Platelet Rich Fibrin Matrix with Facial Collagen Genesis and Epidermal Regeneration

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Abstract

Objective: To determine if Platelet-rich fibrin matrices can induce an improvement in facial volume and mollify epidermal age-related negative remodeling.

Introduction

Dr. Anthony Stefani has demonstrated that PRP is effective for restoring facial volume in such regions as the nasolabial folds. Currently, there has not been any published study which shows that multiple injections of PRP throughout the face could induce collagen genesis or thickening of the dermis as well as overall epidermal rejuvenation in the older phenotype. It is our intent to determine in this pilot study if platelet-rich fibrin matrices are an appropriate aesthetic protocol for facial rejuvenation.

Methods

Twelve subjects between the ages of 44 and 56 who are healthy with a normal BMI as well as non-smokers were selected to participate in this study. We injected these individuals with their own autologous PRP throughout the mid and lower face. These injections were within the dermis and subdermis. The injection sites were three across the face and there were four rows from the zymatic arch to the jaw line. Thus, we injected twelve distinct areas on each side of the face. The amount of PRP plasma was approximately fifteen milligrams, which was calibrated as a function of the mild elevation of the epidermis. This aesthetic strategy was executed twice a month, or nine times during a four month period. Furthermore, facial volume loss was treated with a more robust prophylaxis, as several cc's of PRP was injected into those facial regions.

Results and Discussion

In our view, autologous PRP prophylaxis engendered an improved collagen genesis which translated to enhanced facial volume as well as significant epidermal texture rejuvenation. It is likely in certain phenotypes that PRP cosmetic treatments may be as effective as current laser protocols and fat grafting. We observed no patient down time, and as a function of our questionnaire, subject's satisfaction was overwhelmingly substantial. PRP prophylaxis appears to be a viable strategy with multiple injections to treat a face between the ages of 35 and 55 which exhibits age and sun related facial aberrancies in terms of unattractive and negative remodeling.

Introduction

There has been a myriad of studies demonstrating the efficacy of PRP for orthopedic and cosmetic purposes.1,2,3,4 and 5 Dr. David Crane and Peter Everts, 2008, have stated that "PRP matrix grafts, along with other biological graft techniques are becoming more prevalent in the treatment paradigms of musculoskeletal medicine. These PRP matrix grafts provide effective, safe, and amelioratively low cost treatment options to patients who have time and wherewithal to allow collagen synthesis and maturation at the graft site. PRP matrix grafts appear to restore tissue homeostasis and the biotensegrity of collagen".6 David Karli and B. Robinson, 2010, have likewise stated that the body's own biological modulations can be efficacious in terms of enhancing the wound healing process. The literature is extensive, demonstrating that an increase in growth factor concentrations can be propitious in the wound healing process. Moreover, Karli and Robinson, 2010, purport that "this case report demonstrates sustained, subjective and functional improvement with near complete repair on MRI with a single application of platelet rich plasma in a severe tendon injury".7 J Menetrey and Etal show that higher concentrations of growth factors improved muscle healing.8 Conversely, a recent article in the British Journal of Sports Medicine, 2010, revealed that the IOC consensus paper on the use of platelet-rich plasma in sports medicine was far more conservative in its investigation of the use PRP in the treatment of sports related musculoskeletal perturbations.9 Clinical researchers like Stephen Barret have argued that higher concentrations of platelet rich growth factor provides excellent wound healing profile.10

Molecular medicine in the twenty-first century comprehends that cytokine activation of cellular dynamics is preferable to exogenous nonspecific pharmacological agents as well as highly invasive surgical protocols. Kilroy et al., 2007, purported that "the cytokine expression profile has a direct relevance to adipose tissue function and healing disease".11 The literature is expanding with studies which indicate that higher concentrations of growth factors can have edifying effects on transcriptional and translational activities on a diverse array of human cell types. Elizaveta Kon et al. 2008, have reported that PRP intervention for the utilization of the higher concentration of growth factors engenders a rejuvenation of tissue. This group stated that "this report outlines the first in vivo investigation of the use of autologous growth factors to treat jumper's knee by means of PRP injection, and demonstrate that this is a method to improve tendon healing and promising results".12

The intent of this investigational study is not on the heuristic merit of PRP therapies for wound healing for sports injuries; but rather whether this biological modulatory protocol can actually have significant cosmetic efficacy. Anthony Sclafari's initial 2009 study indicates that PRP therapies can have significant cosmetic benefits.13 This plastic surgeon employed higher concentration of platelets with their growth factors into a patient's nasolabial folds to mitigate deficiency in volume in this particular facial region. His results were promising and cosmetically effective.

