



## A Comparative Study of the Efficacy of PRP Prepared Using Double Spin Technique versus PRP Prepared Using Turn Down – Turn Up (TD-TU) Technique in Combination with Microneedling in the Treatment of Atrophic Acne Scars

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### Abstract

**Background:** Acne scarring has a huge impact on patient's life. Platelet rich plasma (PRP) has been widely used as an adjunct to other procedures like microneedling in the treatment of acne scars. Aim: The aim of the study is to compare the efficacy of microneedling with PRP prepared using double spin technique and microneedling with PRP prepared using Turn down- Turn up technique in treating atrophic post acne scars. **Methods:** A total of 20 patients suffering from atrophic acne scars were included in the study and randomly divided into two groups of 10 each. Group A were treated with microneedling and PRP prepared using Double-spin technique and in Group B patients were treated with microneedling and PRP prepared using Turn down-Turn up technique (TD-TU). A total of 2 sittings were done at 4 weeks interval and the final readings were assessed after 8 weeks. The improvement in scars were evaluated using Goodman and Baron's scale, ECCA scale, Scar quartile gradient and dermatology life quality index scores. Results were analysed using Mann-Whitney test and Wilcoxon signed rank test. **Results:** Significant improvement in the acne scars was noted in both the groups. **Conclusion:** Microneedling in combination with PRP prepared using Turn Down- Turn up technique produces appreciable results comparable to PRP prepared using double-spin technique in the management of atrophic acne scars.

**Keywords:** Atrophic Acne Scars; Microneedling; Platelet Rich Plasma

### Introduction

Acne is a common condition experienced by most of the people especially in the age group of 15 to 30 years, though rarely occurring in older adults. This is of concern as in most of the cases, the severe inflammatory response to Propionibacterium acnes results in permanent, disfiguring scars, mostly appearing on the face. It has been observed that a vast majority of patients suffering from acne develop some degree of facial scarring, thereby having a psychosocial impact on its sufferers (Baldwin, 2002; Layton, Henderson & Cunliffe, 1994).

Though over the past several decades, various innovative techniques have been suggested to reduce the appearance of scarring in those patients suffering from acne (Fabbrocini *et al.*, 2009), the advances in science and technology have paved way for numerous innovative techniques, especially in the field of health and medicine which not only address the clinical situation but also provide a relief from the

cosmetic discomfort. Platelet-rich plasma with microneedling is one such technique which has gained significant importance in acne scar removal.

Microneedling is a procedure done to create micro-puncture in the dermis, which releases various growth factors resulting in collagen synthesis leading to levelling up of atrophic scars (Fabbrocini *et al.*, 2009; Fernandes, 2002). Microneedling has been used in combination with many other treatment modalities such as chemical peels, radio frequency, subcision, PRP, etc (Sharad, 2011; Gadkari & Nayak, 2014).

“Platelet-rich plasma” which was used for the first time by Kingsley *et al.* (1954) refers to the standard platelet concentrate for transfusion. PRP which is obtained from blood contains high concentrations of platelets (Marx, 2001) which is known to have beneficial effects in treatment of skin ulcers, skin rejuvenation and post acne scars. It improves dermal regeneration and acts locally to promote protein and collagen synthesis, which is responsible for enhancing the wound-healing response and reducing the scars (Fabbrocini *et al.*, 2009; Fernandes, 2002).

Since both Microneedling and PRP releases various growth factors, the combination of these two techniques enhances the effects to give better cosmetic results. Various studies have tried these two modalities and it was found that better results were obtained treatment of atrophic acne scars (Gupta, Barman & Sarkar, 2021).

