Original Research Article

Comparison between efficacy of carboxytherapy, plasma gel injection and mesotherapy in treatment of periorbital dark circles

Abstract

Background: Periorbital dark circles (PODC) are also recognized as periorbital melanosis, dark eye circles, and infraorbital darkness. They are a public cosmetic skin disorder with relative darkness of the periorbital areas. This condition affects both sexes, it gets worse by aging which makes people look tired and older so it has a negative effect on the quality of life, particularly in women.

Methods: This comparative, simple randomized, controlled study was carried out on 30 patients whopresented with periorbital dark circlesand had not received any treatment 3 months before joining the study. The patients were equally allocated into three groups **Group A**: treated with bilateral injection of PRP gel, **Group B**: treated with bilateral intradermal injection of CO_2 with a carboxytherapy machine, and **GroupC**: treated with intradermal injection of vitamin C. All patients received 4 sessions with 2 weeks apart.

All patients were subjected to complete history taking, general and dermatological examination, and clinical assessment was done for all patients to determine the type of dark circles

Results: The improvement of PODC in plasma gel injection and carboxytherapy ~~mesotherapy~~ were excellent in 50%,and 40% in vitamin C group. There was a gradual improvement of PODC with time, but therecurrence rate of the studied groups after 6 months was 40%, 30%, 60% in groups A, B and C, respectively, showing a higher recurrence rate in group C patients treated with vitamin C mesotherapy.

Conclusions: Gel injection, carboxytherapy and vitamin C mesotherapy were effective and safe therapeutic modalities in thetreatment of PODC.

Keywords:carboxytherapy, plasma gel injection, mesotherapy, periorbital dark circles

Introduction:

Periorbital dark circles (PODC) are also identified as periorbital melanosis, dark eye circles, and infraorbital darkness. It is a cosmetic skin ailment characterised by relative darkening of the periorbital regions. This condition affects both sexes, it worsens with age, making people appear fatigued and older, and it has a severe impact on women's quality of life [1].

Non-surgical options for the treatment of periorbital pathologies include hyaluronic acid injections, micro-needling, chemical peels, laser-resurfacing, microdermabrasion, nifedipine, botulinum toxin, and carbon dioxide (carboxytherapy) injections. Surgical options include fat transfers, blepharoplasty, and face-lifts [2].

Although there are several therapy options for PODC, the majority of them have negative results. Platelet-rich plasma (PRP) and carboxytherapy may hold promise for face rejuvenation and treatment of PODC in aesthetic dermatology [3].

Centrifugation generates autologous platelets for platelet-rich plasma. The platelet concentration is three to five times the average plasma concentration (1). At least seven important protein growth factors are secreted by platelets to initiate wound healing. The main promise of PRP therapy is to augment the natural regeneration process by administering a concentrated dose of platelets and higher concentrations of growth factors than those found in peripheral blood [4].

CO_2 is injected transcutaneously in carboxy treatment, which is a noninvasive method. By boosting the amounts of growth hormones, this treatment encourages the development of new blood vessels. This increased blood flow delivers oxygen and nutrients to the skin and blood vessels, hence improving the skin's look [5].

Mesotherapy refers to a number of minimally invasive procedures that involve intracutaneous or subcutaneous injections of fluids. Some of the most popular substances injected in mesotherapy are vitamins, such as vitamin C, which can assist in the healing of face skin [5].

This study aims to examine the efficacy and safety of carboxytherapy, plasma gel injection, and vitamin-C mesotherapy in the treatment of PODC.

Patients and Methods:

This was a comparative, simple randomized, controlled study carried out on 30 patients presenting with periorbital dark circleswho had not received any treatment 3 months before joining the study who were admitted to Outpatient Clinic of Dermatology and Venereology Department, Tanta University Hospitals, from December 2019 to December 2020.

The study was started after being approved from Dermatology and Venereology Department, Tanta University Hospitals. Signed consent was obtained from all cases.

Exclusion criteria including pregnant or lactating patients, heavy smokers, patients using anticoagulants, patients suffering from or having a history of blood dyscrasias, active infection or inflammation in the treated area, patients with other systemic or skin diseases and history of keloid formation or foreign body reaction.

The patients were randomly allocated into three groups according to the therapeutic regimen; **Group A**: 10 patients treated with bilateral injection of PRP gel, **Group B**: 10 patients treated with bilateral intradermal injection of CO_2 with a carboxytherapy machine and **GroupC**: 10 patients treated with intradermal injection of vitamin C. All patients received 4 sessions with 2 weeks apart.

All patients were subjected to complete history taking, general and dermatological examination, and clinical assessmentwas done for all patients to determine the type of dark circles.

Plasma gel preparation:a sample of10 mL autologous whole venous blood was collected from an antecubital vein. Each blood sample was centrifuged for 10 minutes at 3000 RPM at room temperature, The plasma layer was centrifuged for another 5 minutes at 1000 RPM to

obtainplatelet-poor plasma (PPP) for injection subcutaneously through filler cannula of size 23G at the point ofnasojugal fold projection line converges with the perpendicular line to the outer canthus of the eye, then treated area was massaged, a cool compress was applied for the next 4 to 6 hours to reduce bruises.

Carboxytherapy approach: The instrument used was a carboxytherapy devicethat includes a CO_2 gas tube, power supply, CO_2 regulator, skin stabilizer, short extension tube, long extension tube, bacteria filterand an injection gun (CONCERTO, BFP Electronique Pole Technoloique de Vimenet 48 100 MONTRODAT – France).The injections were performed in specific points at 2 or 3 punctures, 1 or 2 cm spaced 1 or 2 seconds each, made all along the external inferior border of the ~~eye~~ orbit and at thelateral upper eyelid, bilaterally by a 32G needle ~~to perform the injections~~.The total amount of gas administered was equal to 1-2mL for each side with flow rate 1cc/sec.

