells (RBCs), 6% platelets, and 1% white blood cells. The principle behind PRP treatment is to enrich the platelets through centrifugation, to reverse the RBC-to-platelet ratio to achieve a 94% concentration of platelets and a 5% concentration of RBCs. The high level of growth factors and cytokines in PRP are thought to facilitate tissue rejuvenation and healing. A platelet concentration of approximately 1 million platelets/ $\mu\text{l}$ , which is  $\sim 5$  times the normal concentration of platelets, has demonstrated tissue reparative efficacy [17].

Platelets are most often thought of for their hemostatic functions. However, they also contain a vast reservoir of over 800 proteins which, when secreted, act upon numerous targets including stem cells, fibroblasts, osteoblasts, endothelial, and epithelial cells [19, 20]. Granulation of these factors generally begins within 10 min of platelet activation. Besides platelets and their secreted factors, there are other active components within PRP, importantly fibrinogen and leukocytes. The current thinking is that the therapeutic benefits of PRP come not only from the platelets, but from the combination of constituents and growth factors [19].

### *PRP Preparation*

Current protocols for PRP preparation vary greatly. Although there are several commercially available PRP kits, high cost often precludes their use. Furthermore, even with standardized kits, patient parameters such as hydration status, infection, lipemia, and hematocrit all play a role in the final PRP characteristics [19]. In 2012, a classification system for PRP with 4 distinct "families" of preparations was established by a multidisciplinary consensus conference. The 4 families were determined based

on leukocyte count and fibrin architecture: pure PRP; leukocyte- and platelet-rich plasma (L-PRP); pure platelet-rich fibrin; and leukocyte- and platelet-rich fibrin [21]. A variety of algorithms exist for isolating PRP from whole blood. Despite the variations, all protocols for PRP preparation follow a generic sequence which is summarized below.

#### *PRP Extraction Steps* [20]

- 1 Obtain whole blood by performing venipuncture using vacutainer tubes with an anticoagulant such as citrate dextrose A.
- 2 Centrifuge the tube under a “soft” speed. The sample should separate into a yellow top layer (plasma), a thin middle layer – the buffy coat (platelets and white blood cells) – and a red bottom layer (RBCs).
- 3 For the production of pure PRP, the upper layer and superficial buffy coat should be transferred to an empty sterile tube. For the production of L-PRP, the whole buffy coat layer and a fraction of the RBCs should be transferred.
- 4 Spin a second time at the appropriate force to achieve a “soft pellet” at the bottom.
- 5 Remove the upper 2/3 of fluid from the tube.
- 6 Homogenize the pellet in the remaining 1/3 of fluid. This is ready to use PRP.
- 7 In some cases, an activator is added to the platelet-enriched product.

Dhurat and Sukesh [20] performed a literature review of the various protocols for PRP production evaluating the volume of whole blood used, centrifugal force, and duration for the first and second centrifugation. Each study’s respective platelet count increase was documented. From this study, the authors concluded that 900 g for 5 min for the first centrifugation and 1,000 g for 10 mins for the second centrifugation at 16°C in a refrigerated centrifuge produced the most optimal yield of PRP [22, 23]. Giusti et al. [24] determined that  $1.5 \times 10^6$  platelets/ $\mu\text{L}$  is the optimal concentration of platelets for induction of angiogenesis in endothelial cells. It was suggested that higher concentrations decreased the angiogenic potential of platelets for follicular and perifollicular angiogenesis [24]. Thrombin, collagen, and calcium are common activators added to the enriched PRP, which promote growth factor secretion upon platelet activation [22, 23].

#### *Platelet-Rich Plasma and Hair Regrowth*

PRP contains high concentrations of over 20 growth factors that are actively secreted from the  $\alpha$ -granules of platelets. Among those thought to stimulate hair re-

growth are platelet-derived growth factor, transforming growth factor, vascular endothelial growth factor (VEGF), epidermal growth factor, fibroblast growth factor, connective tissue growth factor, and insulin-like growth factor IGF-1 [25]. These essential proteins regulate cell migration, attachment, proliferation, and differentiation and promote extracellular matrix accumulation [16, 26]. Growth factors in PRP promote hair regrowth by binding to their respective receptors expressed by stem cells of the hair follicle bulge region and associated tissues. Upon ligand binding, stem cells induce the proliferative phase of the hair follicle, producing the anagen follicular unit and facilitating hair regrowth [27, 25]. Further, they activate downstream cascades leading to angiogenesis and stimulation and generation of adnexal structures. Anagen-associated angiogenesis has been linked to the secretion of VEGF by keratinocytes in the outer root sheath and fibroblasts of the dermal papilla. This increased production of VEGF promotes the growth of normal and pathological dermal structures [28]. Activated autologous PRP has also been noted to activate the proliferation of dermal papilla cells by upregulating fibroblast growth factor-7 and  $\beta$ -catenin, in addition to extracellular signal-related kinase and Akt signaling [28].