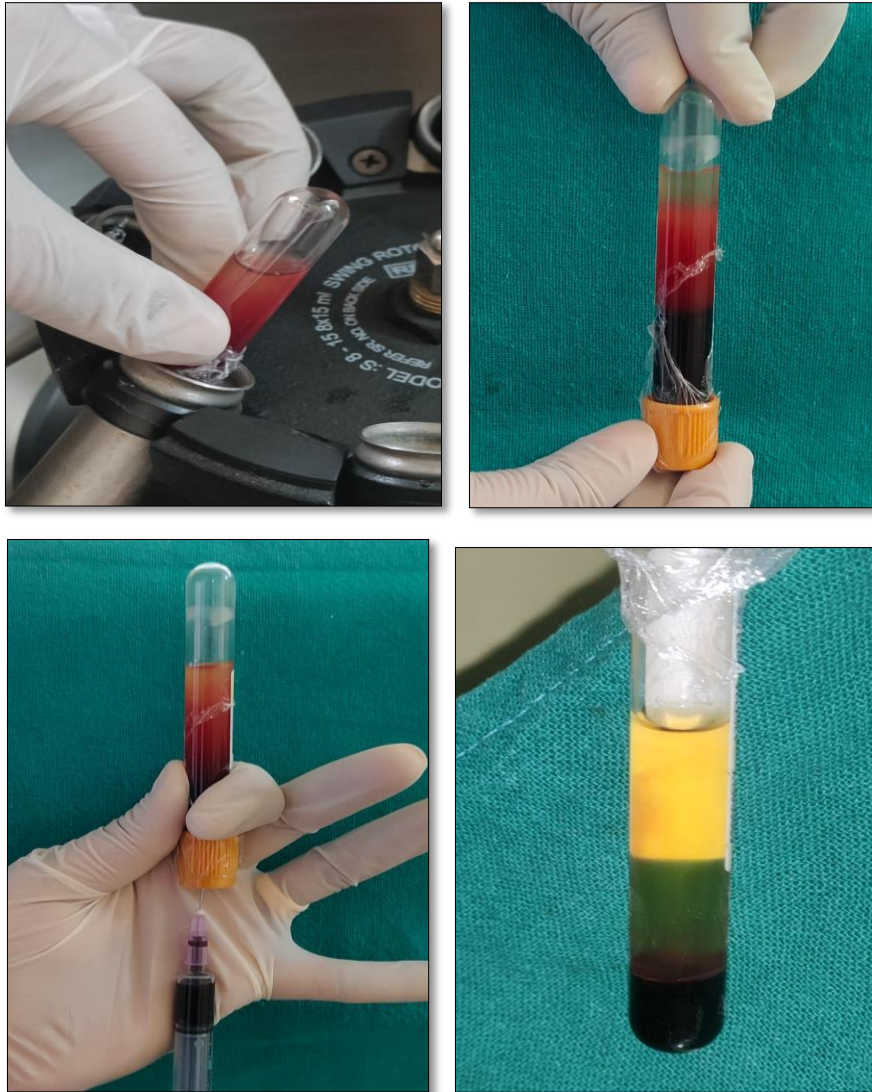
Several protocols for PRP preparation are existing in the literature. It has been shown that PRP preparation as described by Chawla, 2021 using double spin technique yields higher platelet concentrations. Machado *et al.* in 2019 described the procedure for PRP preparation using the 'Turn down-Turn up' technique (TD-TU), which follows the low-speed centrifugation concept (LSCC) to preserve the maximum volume of platelets and growth factors (Choukroun & Ghanaati, 2018). This technique offers the added advantage of low-cost preparation and at the same time reduces the contamination risk, as the blood remains in the same tube during the entire process (Machado *et al.*, 2019).

In this present study our aim was to compare the efficacy of microneedling with PRP prepared using Turn down-Turn up technique and microneedling with PRP prepared using double spin technique in treating atrophic post acne scars.

### **Methodology**

The study was conducted for a period of 8 weeks. The inclusion criteria included patients aged between 18 and 45 years with acne scars of grades II, III and IV according to the Goodman and Baron's grading scale. Patients with active acne, tendency of keloid formation, history of bleeding disorder, patients on oral steroid therapy, Lignocaine hypersensitivity, Pregnant and lactating mothers were excluded from the study. Patients were divided randomly into 2 groups. Group A were treated using microneedling with PRP prepared using Double spin technique and Group B were treated using microneedling with PRP prepared using Turn down- Turn up technique.