Bob Jackson's 2003 article stated that PRP therapy actually significantly reduced senoma formation during the abdominoplasty procedure.14 This cosmetic surgeon stated that "the application of platelet rich plasma as a natural fibrin matrix delivers growth factors to the wound and seems to promote more rapid healing".15 Ferdinand Becker in an unpublished pilot study showed that PRP therapies provided patients with superior aesthetic results after undergoing a cosmetic procedure. He writes that "the use of platelet concentrate has demonstrated excellent results by enhancing and accelerating wound healing. Patient's own platelet concentrate has included experiencing significantly less swelling, bruising, and overall morbidity". 16 Dr. Clevens work in Melbourne, Florida, likewise has shown an enhanced healing time when utilizing PRP in conjunction with a midface surgical procedure.17 Moreover, many others such as Thomas Tzikas have stated that the use of PRP for cosmetic purposes during a surgical procedure improves healing and reduces bleeding.18 Dr. Patrick Abuzeuni and Robert Alexander found that PRP with fat grafting enhances fat transplantation. They discovered that "this technique is intended to promote or accelerate the healing face after grafting, enhanced the

intended augmentation retention value of volume, potentially reduced secondary calcification, and microcystic formation, and maximized the transplant unit volume by reducing extracellular fluids transferred with grafts".19

Walter Tom wrote the following about the use of PRP for the use of aesthetic rejuvenation. "The new paradigm for natural facial rejuvenation is based on revolumizing the aging deflation of the face...an answer may be our own plasma with a concentrated fraction of platelets. Platelet rich plasma [PRP] has the potential not only to fill deflated volume, but may indeed trigger cell migration and differentiation. If this is born out, then we have a relatively inexpensive filler that is autologous with long term benefits and a minimum of side effects". 20 Recently. Katherine St. Louis discussed in a New York Times article that the benefits of the use of PRP for facial rejuvenation.21 The dermatological phenomena of what she referred on the "vampire face lift", which was licensed by Dr. Charles Reynolds; and represents a new frontier in biological medicine. However, the antidotal commentaries which were cited in this article seem puerile, and more hypothetical than a sophisticated clinical procedure.8 It is our purpose in executing this pilot study to utilize a complex methodology to determine if it is possible to reduce and alleviate volume deflation; and aesthetically improve the impoverished epidermal texture in the older phenotype in a manner which is consistent with what is attained with an average SMAS face lift in conjunction with a fractionated laser resurfacing. In summary, it is our intention to determine if many facial injections of PRP can induce dramatic dermatological remodeling of an older cosmetically compromised face. The literature seems to support that autologous platelet growth factors are able to remodel cellular tissue in a manner which is consistent with facial fat grafting.

These are some the essential aesthetic issues which are the etiological basis of this dermatological study. In general, we are interested in understanding whether we can reeducate the transcriptional machinery of senescent cells to behave in a more robust, youthful manner with respect to overall protein translation and synthesis. This study involves nine treatment sessions over a five month period where both the mid and lower face are treated. The second phase of this study will examine how long the aesthetic benefits of multiple PRP sessions are sustained over a six month time period. It appears that growth factors, in most instances, have more ameliorative merit than stem cells and are very important in terms of cellular remodeling.

Methodology

Study Design: Ten subjects were selected who met our criteria. We excluded subjects who were overweight (40 lbs over normal BMI), smokers, and those taking several or more prescribed medications, especially for depression and anxiety. The age range was from 44 to 57. Gender was not a delimiting factor. Some of the subjects previously had minimum cosmetically invasive procedures. One patient underwent a midface lift five years ago with limited cosmetic benefit. Several of the subjects have been treated with fillers, botox, and laser protocols. Their skin types ranged between two and four on the Fitzpatrick rating scale.