In this literature review, we evaluated the effectiveness of PRP treatment for AGA in 12 studies, and conclude that PRP was effective in promoting hair growth in most studies.

## **Materials and Methods**

Due to the growing interest in hair restoration, a number of investigations have been conducted to assess the efficacy of PRP as a treatment modality for AGA. Searching through the PubMed/MEDLINE, Clinicaltrials.gov, and Scopus database without a language or publishing-time restriction, we identified 76 articles using the keyword “Platelet rich plasma AND alopecia.” We included clinical trials with male and female patients diagnosed with AGA, also referred to as male or female pattern hair loss. Eight articles were excluded as they evaluated PRP on other hair disorders such as alopecia areata and lichen planus pilaris, 30 articles were excluded as they were review and/or commentaries on the topic, 5 articles were excluded as they were duplicate studies, 10 articles were excluded as they were off-topic, 3 articles were excluded as they assessed PRP in combination with other procedures/treatments, and 8 articles were excluded as the study was incomplete for a total of 38 excluded articles. Twelve original studies were included in this review. These 12 studies were evaluated and summarized by study characteristics (Table 1), treatment protocols (Table 2), mode of preparation of PRP (Table 3), and study results (Table 1). We elaborate on studies conducted in 2017 and 2016, which to the best of our knowledge, have not been collectively discussed in prior reviews. These studies include Anitua et

**Table 1.** Study design and results of included studies

First author [Ref.], year	Study type				Characteristics of enrolled subjects (completed study)	Objective measures	Objective assessment of hair growth	Subjective assessment of hair growth
	randomized	controlled	blinded	half-head				
Anitua [29], 2017	No	No	Yes	No	19 (19) 12 M, aged 27–60, stage III–VI; 2 F, aged 32–60, stage II–frontal	#1–4 Computerized phototrichogram 1. Hair density 2. Hair diameter 3. Terminal/vellus-like hair ratio 4. Thin/regular/thick hair shafts among terminal follicles 5. Independent observer clinical evaluation (mean improvement score using global macro-photographs) 6. Epidermal thickness perivascular inflammatory infiltrate, rete ride number, terminal/miniarized hair ratio, and collagen, reticular fiber and elastic fiber mesh quantity (3 mm punch biopsies) 7. Proliferative epidermal/follicular cells, newly formed blood vessels, and presence of bulge stem cell niches (immunohistochemistry)	1. Yes ( $p < 0.05$ ) 2. Yes ( $p < 0.05$ ) 3. Yes ( $p < 0.05$ ) 4. Yes ( $p < 0.05$ ) 5. Yes; 0.75/1 <sup>a</sup> 6. Yes ( $p < 0.05$ for most) 7. Yes ( $p < 0.05$ for most)	Patient self-satisfaction score following a Likert scale: 7 = very satisfied, 6 = satisfied, 5 = indifferent, 1 = unsatisfied, and 0 = very unsatisfied; most patients (15/19) declared noticeable hair loss decrease, 13/19 declared noticeable improvement in hair quality and appearance, and 11/19 stated they would continue with PRGF treatment
Alves [33], 2016	Yes	Yes	Yes	Yes	25 (24) 11 M, aged 18–65, stage II–V; 11 F, aged 18–86, stage I–III	#1–6: Phototrichogram & global photography 1. Anagen hair (%) 2. Telogen hair (%) 3. Anagen:telogen ratio 4. Hair density 5. Terminal hair density 6. Hair count	PRP vs. placebo: 1–3, 5, 6. No ( $p > 0.05$ ) 4. Yes, at 3 and 6 mos ( $p < 0.05$ ) PRP vs. baseline: 1–5. Yes ( $p < 0.05$ ) 6. No ( $p > 0.05$ )	n.a.
Gentile [34], 2015	Yes	Yes	Yes	Yes	23 (20) 20 M, aged 19–63, stage IIa–IV	#1–3: Computerized phototrichogram and global photography: 1. Hair count & total hair density 2. Terminal hair density 3. Epidermal thickness & hair follicle density (3-mm punch biopsy) 4. Keratinocyte proliferation and small blood vessel proliferation around hair follicles (immunohistochemistry) 5. Relapse of AGA	1. Yes ( $p < 0.0001$ ) 2. Yes ( $p = 0.0003$ ) 3. Yes ( $p < 0.05$ ) 4. Yes ( $p < 0.05$ ) 5. Four patients reported progressive hair loss at 12–16 mos	Physician and patient global assessment scale – results not reported
Cervelli [28], 2014	Yes	Yes	Yes	Yes	10 (10) 10 M, aged 20–52, stage IIa–IV	#1–4: Computerized phototrichogram & global photography: 1. Hair count 2. Hair density 3. Terminal hair density 4. Epidermal thickness & hair follicle density (3-mm punch biopsy) 5. Percentage of Ki67+ keratinocytes & blood vessel density (immunohistochemistry)	1. Yes ( $p < 0.0001$ ) at 3 mos 2. Yes ( $p < 0.0001$ ) at 3 mos 3. Yes ( $p = 0.0003$ ) at 3 mos 4. Yes ( $p < 0.05$ ) at 3 mos 5. Yes ( $p < 0.05$ ) at 14 wks	Physician and patient global assessment scale – results not reported
Singhal [12], 2015	No	Yes	No	No	20 (20) 16 M, aged 25–32; 4 F aged 32–35	1. Hair count (hair pull test) 2. Hair growth, hair volume, hair quality, fullness (global photographs)	1. Yes, pulled hair count was reduced by 65% (vs 0% in controls) <sup>a</sup> 2. Yes, hair growth noted in 6 patients after 7 days but in 4 patients after 15 days; yet, all patients (10) had good hair growth after 3 mos <sup>a</sup>	n.a.
Gupta [32], 2017	–	No	No	No	30 (30) 30 M, aged 25–35, stage III–VII	1. Hair density (CapilliCare trichoscan) 2. Hair diameter (CapilliCare trichoscan) 3. Independent observer clinical evaluation (global macrophotographs)	1. Yes (39.7 ± 16.5% increase compared to baseline) <sup>a</sup> 2. Yes (39.8 ± 17.2% increase compared to baseline) <sup>a</sup> 3. Average improvement = 30.2 ± 12.2%	Patient self-assessment questionnaire: treatment group reported 30 ± 13.1% mean improvement (range 10–70%); 93.3% reported complete cessation of hair fall by 2 mos; 66.7% reported increase in hair growth; 36.7% reported improvement in hair texture
Schiavone [36], 2014	–	No	No	No	64 (64) 42 M, mean age 28, stage II–V; 22 W, mean age 32, stage I–II	1. Hair count and hair thickness using Jaeschke 15 point scale rating of clinical change (macrophotographs examined by 2 independent evaluators)	1. Yes (mean change in clinical rating of 3.2 and 3.9) <sup>a</sup>	n.a.