The area of interest was cleansed with an alcohol swab and anesthetised using a thick layer of topical anaesthetic cream (a combination of prilocaine and lignocaine) for about 30-45 minutes before the procedure. 10 ml of whole blood was collected and PRP was prepared either by Double spin or by Turn down – Turn up Technique. Preparation of PRP using double spin technique (Chawla, 2014) included an initial spin which was performed at 1500 rpm for 10 min and the second spin was performed at 3700 rpm for 10 min at room temperature (Figure 1).



**Figure 1: TD-TU Protocol (A) Tube Inserted into the Centrifuge with Cap Facing Down (B) After First Spin (C) Aspiration of the Hematic Layer with Cap Facing Down (D) Final Appearance of Tube After Second Spin**

PRP which was prepared using Turn Down-Turn Up Protocol (Machado *et al.*, 2019) included a first spin which was performed at 200 g for 15mins with the tube cap facing down. The tube was then carefully removed from the centrifuge maintaining the downward position without turning the tube. 3.5 ml of the hematic layer is aspirated under aseptic conditions, through the rubber cap and the tube was turned to an upright position (i.e. tube cap facing up). It was again centrifuged at 1600 g for 10 minutes in the upright position with the tube cap facing up. Under 3.5 ml of the superior portion of the material (platelet-poor plasma) was aspirated. The desired amount of PRP (1-2 ml) from the lower portion of the tube was aspirated as in Figure1. The prepared PRP was injected intradermally through a 30 Gauge needle (insulin syringe) deep to each scar on the affected areas. The amount of PRP injected was sufficient to elevate the scar and the end point was taken as blanching and elevation of the scar. After injecting, the site was treated with microneedling using Dermapen device with 36 pin needle cartridges in circular motions for about 4-5 minutes (Nandini *et al.*, 2021). A needle length of 1.5-2.0mm was used in different areas of the face. The length of needles was adjusted according to the severity of the concern, area being treated, the person's discomfort, tolerance and response (Singh & Yadav, 2016).

The acne scars were graded using Goodman and Baron Qualitative (Goodman & Baron, 2006), Quantitative scale and ECCA scale (Goodman & Baron, 2006; Dreno *et al.*, 2007). The degree of

improvement in the acne scars were graded using Scar quartile grading scale (SQGS) (Elsaie, Ibrahim & Saudi, 2018). Response was recorded for Patient's Quality of Life using Dermatology Life Quality Index Questionnaire (DLQI) (Finlay & Khan, 1994). All the scores were graded at baseline and 8 weeks. Digital Photographs using identical camera settings and lighting conditions at 3x magnification were taken at baseline and 8 weeks.

The compiled data were analysed using Wilcoxon signed rank test and Mann-Whitney test,  $p < 0.005$  was considered statistically significant.

### **Ethical Consideration**

This study was approved by I.T.S Institutional Ethics Committee (IIEC), India with registration number: ITSCDSR/IIEC/LD/PERIO/2022-25/002 on September 22<sup>nd</sup>, 2023.

### **Result**

All 20 patients completed the study. Figure 2 and Figure 3 shows the before and after results after microneedling and PRP using Double spin protocol and TD-TU protocol respectively.



**Figure 2: Patient with Atrophic Acne Scars Before and After (8 Weeks) Microneedling with PRP Using Double-Spin Protocol**



**Figure 3: Right Side of the Face Before and After 8 Weeks Microneedling with PRP Using TD-TU Protocol**

The mean age of the participants in double-spin protocol and turn up-turn down protocol groups were  $31.30 \pm 6.60$  and  $28.30 \pm 6.78$  years respectively. There were four males and six females in each group. The demographic profile of the studied individuals is given in Table 1.