All the subjects exhibited excessive epidermal sun damage and aging. Some of them had letiginious lesions and dyschromia. Every subject had considerable volume loss in the nasolabial fold region as well as adjacent facial quadrants. None of the subjects were pregnant, nursing, or with any kind of skin lesion which was inflamed or infected. These individuals did not have a herpes simplex outbreak within the preceding five years. Furthermore, the subjects were not using any kind of dermatological prescription topical agent like retinoid acid. All subjects never reported an increased sensitivity to light or any kind of skin-related debilitating perturbation.

The autologous platelet concentration was prepared from a 60 mL or 20 mL kit with anti-coagulant. The blood was extracted and prepared using the Smart PREP system of Harvest Technology, Plymoth, Mass. This process provides a 9cc or 3.5cc of platelet concentrate with higher levels of growth factors. In general, a 60 or 20cc syringe is prefilled with 5cc of a citrate based on an anticoagulant (ACD-A) which is part of Harvest's disposable kit. Approximately 55cc or 20cc of patient blood is withdrawn from a venous puncture in the upper arm into either a 60cc or 20cc syringe.

The anti-coagulated blood is then placed into a blood chamber of a processing vessel which is disposable. This disposable unit is then set into the centrifuge locator cup of the Smart PREP system. The counter balance weight is placed in the opposite rotator cup, and in most instances, we had a second disposable kit to balance the unit. The lid of the system is closed and the processing of the blood is commenced. The process is automatic and takes twelve minutes. The centrifugation separates red blood cells from the plasma. This process enables platelets to create a pellet at the bottom of the disposable kit's plasmid

chamber.

The disposable kit's plasmid chamber contains red blood cells and in a second part harbors platelet concentrate. The platelet poor plasma (PPP) is removed. We primarily utilize the platelet concentrate. However, there was some degree of platelet poor plasma which was infused to create the concentrated platelet rich plasma or PRP solution. We did not use an activator for the platelet rich plasma; and it was placed by our technician into 31 gauge 1cc syringes. A 20cc kit produced 3.5cc of viable PRP concentrate, whereas a 60cc system yielded 9cc or more of PRP concentrate.

All patients were not treated with lidocaine. For most subjects, the pain of multiple injections was tolerated. One patient got her own ice packs to cope with the perceived pain. Moreover, we did not use any kind of topical lidocaine. A medical assistance cleaned the facial surface of all patients. A plastic surgeon marked which facial regions were to be treated with PRP injections. In general, a large kit of 60cc was used for subjects with excessive volume loss. Most patients had at least two regions on each side of their face which needed to be treated for excessive volume deflation.

The injections involved three points across, and a total of four rows for each side of the face from the zyomatic arch to the lower jaw line. Each injection into a particular mark represents a titration into the dermal and subdermal region; and in most instances the 31 gauge needle only punctured 50% into the facial surface. In most instances, a 1cc 31 gauge syringe treated most of the twelve marks on each side of the face. Each injection was approximately 5 milligrams. The clinical practitioner utilized a direct lateral approach into the surface of the face. Conversely, when treated the nasolabial fold the clinician employed a direct vertical angle, where the needle was placed fully into a specific subdermal vector plane. These individuals also received twelve injections on each side of their face involving 1cc of PRP concentrate. In general, there was some degree of variability in terms of treatment strategies for a particular phenotype as a function of their level of volume deflation. Furthermore, all patients did receive twelve standardized injections on each side of their face, but there was sufficient variability in terms of treatment protocols for facial volume deflation.

The injection sessions were fourteen days apart and took between ten and fifteen minutes. For most patients, the pain was well tolerated. There were nine separate injection sessions over a five month period. We did not observe any severe complications in any of the ten phenotypes who participated in the study. Most patients did not experience pain after each treatment session. There was never a need for the use of prescription medication for any subject who was involved in the study. Some of the subjects did observe in areas of deeper injection, such as in the nasolabial fold region, bruising which was resolved in a few days.

We did not see any extensive erythema or any edema; and limited facial swelling resolved within twelve hours. Thus, these injections were well tolerated by subjects with limited complications. However, one patient seemed to have a heightened pain sensitivity after each session. This woman reported that she was suffering from severe general anxiety syndrome which was not currently being treated. All subjects were given fourteen questions to answer at the final session which was concerned with their satisfaction with these treatment protocols.

Patient Questionnaire for PRP Study

Please circle numbers on the scale of 1 to 10 (1 being lowest 10 the highest) to score how strongly you agree with the following statements.