**Table 1** (continued)

First author [Ref.], year	Study type				Characteristics of enrolled subjects (completed study)	Objective measures	Objective assessment of hair growth	Subjective assessment of hair growth
	rando- mized	controlled	blinded	half-head				
Gkini [40], 2014	No	No	No	No	22 (20) 18 M, aged 24–72, stage II–5a; 2 F, aged 58–72, stage I	1. Hair pull test 2. Hair density & quality (dermoscopic photomicrographs and macroscopic photographs)	1. Yes <sup>a</sup> 2. Yes, $p < 0.001$ ; overall improvement in hair density & quality per photographs	Patient self-assessment questionnaire: mean result rating of 7.1 on a 1–10 scale; 85% reported improvement in hair quality and thickness; 65% reported increases in hair density
Khatu [35], 2014	No	No	No	No	11 (11) 11 M, aged 20–40, stage II–IV	1. Hair pull test 2. Hair count (Trichoscan) 3. Hair loss (clinical examination, macroscopic photos)	1. Yes (81.81% achieved a negative pull test at 12 wks) <sup>a</sup> 2. Yes (average mean gain of 22.09 follicular units/cm <sup>2</sup> ) <sup>a</sup> 3. Yes (moderate improvement in hair volume and coverage with reduction in hair loss) <sup>a</sup>	Patient satisfaction questionnaire: mean overall satisfaction rating of 7 out of 10
Takikawa [37], 2011	–	Yes	No	Yes	26 (26) 16 M, 10 F, aged 28–59, thin hair in the frontal or parietal areas	1. Mean number of hairs (digital and dermoscopic imaging) 2. Mean cross sections of hairs (digital and dermoscopic imaging) 3. Epidermal thickness, collagen and blood vessel density around hair follicles (4-mm punch biopsy)	1. Yes <sup>a</sup> 2. Yes ( $p < 0.01$ ) 3. Yes <sup>a</sup>	Patients reported less depilation when shampooing, greater bounce/resilience of hair, maintenance of healthy hairs
Puig [30], 2016	Yes	Yes	Yes	No	26 (26) 26 F, stage II	1. Hair count (photography) 2. Hair mass index (Cohen HairCheck <sup>®</sup> system)	1. No ( $p = 0.503$ ) 2. No ( $p = 0.220$ )	13.3% of treatment group vs. 0% of control group reported substantial improvement in hair loss, rate of hair loss, hair thickness, and ease of managing/styling hair; 26.7% of treatment group vs. 18.3% of control group reported feeling coarser/heavier hair
Mapar [31], 2016	Yes	Yes	Yes	Yes	19 (17) 17 M, aged 24–45, stage IV–VI	1. Terminal hair count (magnifying glass) 2. Vellus hair count (magnifying glass)	1. No ( $p = 0.25$ at 6 mos) 2. No ( $p = 0.23$ at 6 mos)	n.a.