**Table 1: Demographic Details of the Study Participants**

Variable	Category	Double spin protocol (n=10)	Turn up-Turn down protocol (n=10)
Age		31.30 ± 6.60	28.30 ± 6.78
Gender	Male	4 (40%)	4 (40%)
	Female	6 (60%)	6 (60%)

The intragroup comparison using Goodman Baron Qualitative scar grading scale showed that even though the patients showed improvement, there was no statistical significance from T1 to T2 in both the groups with a p-value of 0.072 in the Double spin and 0.346 for the TD-TU group respectively. Table 2 Intergroup comparison using Mann-Whitney test, showed that there was a non-significant difference in the Goodman & Baron Qualitative Score between the two groups at T1 interval ( $p = 0.91$ ) and T2 interval ( $p = 0.63$ ).

**Table 2: Intragroup Comparison of the Variables - Goodman & Baron Qualitative Score**

Protocol	Interval	Grade 1	Grade 2	Grade 3	Grade 4	Median	p-value
Double Spin	T1	0 (0%)	7 (70%)	0 (0%)	3 (30%)	2	0.072
	T2	2 (20%)	5 (50%)	2 (20%)	1 (10%)	2	
TD-TU	T1	0 (0%)	6 (60%)	2 (20%)	2 (20%)	2	0.346
	T2	0 (0%)	7 (70%)	2 (20%)	1 (10%)	2	

Wilcoxon Signed Rank Test

The intragroup comparison using Goodman Baron Quantitative scar grading scale showed that the mean score for double-spin group was  $9.30 \pm 6.95$  and  $7.70 \pm 5.62$  at T1 and T2 respectively. On intragroup comparison, there was statistically significant decrease in the scores from baseline to 8 weeks ( $p = 0.026$ ).

Similarly, the mean score for TD-TU group was  $10.10 \pm 5.51$  and  $7.90 \pm 4.91$  at T1 and T2 respectively. On intragroup comparison, there was statistically significant decrease in the scores from baseline to 8 weeks ( $p = 0.011$ ) Table 3.

However, on Intergroup comparison, the mean scores were statistically non-significant between both groups at T1 ( $p = 0.436$ ) and T2 (0.796).

**Table 3: Intragroup Comparison of the Variables - Goodman & Baron Quantitative Score**

Variable	Protocol	T1		T2		p-value
		Mean	SD	Mean	SD	
Goodman & Baron Quantitative Score	Double Spin	9.30	6.95	7.70	5.62	0.026*
	TD-TU	10.10	5.51	7.90	4.91	0.011*

Wilcoxon signed rank test; \*Indicates a significant difference at  $p \leq 0.05$

Using the ECCA grading, on intergroup comparison, the mean scores were statistically insignificant between both the double spin and TD-TU groups at baseline ( $p = 0.579$ ) and 8 weeks ( $p = 0.739$ ).

Another assessment parameter used was Scar quartile grading scale. In the Double spin protocol group, out of the 10 patients 4 patients showed no improvement, 3 patients showed > 25% improvement and 3 patients showed 26-50% improvement whereas in the Turn down-Turn up protocol group, 2 patients showed no improvement, 5 patients showed > 25% improvement and 3 patients showed 26-50% improvement. The differences between the 2 groups were statistically not significant ( $p = 0.631$ ) (Table 4).

**Table 4: Intergroup Comparison of Scar Quartile Grading Score**

Protocol	Grade 0	Grade 1	Grade 2	Grade 3	p-value
Double Spin	4 (40%)	3 (30%)	3 (30%)	0 (0%)	0.631
TD-TU	2 (20%)	5 (50%)	3 (30%)	0 (0%)	