Question 1: I've received significant facial rejuvenation:

1 2 3 4 5 6 7 8 9 10

Question 2: The epidermal texture of my face has improved:

1 2 3 4 5 6 7 8 9 10

Question 3: These injections made me look younger:

1 2 3 4 5 6 7 8 9 10

Question 4: My face looks like I had a facelift:

1 2 3 4 5 6 7 8 9 10

Question 5: This procedure added volume to my face:

1 2 3 4 5 6 7 8 9 10

Question 6: This cosmetic strategy enabled me to become more attractive:

1 2 3 4 5 6 7 8 9 10

Question 7: I would undergo these treatments again:

1 2 3 4 5 6 7 8 9 10

Question 8: I will recommend PRP facial treatments to my friends:

1 2 3 4 5 6 7 8 9 10

Question 9: This cosmetic strategy improved my quality of life:

1 2 3 4 5 6 7 8 9 10

Question 10: I prefer this treatment over a facelift:

1 2 3 4 5 6 7 8 9 10

Question 11: The aesthetics of my face are still improving with multiple treatments:

1 2 3 4 5 6 7 8 9 10

Question 12: I think this medical protocol is appropriate for the general public in terms of enhancing facial beauty:

1 2 3 4 5 6 7 8 9 10

Question 13: I think this aesthetic therapy is a superb strategy for reducing the effects of facial aging:

1 2 3 4 5 6 7 8 9 10

Question 14: I am very satisfied with the cosmetic results I achieved from participating in this study:

1 2 3 4 5 6 7 8 9 10

Results

This initial pilot study involving twelve subjects was to determine if platelet-rich plasma actually had sufficient cosmetic efficacy in terms of observable and viable aesthetic changes. It is essential in a larger cohort study to begin doing genetic analysis of the cellular system protein expression changes in the dermis; and even 3-D imaging in order to understand the degree of dermal thickening as well as other molecular ameliorative modifications. Moreover, histological analysis and 3-D imaging was not utilized in this study, but would be an essential facet of any other future investigation. In general, we relied on a patient satisfaction rating scale and their overall perceptions to verify the effectiveness of this innovative cosmetic protocol.

All patients who participated in this study perceived that they received facial rejuvenation; and they all rated their results at the highest level. These subjects evaluated the first seven questions of this study with the highest rating possible. In general, they were extremely satisfied with the improvement of their epidermal texture; and they felt that these injections enabled them to look younger in appearance. They all believed that this procedure induced their face to appear as though they underwent some type of invasive surgical procedure.

All subjects regarded that this cosmetic strategy significantly added volume to their face; and therefore they felt more attractive. All eight subjects who completed this investigative study would unanimously undergo these treatments again. Thus, the eight patients of this study answered the questions concerned with their satisfaction with the highest rating possible in terms of improvement of facial appearance. They all perceived their epidermal texture as improving, looking younger with greater volume, and would undergo these treatments again in order to become more attractive.

Two of the eight subjects answered seven and eight whether they would recommend this procedure to their friends. These two women did not enjoy having these injections without lidocaine; and they believed that their friends would not necessarily be able to cope with the discomfort associated with this protocol. Six of these patients stated that this procedure absolutely improved their quality of life, and one responded with a nine rating, one with an eight, and another with a seven. All seven subjects stated that they preferred this strategy over that of a face lift, and one woman responded with a score of an eight. It seems that all the subjects who completed this study preferred multiple treatments of PRP to that of having a highly invasive surgical procedure. Six of the eight subjects rated this question a ten to the fact that their face is still improving, and one person responded with a seven and another with a score of eight. All seven subjects expressed a rating of ten in terms of the appropriateness of this procedure for the general public. These subjects evaluated the use of PRP treatments as an effective strategy for reducing facial aging. Furthermore, the subjects who participated in this study were 100% satisfied with their cosmetic results; However, one woman rated her satisfaction with a score of nine.

Clinical Observations of Medical Practitioners who Participated in this Investigational Study

We all concurred that four to six injections of PRP in the older phenotypes who participated in this study was highly efficacious in terms of cosmetic benefits. In certain subjects, multiple injections seemed to be as effective as a robust fractionated CO2 treatment with respect to facial rejuvenation of the epidermal texture and mitigating skin laxity. However, the results may not necessarily be consistent with a larger cohort population. We believe that there is a certain degree of variability in the subject population in terms of the degree of skin tightening, which is affected by age and overall physical health status. Furthermore, we all observed results in the majority of subjects consistent with one pass of a fractionated CO2 treatment.