M, male; F, female; wks, weeks; mos, months. <sup>a</sup>  $p$  value not reported.

al. [29], Puig et al. [30], Mapar et al. [31], Gupta et al. [32], and Alves and Grimalt [33]. Studies from 2015 (Singhal et al. [12] and Gentile et al. [34]), 2014 (Gkini et al. [27], Khatu et al. [35], Schiavone et al. [36], and Cervelli et al. [28]), and 2011 (Takikawa et al. [37]) are also compared.

## Review of the Literature

### Studies with Positive Results

A recent pilot study by Anitua et al. [29] evaluated the use of plasma rich in growth factors in 19 subjects with AGA. Subjects were given 5 injections of PRP enhanced with platelet-rich growth factor (PRGF) activator to provoke release of growth factors and morphogens from the specimen (Table 3). Compared to baseline, all outcome measures showed positive results after 1 year of follow-up. Mean hair density, hair diameter, and terminal/vellus

hair ratio were among the measures showing statistically significant improvement ( $p < 0.05$ ). Histomorphometric evaluation also favored the use of PRP, showing improvement in epidermal thickness, perifollicular neoangiogenesis, and terminal/miniaturized hair ratio (Table 1), as well as decreased perivascular inflammatory infiltrates. Overall, patients were satisfied with their clinical improvement.

Alves and Grimalt [33] led a 25-subject randomized, blinded, half-head investigation, among which 22 completed the trial. The subjects were divided into 2 groups: group A, which received 3 mL of PRP on the right half of the head and 3 mL of saline placebo on the left, and group B, which received the same 2 solutions on opposite sides of the head. After 3 and 6 months, statistically significant improvements were detected in mean anagen hairs, mean telogen hairs, hair density, and terminal hair density in PRP-treated areas when compared with baseline ( $p <$

**Table 2.** Treatment protocols for included trials

First author [Ref.], year	PRP treatment sessions	Interval between sessions	Total follow-up period	Description of PRP application
Anitua [29], 2017	5	1 mo for first 4 sessions; final session 7 mos after start point	1 year	Intradermal injections of PRGF into hair-depleted areas
Alves [33], 2016	3	1 mo	6 mos (at 3-mo intervals)	Injections (0.15 mL/cm <sup>2</sup> ) within four 1×1 cm selected circular areas of the frontal & occipital scalp (marked with a dot tattoo) depending on the treatment-designated side of the scalp (vs. control side of the scalp received placebo (normal saline); no local anesthesia was used
Gentile [34], 2015	3	30 days	2 years (at baseline and 2, 6, 12, 16, and 23 mos after initial treatment)	Interfollicular injections of PRP (0.1 mL/cm <sup>2</sup> ) within 2 of the 4 selected areas of the scalp (physiologic solution into the other 2 areas), after cleaning skin with 70% alcohol; target areas were marked with semi-permanent tattoos for subsequent treatment and evaluation; local anesthesia was not used
Cervelli [28], 2014	3	1 mo	1 year (at baseline and 14 wks, 6 mos, and 12 mos after initial treatment)	Intradermal injections (0.1 mL/cm <sup>2</sup> ) into 2 of the 4 selected halves (e.g. frontal or parietal) (placebo was injected into the other 2 halves) after the scalp was cleansed with 70% alcohol; local anesthesia was not used
Singhal [12], 2015	4	2–3 wks	3 mos (at 1-wk intervals)	Injections using nappage technique (multiple small injections in linear pattern 1 cm apart) after area was cleansed with spirit and povidone-iodine
Gupta [32], 2017	6	15 days	6 mos	Scalp was activated by microneedling; then, PRP was massaged into the vertex of the scalp (10 cm from the glabella)
Schiavone [36], 2014	2	3 mos	6 mos	After local anesthesia (xylocaine 1%, with adrenaline 1:100,000) was administered, cutaneous inflammation was induced via application of gentle pressure using 1.0-mm-deep Scalproller to favor activation of injected platelets; then, superficial injections were administered 1 cm apart
Gkini [40], 2014	3 (+1 booster)	21 days (booster 6 mos after onset)	1 year	Injections (0.05–0.1 mL/cm <sup>2</sup> ) were performed using nappage technique in affected areas to a depth of 1.5–2.5 mm; a specific area was checked at all times by defining a “V” (Kang’s point) as proposed by Lee et al. [43]
Khatu [35], 2014	4	2 wks	12 wks	Nappage technique injections (2–3 ml) into a prefixed 1 × 1 cm squared area over the right parietal area; anesthetic cream was applied before each treatment after cleaning the skin with cetavlon, spirit, and povidoneiodine
Takikawa [37], 2011	5	2–3 wks	12 wks	Subcutaneous injection (3 mL) into selected 1 × 1 cm areas measured from the nasal tip and upper part of the auricular base
Puig [30], 2016	1	n.a.	26 wks (at 4-wk intervals)	Single subcutaneous injection within the 4 cm <sup>2</sup> area in the central scalp (termed the “hair check data box”), after anesthesia (2% lidocaine and 0.5% bupivacaine) was administered
Mapar [31], 2016	2	1 mo	6 mos (at 1, 3, and 6 mos after initial treatment)	Injections (1.5 mL of PRP) within one of two 2.5 × 2.5 cm square regions, at least 3 cm apart, in the scalp randomly assigned to be a case square (control square received 1.5 mL of normal saline); randomization of case and control squares was performed using a random number table; iron oxide- and titanium dioxide-containing substances were used to tattoo the corners of the squares

