Improvement in the Youthfulness of Facial Skin after a Single Treatment with Platelet Rich Plasma

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Platelet-Rich Plasma for Facial Rejuvenation

The single treatment of **Platelet rich plasma (PRP)** injection demonstrated the safety profile and effectiveness for facial rejuvenation.

Significant improvement in skin elasticity was observed at 1 and 2 weeks after treatment. Our subjects perceived themselves to appear younger than their actual age, and reported about decreased wrinkles, refined pores, whitened skin, skin softening, hydration, and a decrease in post inflammatory hyperpigmentation from acne.

40 Thai patients (female 97.5%) Fitzpatrick skin type 3–4 **Average age: 36.4 years.**

Every subjects were received a single PRP facial injection (intradermally) at 30 locations (volume of 0.1 mL per location).

The outcomes were assessed by using the patient-perceived age visual analog scale (VAS), FACE-Q assessment, and elasticity index (EI)

After treatment, the patient-perceived age VAS significantly decreased (patients feeling that they look 2.71 years younger than their actual age), FACE-Q assessment for the satisfaction with the skin, and the outcome were high (74.29 and 73.41 points respectively).

feeling **2.71** years younger

The skin elasticity was significantly improved at the first two week after treatment.
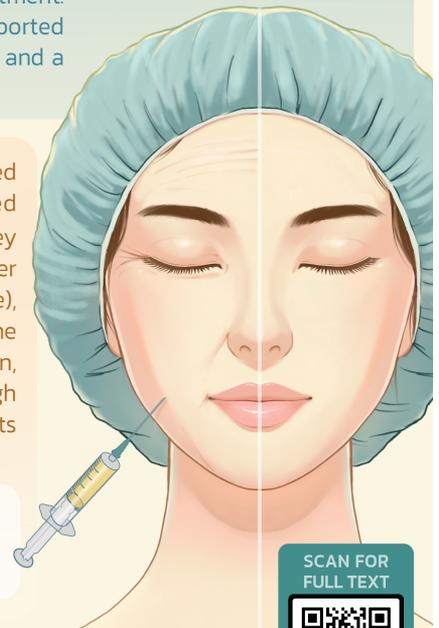
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ABSTRACT

Objective: Platelet rich plasma (PRP) is an autologous substance widely used to stimulate dermal collagen synthesis. This study aimed to investigate the efficacy of a single PRP treatment for facial rejuvenation.

Materials and Methods: This study was conducted at the Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, Thailand from August 2020 to March 2021. Enrolled patients underwent a single intradermal injection treatment of 0.1 mL PRP at 30 locations on the face. Outcomes were assessed at 1, 2, and 4 weeks post-intervention using the patient-perceived age visual analog scale (VAS), FACE-Q assessment, and elasticity index (EI) with a Cutometer.

Results: Forty Thai patients with an average age of 36.4 years, Fitzpatrick skin type 3-4 were enrolled. The mean Patient-perceived Age VAS significantly decreased one month after treatment (-2.71±2.42, $p<0.001$). FACE-Q satisfaction with facial skin was 74.29±14.49, and satisfaction with the outcome was 73.41±16.26 (scale 0-100). The PRP significantly affected skin EI, with the peak improvement observed at 1-2 weeks post-injection, predominantly at the cheek, crow's feet, and perioral area (all $p<0.05$). All patients had an overall improvement in satisfaction scores throughout the study.

Conclusion: A single treatment of PRP injection resulted in overall satisfaction in facial skin rejuvenation as measured by patient-perceived age VAS and FACE-Q assessment. The PRP showed positive outcomes in EI with peak performance at 1-2 weeks post-treatment.