The two clinicians who participated in this study have performed many facial fat grafting procedures. They observed that after multiple treatments of a PRP protocol, that volume restoration for several patients is as judicious as employing facial fat grafting. PRP injections provides a patient with a very naturalistic symmetry as well as ameliorating facial volume loss compared to the unpredictability associated with facial fat grafting. There appears to be sufficient evidence from this initial pilot study that multiple PRP injections in a particular facial quadrant may be preferable in some instances to facial fat grafting.

In the eight phenotypes who participated in this study, only two subject had aesthetic results which would be applicable to a SMAS mid facelift. These two women are in their late forties, and demonstrated at the initial phase of the study limited facial laxity and had only moderate volume deficiency. We observed after multiple PRP treatments that there was a mollification of facial laxity and restoration of facial volume loss as well as robust epidermal remodeling. In our view, eight injections in phenotypes over fifty would not be comparable to a mid-facelift in most phenotypes. However, in some patients in their late forties with a healthy lifestyle, they could possibly achieve the aesthetic benefits consistent with a mid-facelift. This particular cohort population now have the option to undergo multiple treatments of PRP in lieu of a highly invasive surgical procedure.

Three subjects were eliminated from this investigation for missing two consecutive treatment sessions. One woman, who was 57, with excessive skin laxity dropped out, as her results were more minimum than the other cohorts who completed this research study. This female was advised as a function of her aberrancies in soft tissue remodeling that she should pursue an invasive mid-facelift procedure as it would be the only cosmetic protocol to mitigate her inordinate facial aging. Thus, in our opinion, multiple PRP treatments are not appropriate for anyone who is over sixty with excessive skin laxity or in general with pejorative soft tissue remodeling. This biological cosmetic protocol is most appropriate for a phenotype with sun damage in their late thirties to early fifties without excessive soft tissue aberrant remodeling.

We have discerned that younger subjects who are physically active in their forties will have the most dramatic aesthetic results with multiple treatment sessions. Moreover, two younger females in their late twenties with non-age related volume deficiencies were treated twice with PRP injections; and their aesthetic results were dramatic in terms of improving their overall appearance. Thus, we believe that younger women could have significant aesthetic enhancement with two treatment sessions of PRP therapy. This innovative cosmetic protocol is definitely efficacious with respect to improving facial beauty in the younger phenotype.

One male patient who was treated twice with PRP injections in the scalp has demonstrated a greater hair density in areas of his scalp which were formerly very thin. In our view, it is necessary for other investigational studies to be conducted in a larger male cohort population, if PRP injections can actually thicken hair density. In summary, patients receiving the greatest benefit of PRP injections had to be treated at least five times. We did view smaller cosmetic benefits with several more injections. In the second phase of this study, we will comprehend how long these cosmetic benefits will be sustained after six months.





Discussion

Twenty-first century cosmetic medicine will be in the ensuing decades, much more reliant on the body's own endogenous molecular machinery to ameliorate protein expression and overall tissue remodeling. Cytokine matrices will be the molecular modulatory agents to govern cellular transcriptional processes in a manner which would be cosmetically beneficial to the patient. It is likely that highly invasive surgical aesthetic procedures will be replaced to a certain degree by cosmetic molecular science. This current pilot study showed that ten or more of the body's own endogenous growth factors expressed by platelets in higher concentrations can attenuate the aberrant physiological remodeling associated with excessive sun damage and molecular aging.