mos, months; wks, weeks.

0.05). Mean total hair density was the only measure found to be significantly increased in PRP versus placebo-treated areas ( $p < 0.05$ ).

Another randomized, blinded, half-head study performed by Gentile et al. [34] evaluated treatment outcomes of PRP in 20 male subjects. PRP was injected on half of the affected scalp of each patient, while the other side received physiological solution as control (Table 2). The study found a statistically significant increase in all

outcome measures, including mean hair count, hair density and terminal hair density, after 3 months of PRP treatment compared to placebo. Cervelli et al. [28] performed a very similar study to that of Gentile’s group with 10 men, and found similar positive results after 3 months of PRP. All outcome measures showed statistically significant improvement (Table 1). In both studies, microscopic analysis revealed that epidermal thickness and density of follicles were both increased compared to base-

**Table 3.** PRP preparation protocol in included trials

First author [Ref.], year	Mode of preparation	Activators	Centrifugation	Time of centrifugation	Platelet enrichment (fold)	Blood volume	PRP volume
Anitua [29], 2017	Single spin method	PRGF activator (BTI Biotechnology Institute)	580 rpm	8 min	$\times 2 \pm 0.3$ mean	18 mL	3–4 mL
Alves [33], 2016	Single spin method	Calcium chloride (10%, 0.15 mL)	460 g	8 min	$\times 3$	18 mL <sup>a</sup>	3 mL
Gentile [34], 2015	a. Cascade-Selphyl-Esforax system b. PRL platelet-rich lipotransfert system	Ca <sup>2+</sup>	a. 1,100 g b. 1,200 rpm	a. 10 min b. 10 min	–	a. 18 mL b. 60 mL	a. 9 mL b. 20 mL
Cervelli [28], 2014	Cascade-Selphyl-Esforax Kit	Ca <sup>2+</sup>	1,100 g	10 min	–	18 mL	9 mL
Singhal [12], 2015	Double spin method	Calcium chloride (9:1 ratio)	a. 1,500 rpm b. 2,500 rpm	a. 6 min b. 15 min	–	20 mL	8–12 mL
Gupta [32], 2017	Double spin method	–	–	–	–	–	–
Schiavone [36], 2014	GPS III Platelet Separation System a. Single spin at baseline b. Double spin at 3 months	No (Scalproller used to favor–platelet activation (see Table III))	–	–	a. $\times 6-7$ ( $\times 3.5-4$ with addition of plasmatic protein concentrate) b. $\times 4$ with addition of plasmatic protein concentrate	a. 60 mL b. 40 mL	a. 6–8 mL PRP + 3–4 mL of plasmatic protein concentrate = 9–12 mL; 0.2–0.3 mL per injection b. Same as above
Gkini [40], 2014	RegenA-PRPCentri (Regenlab) Single spin method	Calcium gluconate (0.1 mL per 0.9 mL of PRP; 1:9 ratio)	1,500 g	5 min	$\times 5.8$	16 mL	6 mL (0.05–0.1 mL/cm <sup>2</sup> )
Khatu [35], 2014	Manual Double Spin	Calcium chloride (1:9 ratio)	a. 1,500 rpm b. 2,500 rpm	a. 6 min b. 15 min	–	20 mL	2–3 mL
Takikawa [37], 2011	Manual Double Spin	–	a. 1,700 rpm b. 3,000 rpm	a. 15 min b. 5 min	$\times 6$	15 mL	3 mL
Puig [30], 2016	Angel PRP system (Cytomedix)	No	–	–	$\times 2.75-3.4$	60 mL	10 mL
Mapar [31], 2016	Double spin method using Tubex PRP tube (Moohan Enterprise)	Calcium gluconate (0.1 mL per mL of PRP)	a. 3,000 rpm b. 3,300 rpm	a. 6 min b. 3 min	$\times 3$	9 mL <sup>b</sup>	1.5 mL