Mann Whitney Test

The interpretation of DLQI scores was performed for all the patients. The DLQI scores ranged from 3 to 14 with a mean score of  $2.9 \pm 0.88$  for the double spin group and  $2.50 \pm 0.85$  for the Turn down-Turn up protocol group at baseline which reduced to  $2.20 \pm 0.79$  for the Double spin group and  $2.20 \pm 0.63$  for the Turn down-Turn up protocol group respectively at 8 weeks. The p values obtained was 0.034 and 0.083 for the double spin group and Turn down-Turn up group respectively which was statistically significant for the Double spin protocol as given in Table 5

**Table 5: Intragroup Comparison of the Variables – DLQI Score**

Variable	Protocol	T1		T2		p-value
		Mean	SD	Mean	SD	
DLQI Score	Double Spin	2.90	0.88	2.20	0.79	0.034*
	TD-TU	2.50	0.85	2.20	0.63	0.083

Wilcoxon signed rank test; \*Indicates a significant difference at  $p \leq 0.05$

## Discussion

In the present study, 20 patients presenting with Goodman and Baron Grade II, III and IV atrophic acne scars were treated with a combination of microneedling with PRP. This study aimed to compare the effectiveness of microneedling with PRP prepared by using two different protocols namely the “Double-spin” protocol (Chawla, 2014) and “Turn down- Turn up” protocol (Machado *et al.*, 2019) and determined its influence on the treatment response, as both protocols have shown to yield higher platelet concentrations and growth factors as per reviewed literature. To the best of our knowledge, there are no reported literature comparing the responses using these two protocols. In our study all the patients having scars of varying duration showed significant response to PRP with microneedling in both the groups.

Acne vulgaris is a common physiologic skin condition eventually resulting in scarring in a significant proportion of the population. In a survey among 4000 patients in a French population Poli, Dreno and Verschoore (2001) found that 49% of the total patients with acne had acne sequelae resulting in acne scars and/or pigmented macules. Layton, Henderson and Cunliffe (1994) found that even though all types of acne are associated with scarring, the nodulocystic acne are more commonly associated with acne scars and the most common site for these scars is the face followed by the trunk region.

Microneedling process which involves repetitive skin punctures using sterile micro needles has been proved as an effective treatment for the management of acne scars (El-Domyati, Abdel-Wahab & Hossam, 2018). Platelet-rich plasma (PRP) have been used clinically in humans for its healing properties attributed to the presence of growth factors. Fabbrocini *et al* (1994) in their study found that PRP combined with microneedling was more effective in the treatment of atrophic acne scars than using microneedling alone.

In the present study, PRP with Microneedling was well tolerated in both the groups with only mild erythema which lasted for a day or two and all the patients having scars of varying duration showed appreciable response to PRP with microneedling which is in accordance with the study by Dogra, Yadav and Sarangal (2014). The established Goodman and Baron’s grading were chosen for analysis of the present study, as it encompasses all the various types of post acne scars and uses simple clinical examination as a tool for the grading of the acne scars.

Patients were assessed twice once at baseline level (T1) and once after 8 weeks (T2). The study ensured similar subject inclusions in both the groups at level T1, so that further improvement in both the groups can be compared at T2 level. The significance of the outcomes of the study was assessed by calculating the p value and considering  $p < 0.05$  as statistically significant.

The established Goodman and Baron’s qualitative score was used to study the response in the subjects in both the groups of Double spin protocol and the TD-TU protocol. Our analysis revealed that even though there was an improvement in all the patients, the intragroup and intergroup statistical analysis was insignificant. This could be because of the analysis being performed after 8 weeks in comparison to the 32 weeks reported in the previous studies.

The Goodman and Baron Quantitative grading system was based on lesion counting. In a study by Chandrashekar *et al.* (2014) using the Goodman and Baron's grading system revealed moderate improvement in 58% of the patients, 29% minimal improvement, good improvement in 9%, and very good improvement in 3% of the patients. Although all the patients in our study reported improvement in the scores from T1 to T2, the analysis did not reveal statistical significance in both the groups as almost 90% had minimal reduction in the Double Spin group and all the 100% showed minimal reduction in the TD-TU group.