Cytokines or growth factors expressed by platelets are normally involved with body's wound healing process. PRP rich plasma has at least five times greater concentration of growth factors than would normally occur in circulation. PRP rich concentrate has been shown to impact cellular and molecular processes of soft tissue. For example, Timothy Foster et al. 2009 writes the following concerning the positive effects that growth factors have on gene expression. "Several recent studies have clearly shown that PRP positively affects gene expression and matrix synthesis in tendon and tendon cells. Cell proliferation and total collagen production is increased in human tenocytes...explants cultured in PRP showed enhanced gene expression of type I collagen, types III collagen, and cartilage oligometric matrix protein... Several cytokines contained in PRP had a positive effect on muscle healing. For example, FGF [BFGF] and IGF-1 improved muscle healing in a gastrocnemius muscle laceration model in mice."22 The following growth factors are the prevalent cytokines which are released by higher concentrations of platelets as a result of PRP injections; TGF-B promotes matrix synthesis, TDGF is related to cell proliferation; IGF-1 and IGF-2 induces cellular synthesis as well as anabolic signaling; FGF causes angiogenesis and fibroblast proliferation; EGF is associated with cell proliferation; VEGF is involved with angiogenesis: and ECGF induces cell activation and angiogenesis; and fibronectin engenders cell growth. These are some of the essential growth factors which are secreted by platelets. In general, these cytokines impact many other cellular transcriptional activities, and most importantly, the nature of post-translational protein synthesis. It is likely that higher concentrations of these growth factors likewise impact

how seemingly unrelated proteins are also being translated both intra and extra-cellular. The matrix complexity of all these interactions is far too abstruse to establish a paradigm which explicates these biological processes.

Patrick Abuzei and Robert Alexander in an earlier article on PRP also showed the ameliorative molecular dynamics of platelet-rich plasma as a way to engender positive soft tissue remodeling. These medical researchers have written the following. "It appears that PDGF and TGF-B1 are among the most important growth factors in wound healing...PDGF appears to have a direct mitogenic influence on the target cells by binding to cell surface receptors and by indirectly enhancing the proliferative response of cells lacking detectable TDGF receptors...TGF-B1 is a chemotactic for macrophages and fibroblasts and is well established to be a potent stimulator of granulation tissue formation."23

Katherine St. Louis' article in the New York Times, published March 22, 2011, substantiated that there is now tremendous interest in the public and the cosmetic medical community in the use of PRP for facial rejuvenation. However, there are not any published studies aside from inflated antidotal commentary on the supposed efficacy of PRP injections into the face. Anthony Sclafari stills remains the only published investigative study employing PRP as facial filler. This particular research study involved one injection of PRP into the nasolabial fold. Conversely, we have concluded that one injection of PRP into the nasolabial fold would not be very effective in patients over 30 years of age. Furthermore, there are not any viable studies which involve a comprehensive cosmetic prophylaxis of mid and lower facial regions in the older phenotype. This study does indicate that multiple treatment sessions of PRP can have provocative aesthetic remodeling in a cosmetic manner similar to facial fillers, fat grafting strategies, and even fractionated CO2 laser protocols. We have not determined how long these results can be sustained. It is possible that there could be greater improvement over time or after stopping these treatment protocols, as the tissue structures will continue to remodel in a catabolic fashion since they are not being titrated with high concentrations of growth factors to modulate cellular transcriptional processes.

We have found that all patients with multiple PRP treatment session exhibit a more robust and renewed epidermal texture consistent with a younger phenotype. Most subjects showed a pronounced regeneration in facial volume concordant with fat grafting or the use of fillers. All patients displayed a certain degree of skin tightening. However, we did not observe this dermatological phenomenon when the subject had inordinate skin laxity. We have concluded that multiple PRP injections is most propitious in patients between ages 33-55 who do not exhibit the effects of excessive sun damage or skin laxity.

In our view, our PRP facial prophylaxis or treatment protocols established in this study is an efficacious strategy for inducing overall skin tightening, alleviation of rhytides, the enhancement of facial volume, as well as improving the epidermal texture. We believe for certain phenotypes this cosmetic procedure could be utilized in lieu of a more invasive surgical protocol. We have not determined if PRP cosmetic intervention can be sustained over a year. It has been discussed in the introduction of this study that most of these studies concerning PRP therapy's effectiveness is related to musculoskeletal perturbation. The literature does indicate that these medicinal benefits, in terms of soft tissue healing of tendon or ligament, are long-lasting.

In conclusion, biological modulation of soft tissue will be replacing other more highly invasive cosmetic therapies in the ensuing decades. There needs to be future studies to demonstrate that PRP injections can actually induce hair density in the scalp. It is not known which molecular pathways are activated in terms of collagen genesis from the exogenous PRP injections. In addition, there needs to be 3-D imaging to substantiate the nature of soft tissue remodeling associated with this cosmetic procedure. In our view, there is considerable benefit from multiple treatment sessions of PRP as a facial aesthetic therapy.

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