<sup>a</sup> Plus 2 mL of sodium citrate. <sup>b</sup> Plus 1 mL of acidic citrate dextrose.

line ( $p < 0.05$ ) 2 weeks after completion of PRP therapy. Immunohistochemistry was performed, and the percentage of Ki67+ cells was also increased in both basal keratinocytes of the epidermis and hair follicle bulge cells at 2 weeks after PRP treatment ( $p < 0.05$  compared with baseline), suggesting an increase in keratinocyte proliferation. Investigators also observed an increase in small blood vessel count around the hair follicles ( $p < 0.05$  at 2 weeks after PRP compared with baseline), supporting the notion that PRP promotes angiogenesis via the release of vascular growth factors.

Singhal et al. [12] conducted a small controlled clinical trial to compare PRP with medical treatment in 20 subjects, 8 males and 2 females in each treatment group. Hair growth was seen in 6 subjects after just 7 days, but in 4 subjects after 15 days. Yet, by the end of 3 months, all evaluated parameters (Table 1) showed superior outcomes in PRP-treated subjects than in control subject, although no statistical analysis was reported on the observed data. In comparison, the subjects managed with medical treatment showed no improvement in hair pull test or hair growth.

An open-labeled pilot study conducted by Gupta et al. [32] involved 30 male participants. Each participant received 6 PRP massage treatments after the scalp was first activated by microneedling [38, 39]. After 6 months of follow-up, Trichoscan evaluation of the vertex (10 cm away from glabella) showed significant increase in hair diameter and hair density. Evaluation of global photographs by a blinded observer showed an average improvement of  $30.2 \pm 12.2\%$ . Self-assessment questionnaires likewise revealed improvement with PRP treatment (Table 1). Treatment response was more significant in those with lower-grade alopecia in terms of hair diameter ( $p = 0.0446$ ) and hair density ( $p = 0.0196$ ). Efficacy was more pronounced in subjects who had shorter duration of disease prior to therapy and subjects without family history of alopecia, with improvement in hair diameter ( $p = 0.0485$  and  $p = 0.0272$ , respectively) and hair density ( $p = 0.0096$  and  $p = 0.0114$ , respectively).

Multiple preliminary and observational studies performed in 2014 all concluded that PRP could have a positive therapeutic effect for male and female subjects with AGA. Schiavone et al. [36] led an observational study in which 64 subjects received 2 injections of L-PRP mixed with plasmatic proteins 3 months apart. Hair count and thickness were visibly improved after 6 months of PRP treatment; approximately 40.6% of study participants reached at least a moderate level of improvement.

Gkini et al. [27] performed a prospective cohort study with 22 subjects, of which 20 completed the study. After 3 treatments, they reported increased hair density compared to baseline at 3, 6, and 12 months after PRP ( $p < 0.001$ ), as well as improvements in density and thickness. In this study, milder forms of alopecia (Norwood-Hamilton grade II–III) responded better to PRP treatment than more advanced cases. In addition, subjects with vellus hair had better results. Investigators also suggested that the PRP treatment appeared to lead to increases in hair diameter more than hair count. Khatu et al. [35] also led a small prospective cohort study to investigate PRP efficacy in 11 subjects. After 4 sessions of PRP, 9 subjects reverted to having a negative hair pull test. Hair volume, coverage and follicular hair unit count were improved. Hair counts were noted to be increased from 71 to 93.09 on average. Significant reduction in hair loss was evident per patient questionnaires. Both Gkini et al. [27] and Khatu et al. [35] assessed patient satisfaction, and found the reported means of 7.1 and 7.0 out of 10, respectively.

In 2011, Takikawa et al. [37] led one of the earliest controlled clinical trials of PRP containing dalteparin and

protamine microparticles (D/P MPs) in 26 subjects suffering from frontal or parietal hair loss. Solutions of either PRP with D/P MPs, or PRP and saline, were injected at sites of hair loss (13 subjects each), with controls being opposing sides with equal hair density. After 5 treatments over 12 weeks, increased mean hair count was observed in both PRP- and PRP&D/P MP-treated areas relative to control areas. Additionally, significantly increased hair cross section was observed in both PRP- and PRP&D/P MP-treated areas relative to control areas. Subjective reports by participants noticed less hair loss with shampooing, and greater bounce and resilience of their hair texture. Histological analysis of punch biopsy revealed thickened epidermis, proliferation of collagen fibers and fibroblasts, and greater number of blood vessels around hair follicles in PRP-treated areas. No severe side effects, including infection and hematoma, were reported, though participants noted some temporary pain at the injection sites.