Keywords: Youthfulness; platelet rich plasma; facial rejuvenation; aging face; improvement (Siriraj Med J 2024; 76: 781-788)

INTRODUCTION

Aging of the face is a multifactorial process influenced by both endogenous and exogenous factors. Endogenous factors are modified by individual genetic variation and epigenetics. Ultraviolet (UV) rays significantly contribute to dermal collagen degradation via enzymatic activation of matrix metalloproteinase (MMP).[1-5] Several studies have explored the prevention and treatment of skin aging using platelet rich plasma (PRP). Cho, et al. reported a significant reduction of UVB-induced wrinkles in nude mice following PRP injection, which increased collagen and dermal fibroblasts.[6]

Skin aging involves atrophy and laxity of the skin, a decrease in the number of fibroblasts, and a decrease in collagen production. Hence, treatments that stimulate fibroblasts play an important role in skin rejuvenation.[7] Autologous PRP is centrifuged plasma with a much higher concentration of platelets. The normal range of platelet concentration in plasma is 150,000-400,000/microliter, and this concentration is increased 4-7-fold in PRP.[3, 8-12]

PRP contains several growth factors, cytokines, and chemokines in the α-granules of platelets that can stimulate various types of cells, such as dermal fibroblasts, myofibroblasts, and epidermal cells. Growth factors, such as epidermal growth factor (EGF), fibroblast growth factor (FGF), platelet-derived growth factor (PDGF), transforming growth factor beta (TGF-β), and vascular endothelial growth factor (VEGF), can synchronously promote dermal collagen production.[13-19] (Table 1)

This study aimed to investigate the efficacy of a single PRP injection treatment for the rejuvenation of facial skin.

MATERIALS AND METHODS

This study was conducted at the Division of Plastic Surgery, Department of Surgery, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, Thailand. Adult patients were enrolled from August 1, 2020, to March 31, 2021, to undergo PRP facial injection by a single physician (NA). Patients with a history of connective tissue disease, facial palsy, coagulopathy, conditions affecting wound healing, pregnancy, or active facial skin infection were excluded. Patient demographics, comorbidities, and history of facial skin surgery and treatment were collected and recorded. Additionally, details of the PRP, skin elasticity index, FACE-Q assessment scores, and adverse events were also documented. The Siriraj Institutional Review Board (SIRB) of the Faculty of Medicine Siriraj Hospital, Mahidol University approved this study (approval number 228/2564 [IRB3]), and all study patients provided written informed consent to participate.

TABLE 1. Growth factors found in α-granules of platelets[10-13]

Growth factors	Effects
Epidermal growth factor (EGF)	Fibroblast and keratinocyte proliferation, Re-epithelialization, Tissue tensile strength
Fibroblast growth factor (FGF)	Stem cell differentiation, Proliferation and activation of fibroblasts, Collagen production, Angiogenesis
Platelet-derived growth factor (PDGF)	Proliferation and chemotaxis in fibroblasts and monocytes-macrophages, Angiogenesis
Transforming growth factor-beta (TGF-β)	Proliferation and differentiation of fibroblasts, Collagen formation, Angiogenesis, Extracellular matrix deposition and remodeling, Re-epithelialization
Vascular endothelial growth factor (VEGF)	Angiogenesis
The key growth factors, found in the α-granules of platelets, can stimulate fibroblast activity and support new collagen production in the skin through various mechanisms.	