The intragroup comparison of the Goodman Baron quantitative score reduced from 9.3 to 7.7 in group using double spin protocol and from 10.10 to 7.9 in the other group using Turn up - Turn down protocol which revealed that there was a significant improvement in both the groups, but the difference of the improvement between the two modalities was statistically insignificant. The appreciable improvement in the subjects belonging to both the group could be attributed to higher platelet concentrations in PRP, used with microneedling.

The analysis of the results of the present study using ECCA scale revealed that the combined therapy of Microneedling with PRP gives better results and is better tolerated in all types of atrophic acne scars, which was concurring with results reported by Ismail, Khella & Abou-Taleb (2022). Even though both the protocols exhibited statistically significant improvement ( $p = 0.066$  and  $0.034$  for Double spin and TD-TU protocol respectively) in the degree of scar severity before and after treatment, there was an insignificant difference between both the protocols denoting that both protocols yielded the same net results. Gupta *et al.* (2021) in their study revealed that even though there was a reduction in the ECCA scores, it was not statistically significant with a  $p$  value of 0.058.

Amer, Elhairy and Al-Balat (2021) in their study assessed the scars of 41 patients using the Scar Quartile grading where fourteen patients (34.1%) had mild response to the treatment, seventeen patients (41.5) had good response, two patients (4.9%) each had very good and excellent. In our study the statistical analysis of " $p$ " value as 0.631, which reveals insignificant difference between the responses in both the groups, it is seen from the analysis that the percentage of subjects showing "no improvement" has significantly reduced in the group following the "Turn down-Turn up" protocol which could be attributed to the possibility of preservation of the maximum volume of platelets and growth factors due to low speed centrifugation.

Hayashi, Miyachi and Kawashima (2015) observed scarring in 90.8% cases with acne and suggested that acne scars had a negative impact on patient's QoL. In a similar study conducted by Hazarika and Rajaprabha (2016), the DLQI scores showed a large effect on 13 out of 29 cases with severe acne scars. A study by Porwal, Chahar and Singh (2018) in his study indicated that acne scars had a very large effect on patient's quality of life. In our study the interpretation of DLQI scores was performed for all the patients belonging to both the groups and the analysis revealed that the number of subjects expressing very large effect of impairment reduced significantly in both the groups.

The low scores obtained in the DLQI assessment from the younger age group, can be attributed to their expectation of a flawless radiant skin by them. Considering the smaller sample size, studying the differences by age or sex was a constraint. The results of the present study are comparable to the established double spin technique with non-significant difference statistically. However clinically TD-TU Technique is a reproducible and more economical method of PRP preparation.

Few limitations of the study can be a grading scale which was subjective with a very high index of observer bias, and the limited number of sessions given to the patients. Platelet count estimation and comparative analysis of patients enrolled in the study could have also been done to remove bias. The future studies should be directed to enrol a larger sample size and longer follow-up and a greater number of sessions that would probably shed more light on the outcomes of the techniques suggested in the treatment of acne scars.

## Conclusion

This study highlights that microneedling combined with PRP, whether prepared by the double-spin or the Turn Down-Turn Up (TD-TU) technique, is effective in the treatment of atrophic acne scars. Both techniques led to notable improvements in scar appearance and patient satisfaction. While the double-spin technique showed a statistically significant reduction in Dermatology Life Quality Index (DLQI) scores, indicating a positive impact on patient quality of life, both methods ultimately yielded similar aesthetic results across other metrics, such as Goodman and Baron scales, ECCA scale, and Scar Quartile Grading. Given its lower cost and reduced contamination risk, the TD-TU technique may offer an accessible alternative to the double-spin approach, particularly in resource-limited settings. Overall, the findings support the integration of microneedling with PRP into clinical practice as a viable and effective option for enhancing scar appearance and patient confidence. Further research with larger samples could deepen understanding of the comparative benefits and refine PRP preparation techniques for optimal therapeutic outcomes in acne scar management.

## Conflict of Interest

The authors declare that they have no conflicting interests.

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