#### *Studies with Negative Results*

Two studies did not show statistically significant improvement in the outcomes assessed. Puig et al. [30] carried out a double-blind, randomized, placebo controlled, multicenter trial involving 26 women with female pattern hair loss. Fifteen women were randomized to the PRP group and 11 to the placebo group. Investigators marked a 4-cm<sup>2</sup> area in the central scalp, where hair was repeatedly analyzed for hair mass during the study, using the HairCheck. Study participants received one injection of either PRP or normal saline within 4 cm from this area at week 0 (Table 2). At week 26, no statistically significant difference was found between treatment and control groups in terms of hair count and hair mass index (Table 1). PRP-treated subjects did, however, report subjective improvement in hair loss, rate of hair loss, hair thickness, and ease of hair styling, which none of the placebo-treated subjects noted. This was the only study in which subjects received only one PRP or placebo treatment.

The second study was a prospective, half-head comparative pilot study carried out by Mapar et al. [31] on 17 men with AGA. Investigators injected PRP or normal saline (Table 2) during 2 sessions 1 month apart. Results showed a mean decrease in the number of terminal and vellus hairs after 6 months of PRP treatment, assessed using a magnifying glass. Consequently, investigators found no statistically significant difference in the outcomes assessed between treatment and baseline.



## Discussion

After careful evaluation of the 12 investigations cited in this review, the available evidence suggests a promising use for PRP as an alternative treatment for AGA. Ten of the 12 studies remarked positively on the therapeutic potential of PRP for the treatment of AGA. Among them, 6 studies demonstrated a statistically significant improvement following treatment with PRP using objective measures [28, 29, 33, 34, 37, 40] and 4 additional studies showed hair improvement (e.g., hair density, diameter) with PRP, although no *p* values or statistical analysis was described [12, 32, 35, 36]. Two studies noticed greater improvement in lower-grade AGA [27, 32], while one noted increased improvement in higher-grade AGA [36]. Only one study, by Mapar et al. [31], concluded that PRP was not effective in treating AGA through analysis of terminal and vellus hair count. However, in this study, only 2 treatments were administered, and outcomes were assessed under magnifying glass, which may not be the best method to measure results. Further, neither a physician nor a subject self-assessment was performed. Another study, by Puig et al. [30], did not detect significant improvement using objective measures (hair count or hair mass index) after PRP treatment. In this study, however, only one treatment was administered and, of great importance, the PRP used was not activated, thereby impeding its full therapeutic potential. Nevertheless, they did document subjective improvement (such as less hair loss and improved hair thickness) [30]. In the studies where no statistical analysis was reported [12, 32, 35, 36], the authors remarked positively on hair growth, volume, coverage, and mean hair density. Overall, all the studies in which a minimum of 3 PRP treatments were administered showed improvement in at least one objective measure.

Among the studies reviewed, it is evident that there lacks a standardized treatment protocol for the application of PRP as well as standardized evaluation methods. Without such parameters, it is difficult to adequately assess the effectiveness of PRP for its restorative potential on AGA and compare the results between studies. In conducting this analysis, certain methodological differences were noted. With the initial PRP preparation, there is a lack of consensus regarding the mode of preparation, addition of activators, centrifugation time and speed, platelet concentration attained, and volume of blood and PRP used (Table 3). Three studies, for example, used calcium chloride as an activator [12, 33, 35], while 2 studies used calcium gluconate [31, 27], 1 study used PRGF activator [29], and 2 other studies did not specify the chemical that

provided  $\text{Ca}^{2+}$  [28, 34]. Treatment protocols also varied in the number of sessions, time interval between treatments, administration procedure, and follow-up period (Table 2). In terms of research study designs, they ranged from pilot studies to randomized blinded trials (Table 1).

### *Study Design*

The variations in study design have contributed to difficulty in interpreting results across included studies. The studies were stratified not only through isolated PRP or PRP in combination with other treatments, but also by subject sex, severity of alopecia, sample size, randomization, and control groups, further obscuring PRP treatment results. Each study employed a very unique treatment protocol. Of the studies reviewed, 7 mentioned the use of a control group [12, 28, 30, 31, 33, 34, 37], and 5 conducted the experiment without a control [27, 29, 32, 35, 36]. Five studies mentioned randomization of subjects into treatment or control groups [28, 30, 31, 33, 34], while 3 specifically mentioned that they did not randomize subjects [12, 27, 35], potentially introducing bias. For instance, although a good portion of studies included both male and female subjects [12, 27, 29, 33, 36, 37], others included only males [28, 31, 32, 34, 35] or only females [30]. Since male and female pattern hair loss has different manifestations and may have different mechanisms, it may be inappropriate to extrapolate the results to both sexes in studies examining only a single sex. Moreover, the power of most of the studies was compromised due to the small sample sizes. Most studies enrolled only 10–30 subjects [12, 27–35, 37], and the largest study examined 64 subjects [36]. All studies had inclusion and exclusion criteria.