Preparation of PRP

Twenty milliliters (mL) of venous blood was collected from each patient and stored in 3.2% sodium citrate 3.0 mL sandwich tubes (VACUETTE®; Grenier Bio-one, Monroe, NC, USA). Two rounds of centrifugation were performed sequentially. The first round aimed to separate the blood cells from plasma by centrifugation at 1,500 revolutions per minute (rpm) for 10 minutes. The supernatant was then transferred to new citrate tubes. In the second round, the new plasma tubes were centrifuged at 3,000 rpm for 10 minutes to separate the platelets from the plasma. After the second round of centrifugation, the upper 70% of the supernatant was removed, leaving the platelet pellets. The platelet pellets were then mixed with the remaining 30% of the supernatant located at the bottom of the tube to create PRP for facial injection (Fig 1).
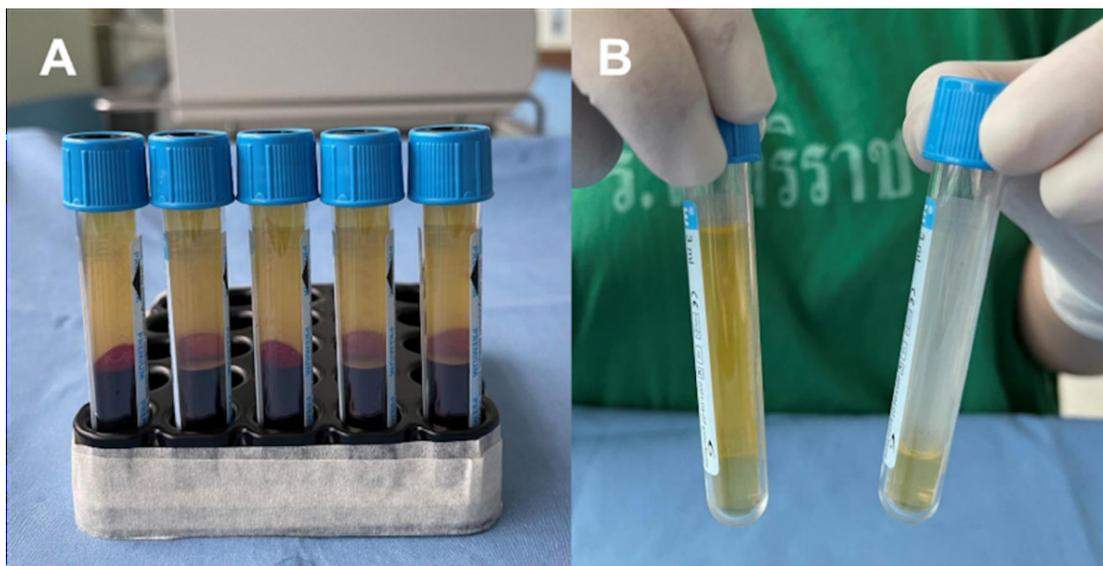
Fig 1. (A) Centrifuged venous blood after the first round of centrifugation at 1,500 rpm for 10 minutes. The supernatant from the first centrifugation was transferred to new citrate tubes. (B) The second centrifugation was performed at 3,000 rpm for 10 minutes. After completion of the second round of centrifugation, the upper 75% of the supernatant was removed and platelet pellets were produced. The platelet pellets were then mixed with the remaining 25% of the supernatant located at the bottom of the tube to create PRP for facial injection.

PRP facial injection

After the facial skin was cleaned, topical anesthesia (EMLA®; AstraZeneca LP, Wilmington, DE, USA) was applied for 60 minutes before the start of PRP injections. A volume of 0.1 mL of PRP was injected intradermally at 30 locations on the face, for a total PRP dose of 3.0 mL per patient (Fig 2). After treatment, the patient was observed in the recovery room for 15 minutes and discharged home if no adverse reactions were observed.

Outcome measurements

The elasticity index (EI) of the skin was measured using the Cutometer Dual MPA580® (Courage & Khazaka GmbH, Cologne, Germany) before and at 1, 2, and 4 weeks after PRP treatment. Ten surrogate reference points were assigned bilaterally at the forehead, lower eyelid, crow's feet, cheek, and perioral region.

The forehead region was measured at 1 cm above the brow ridge in the mid-pupillary line. The lower eyelid region was measured at 1 cm below the lower eyelid rim in the mid-pupillary line. The crow's feet region was measured at 1 cm lateral to the lateral canthus. The cheek area was measured at 5 cm below the lower eyelid rim in the mid-pupillary line. Finally, the perioral region was measured at 1 cm above Cupid's bow and the mid-philtrum line, and at 1 cm below the lower lip border and mid-philtrum line (Fig 3).