### *Study Duration and Treatment Protocols*

A sufficiently long duration of follow-up facilitates accurate assessment of clinical improvement in measures such as hair counts/density, hair thickness, and overall alopecia as well as evidence of relapse. Many of the clinical trials evaluating the effectiveness of hair growth promotion monitor the subjects for 24–26 weeks [13, 41]. Yet of the 12 studies reviewed, 2 evaluated subjects through a short period of 12 weeks [35, 37], another one for 3 months [12]. An overwhelming majority of studies (8 studies) lasted for 6 months to 2 years. Interestingly, in the study with the longest follow-up (2 years), Gentile et al. [34] reported 4 cases of relapse at 1 year and noticed progressive hair loss was even more evident at 16 months, thereby providing evidence for the importance of following subjects for at least 12 months to monitor relapse, and

for the necessity of repeated PRP treatment. Additionally, treatment sessions varied from 1 [30] to 6 sessions [32]. Statistical significance was not established in 2 out of the 3 studies that provided 2 treatments or less [31, 30], suggesting that greater than 2 PRP sessions are likely necessary for true efficacy.

#### *Outcome Measures*

Objective assessments detailed in the various studies included hair count/density, hair mass, hair volume, hair growth, hair diameter/thickness, cross section, anagen:telogen ratio, epidermal thickness, follicular quantity, Ki67+ keratinocyte count, blood vessel quantity/density, and hair loss evaluated through the use of phototrichograms, global photographs, magnifying lenses, Hair-Check<sup>®</sup>, histology and immunohistochemistry, and hair-pull tests. Although most studies used quantitative and qualitative methods to evaluate measures such as hair count, hair density, and hair thickness, the assessments used varied widely, making it difficult to accurately evaluate and compare clinical improvement of AGA. While no universal assessment can be recommended, additional studies may benefit from standardizing their quantitative and qualitative evaluation methods to include those known to be reliable such as global photographs and phototrichograms [42]. Further, only 8 of the studies qualitatively measured patient satisfaction with evaluations such as surveys and questionnaires [27–30, 32, 34, 35, 37], although 2 did not report the findings [28, 34] (Table 1). Patient satisfaction is currently a top priority at health systems; therefore, documenting patient satisfaction and self-reported outcomes can prove advantageous in determining not only which PRP treatments are effective, but also those that will result in high compliance.

#### *Side Effects*

There were no major adverse effects such as scarring, progressive worsening, or infections reported in any of the above studies. Notably, mild headache, tolerable and temporary pain during treatment, mild itching and desquamation, and transient edema were reported by some subjects after PRP treatment.

#### *Suggested Treatment Protocol*

Current protocols for PRP preparation and administration are highly varied. After careful evaluation of published studies, we have the following suggestions for using PRP as treatment for AGA. We would recommend protocols for PRP preparation that include a double-spin centrifugation method, consisting of a first spin at 1,500–

1,700 rpm for 6–10 min followed by a second spin at 2,500 rpm for 10–15 min [20]. Other studies, however, used a single spin method (ranging from 460 to 1,500 g/580 rpm for 5–8 min) and also reported beneficial outcomes. The use of an activator, preferably a Ca<sup>2+</sup>-containing compound such as calcium chloride or calcium gluconate, would activate platelets for release of growth factors and cytokines and would likely provide better results. The amount of total PRP injected varied greatly between the studies reviewed, making it difficult to deduce which volume was necessary for optimal results. Based on studies with positive results, an average volume of 6.2 mL (range of 3–12 mL) of pure PRP with a mean platelet enrichment of 3–6× should be injected. Intradermal injections should be made at approximately 0.1 mL/cm<sup>2</sup> in selected scalp areas using the nappage technique (multiple small injections in a linear pattern 1 cm apart to a depth of 1.5–2.5 mm). Anesthesia is not required; however, we recommend it based on reports of pain during the procedure. A minimum of 3 sessions at 1-month intervals are recommended, although 2- and 3-week intervals were also a common regimen that gave positive results.

#### **Conclusion**

PRP has demonstrated therapeutic effectiveness for AGA in 10 of the 12 reviewed studies. Although our literature review suggests PRP is a potential treatment option for AGA, several study design limitations need to be addressed before PRP is widely introduced as a treatment option in the clinical setting. The field would benefit from additional large-scale double-blind, randomized controlled studies treating both men and women, with standardized PRP preparation methods and administration protocol, repeated treatments, standardized objective data documentation and evaluation, physician and subject assessment, isolating the effects of PRP in different grades of AGA, and performing long-term follow-up.

#### **Disclosure Statement**

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