For the follow-up evaluation protocol, subjects cleansed their face and acclimatized for 15 minutes at room temperature, ranging from 21°C to 25°C, and a humidity level of 31% to 50%. R2 and R7 parameters were used as the main parameters to assess skin elasticity and aging. The R2 parameter represents the gross elasticity of the skin, and the R7 parameter represents biological elasticity, which is the ratio of immediate retraction to total distension.[20]

Study patients completed the FACE-Q assessment questionnaire, a validated patient-reported outcome measurement, one month after PRP treatment. The subscales used in this study included FACE-Q satisfaction with skin, FACE-Q satisfaction with outcome, and patient-perceived age visual analog scale (VAS). The first two subscales are scored from 0-100, with higher scores reflecting higher satisfaction. The age VAS was reported in years, with a more negative value indicating that the respondent feels they look younger than their actual age.[21-23]

Statistical analysis

Demographic data, elasticity index, and FACE-Q results were presented using descriptive statistics, including number and percentage, mean ± standard deviation, and range. Differences in the elasticity index at each time point and the decrease in patient-perceived age VAS were calculated using a paired *t*-test from SPSS Statistics for Windows, version 26.0 (SPSS, Inc., Chicago, IL, USA). A *p*-value lower than 0.05 was considered statistically significant.

RESULTS

Forty Thai patients aged from 22 to 53 years (mean: 36.4 years) were enrolled. The patients had Fitzpatrick skin type 3-4. Most patients were female (97.5%, 39 of 40). Patients were available for cutometer evaluation in 67.5% (27 of 40) of cases. Significant improvement in skin elasticity (as measured by elasticity index [EI])

Fig 2. The 0.1 mL of PRP was injected into the intradermal plane at 30 points on the face (purple crosses).

Fig 3. Reference points for elasticity index measurement by cutometer. Ten surrogate reference points (blue dots) were assigned bilaterally at the forehead, lower eyelids, crow's feet, cheeks, and perioral regions.

was observed at 1 and 2 weeks after PRP treatment in the crow's feet, perioral, and cheek area (all $p<0.05$). The effects of PRP had an impact on the gross and biological elasticity of the cheek area. However, the effect on the crow's feet only affected biological elasticity at 1-week post-treatment and can be seen through gross elasticity for the perioral area at 2 weeks post-treatment. These significant improvements were not sustained at 4 weeks after PRP treatment. The EI values in other areas of the face did not show significant differences between baseline and after treatment (Table 2).

Regarding patient satisfaction, the FACE-Q assessment for satisfaction with the skin was 74.29±14.49 points, and satisfaction with the outcome was 73.41±16.26 points, both measured one month after receiving a single PRP treatment. Subjects reported significant improvement in the patient-perceived age visual analog scale one month after treatment compared to baseline (-2.71±2.42 years *vs.* -0.05±2.87 years, respectively; $p<0.01$). This translates to patients feeling that they look 2.71 years younger than their actual age (Table 3).

At one week after PRP treatment, 26.5% of patients reported a decrease in wrinkles, and 23.5% noted refined pores and whitened skin. At two weeks after PRP treatment, all patients reported perceiving the rejuvenating effects of PRP. The number of patients experiencing skin whitening, decreased wrinkles, hydrated skin, and softening of the skin had further increased at four weeks after treatment compared to two weeks after treatment (Table 4). The number of patients who reported skin whitening continuously increased to 40% by 4 weeks after treatment (Fig 4). Additionally, some patients reported a reduction in post-inflammatory hyperpigmentation (PIH) from acne.

DISCUSSION

PRP rejuvenates the skin by stimulating DNA synthesis, delaying cell apoptosis, and promoting the expression of genes associated with tissue regeneration. Kakudo, *et al.* proposed that PDGF and TGF-β contribute to the proliferation of adipose-derived stem cells and dermal fibroblasts.[24] Several studies reported the efficacy of PRP for skin rejuvenation with satisfactory outcomes and other clinical applications Since autologous PRP is harvested from the patient receiving the injections, it carries a low risk for allergic reactions and blood-related infection transmission.[25]

Significant improvement in skin elasticity was observed at 1 and 2 weeks after PRP treatment compared to baseline measurements (cheek, crow's feet, and perioral area [$p<0.05$]). However, these effects appear transient as the significant improvement was not sustained at 4

weeks after treatment. Further studies should explore the potentially cumulative effect of two or more sessions of PRP injections to determine if they have long-lasting effects and significantly improved outcomes.

Maisel-Campbell, *et al.* conducted a systematic review of the effects of PRP on skin aging. The review demonstrated that PRP can temporarily improve facial skin appearance, texture, lines, and pigmentation.[26] However, the review included 24 studies and was limited by variations in PRP preparation, follow-up evaluations, and the lengthy evaluation period of over 4 weeks. Our study conducted follow-up evaluations at 1, 2, and 4 weeks intervals after a single PRP treatment, using FACE-Q, VAS, and EI for a more precise evaluation at each visit. This approach allowed the research to observe the dynamic effects of PRP progression, which had not been thoroughly discussed in previous studies.

Despite the lack of significant differences in many elasticity indices, most participants reported being satisfied with the results. FACE-Q scores for satisfaction with skin and satisfaction with outcome were both high one month after a single PRP treatment. Subjects perceived themselves to appear 2.7 years younger than their actual age. These subjective outcomes indicate the rejuvenating effects of PRP. Moreover, our patients reported decreased wrinkles, refined pores, and whitened skin in just one week after PRP treatment. The number of patients experiencing satisfactory results increased by the four-week mark. Improvements were also reported in skin softening, hydration, and a decrease in PIH from acne.

Limitations

Many participants reported improvement in periorbital darkness, suggesting that further objective measurement of this parameter should be considered. The drawback of the study was that the number of participants eligible for cutometer measurement was lower than anticipated due to limitations and restrictions caused by the COVID-19 pandemic.[27] If more study patients had undergone elasticity index evaluation, it is possible that the analysis would have yielded more statistically significant results.

CONCLUSION

A single treatment of platelet rich plasma (PRP) injection demonstrated the safety profile and effectiveness for rejuvenating facial skin. This is evidenced by a marked reduction in the Patient-Perceived Age Visual Analog Scale (VAS), enhancements in various facial skin parameters, and significant improvements in the elasticity index outcomes.

TABLE 2. *P*-values from paired samples *t*-test of the elasticity index from Cutometer for each area of the face at different time points compared to baseline.

Area	Evaluation parameter	Time point	Mean	SD	P-value
Forehead	R2 (Ua/Uf)	Pre-treatment	0.4644	0.2914	
		1 week	0.5867	0.2384	0.526
		2 weeks	0.5938	0.1808	0.308
		4 weeks	0.7128	0.0865	0.920
	R7 (Ua/Uf)	Pre-treatment	0.2240	0.1649	
		1 week	0.2171	0.1481	0.315
		2 weeks	0.3054	0.1725	0.392
		4 weeks	0.1369	0.0853	0.072
Lower eyelid	R2 (Ua/Uf)	Pre-treatment	0.4688	0.2636	
		1 week	0.5484	0.1927	0.835
		2 weeks	0.5084	0.1820	0.788
		4 weeks	0.6875	0.1694	0.833
	R7 (Ua/Uf)	Pre-treatment	0.2169	0.1124	
		1 week	0.2054	0.1715	0.753
		2 weeks	0.2981	0.2019	0.232
		4 weeks	0.2086	0.1780	0.653
Crow's feet	R2 (Ua/Uf)	Pre-treatment	0.4334	0.2375	
		1 week	0.5447	0.2007	0.277
		2 weeks	0.4939	0.2095	0.718
		4 weeks	0.7040	0.0929	0.113
	R7 (Ua/Uf)	Pre-treatment	0.1996	0.1220	
		1 week	0.2803	0.1161	**0.035***
		2 weeks	0.2811	0.1438	0.104
		4 weeks	0.2664	0.1971	0.674
Cheek	R2 (Ua/Uf)	Pre-treatment	0.4522	0.2145	
		1 week	0.5851	0.2030	**0.018***
		2 weeks	0.5746	0.2051	0.087
		4 weeks	0.7273	0.1158	0.227
	R7 (Ua/Uf)	Pre-treatment	0.2503	0.1352	
		1 week	0.2546	0.1389	0.861
		2 weeks	0.3586	0.2035	**0.017***
		4 weeks	0.2406	0.2195	0.787
Perioral	R2 (Ua/Uf)	Pre-treatment	0.4367	0.2392	
		1 week	0.5160	0.2085	0.623
		2 weeks	0.5176	0.2023	**0.034***
		4 weeks	0.6018	0.2098	0.780
	R7 (Ua/Uf)	Pre-treatment	0.2302	0.1902	
		1 week	0.2244	0.1380	0.995
		2 weeks	0.2968	0.2242	0.171
		4 weeks	0.1278	0.0805	0.429

The paired T-test was done by comparing each parameter at each time point to the baseline values before treatment. *A p-value less than 0.05 was considered statistically significant.

TABLE 3. Mean FACE-Q assessment scoring.

| FACE-Q scale	Mean ± S.D.	
FACE-Q satisfaction with skin	74.29±14.49	
FACE-Q satisfaction with outcome	73.41±16.26	
Patient-Perceived Age VAS		**p-value**
Before treatment	-0.05±2.87	
One month after treatment	-2.71±2.42	**<0.001***

The FACE-Q satisfaction with skin and satisfaction with outcome scores range from 0-100. The Patient-Perceived Age VAS statistically significantly decreased after 1 month after a single PRP treatment (p<0.001).
Abbreviation: VAS, visual analog scale.

TABLE 4. Subjective patient-reported perceptions after PRP treatment.

Effects	1st week	2nd week	4th week
Decreased wrinkles	26.5%	25.8%	33.3%
Refined pores	23.5%	29.0%	26.7%
Whitening of skin	23.5%	29.0%	40.0%
Softening of skin	20.6%	19.4%	20.0%
Hydrated skin	20.6%	19.4%	20.0%
Decreased PIH after acne	17.6%	22.6%	20.0%
No change	8.8%	0.0%	0.0%

Abbreviation: PIH, Post-inflammatory hyperpigmentation.
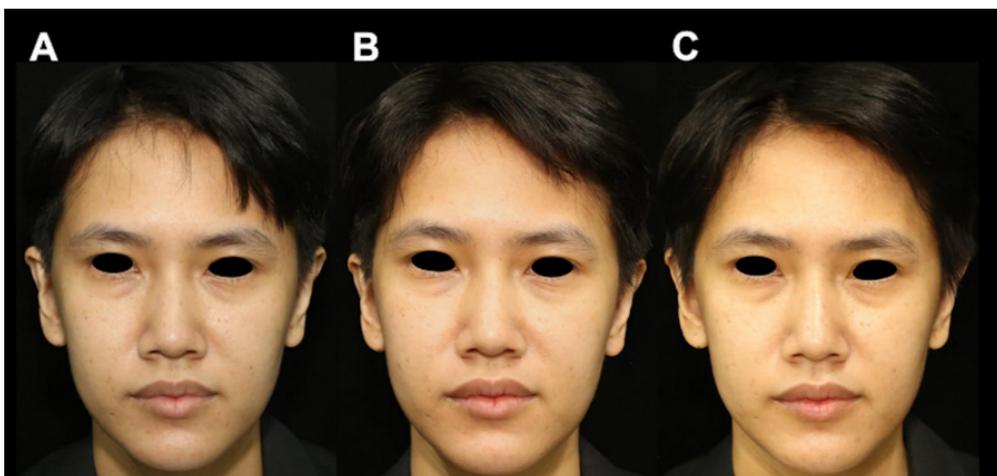
Fig 4. The patient initially presented with post-inflammatory hyperpigmentation (PIH) from acne, fine wrinkles, and periorbital darkness. The results of PRP injection were compared between (A) pre-treatment, (B) two weeks post-treatment, and (C) four weeks post-treatment.

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