Statistical analysis

Statistical analysis was done by SPSS v20 (Armonk, NY: IBM Corp). Quantitative variables were compared by Chi-square test and Monte Carlo correctionand were presented as range (minimum and maximum), mean, standard deviation, median and interquartile range (IQR). Qualitative variables were presented as numberand percentage (%).ANOVA test, Kruskal Wallis test, McNemar Bowker test were used to compare between more than 2 groups and Spearman correlation for non-parametric variables. P value ≤ 0.05 wasstatistically significant

Results

The demographic data were illustrated in Table 1

Table 1: Demographic data in studied groups.

Variables		Group A (n = 10)	Group B (n=10)	Group C (n=10)	P
Sex	Male	3(30%)	3(30%)	2 (20%)	1.000
	Female	7(70%)	7(70%)	8 (80%)	
Age (yr.) Mean ± SD.		38.1 ± 10.60	40.5 ± 6.69	30.6 ± 4.33	0.020*

Age groups	Early adulthood Middle adulthood	6 (60%) 4 (40%)	5 (50%) 5 (50%)	10 (100%) 0 (0.0%)	0.077
Duration (yr.) Mean ± SD.		6.5 ± 1.96	7.3 ± 1.77	7.1 ± 1.79	0.524
Family history	- ve + ve	6(60%) 4 (40%)	4 (40%) 6(60%)	5(50%) 5(50%)	0.670

Data was presented as mean ± SD or frequency and percentage, * $p \leq 0.05$ is statistically significant

There was a statistically insignificant relation between the studied groups regarding type of PODC (P value: 0.301). Table 2

Table 2: Type of PODC

Variables	Group A	Group B	Group C	P
Type of PODC				0.301
Structural	2 (20%)	0 (0.0%)	0 (0.0%)	
Pigmented	2 (20%)	1(10%)	5 (50%)	
Vascular	2 (20%)	2 (20%)	1(10%)	
Mixed				
• (VS)	2 (20%)	1(10%)	0 (0.0%)	
• (PS)	1(10%)	2 (20%)	2 (20%)	
• (PV)	1(10%)	4 (40%)	2 (20%)	

Data was presented as frequency and percentage, * $p \leq 0.05$ is statistically significant

Cases distribution according to PODC grading before and after treatment was showed in Table 3

Table 3: Distribution of group A, B and C patients according to PODC grading before and after treatment:

PODC grades	Group A		Group B		Group C	
	Before	**After**	**Before**	**After**	**Before**	**After**
Grade 0	0 (0.0%)	3(30%)	0 (0.0%)	2 (20%)	0 (0.0%)	1 (10%)
Grade I	0 (0.0%)	2 (20%)	0 (0.0%)	4(40%)	0 (0.0%)	5(50%)
Grade II	4(40%)	5(50%)	4(40%)	4(40%)	1 (10%)	4(40%)
Grade III	6 (60%)	0 (0.0%)	4(40%)	0 (0.0%)	8 (80%)	0 (0.0%)
Grade IV	0 (0.0%)	0 (0.0%)	2 (20%)	0 (0.0%)	1 (10%)	0 (0.0%)
P	0.036*		0.112		0.021*	

Data was presented as frequency and percentage, * $p \leq 0.05$ is statistically significant

Cases distribution according to degree of improvement in relation to PODC type was showed in Table 4

Table 4: Distribution of group A, B and C patients according to degree of improvement in relation to PODC type.

PODC	Group A			Group B			Group C			
	Excel-lent 50%	**Fair 20%**	**Poor 30%**	**Excell -ent 50%**	**Good 10%**	**Fair 40%**	**Excell -ent 40%**	**Good 30%**	**Fair 20%**	**poor 10%**

Structural		2 20%	-	-	-	-	-	-	-	-	-
Pigmented		-	-	2 20%	-	-	1 10%	4 40%	1 10%	-	-
Vascular		-	-	1 10%	2 20%	-	-	-	-	-	1 10%
Mixed	**VS**	2 20%	-	-	-	-	1 10%	-	-	-	-
	PS	1 10%	-	-	3 30%	1 10%	-	-	1 10%	1 10%	-
	PV	-	1 10%	-	-	-	2 20%	-	1 10%	1 10%	-
P		0.081			0.228			0.024*			

Data was presented as frequency and percentage, * $p \leq 0.05$ is statistically significant

Comparison between the three studied groups according to the degree of improvement showed no statistically significant difference between them (P= 0.245). Table 5

Table 5: Degree of improvement between studied groups

Degree of improvement	Group A (n = 10)	Group B (n=10)	Group C (n=10)	P
Excellent	5 (50%)	5 (50%)	4 (40%)	0.245
Good	0 (0%)	1 (10%)	3 (30%)	
Fair	2 (20%)	4 (40%)	2 (20%)	
Poor	3 (30%)	0 (0%)	1 (10%)	

Data was presented as frequency and percentage, * $p \leq 0.05$ is statistically significant

There was a statistically significant relation between the studied groups regarding duration of PODC (P <0.001). Table 6

Table 6: Relation between degree of improvement and duration of PODC in studied groups

Variables	Degree of improvement				P
	fair	Poor	Good	Excellent	
Duration (yr.) Mean ± SD.	8.5 ± 2.38	8.0 ± 1.93	6.0 ± 1.22	3.0 ± 1.15	<0.001*

Data was presented as mean ± **SD**, * $p \leq 0.05$ is statistically significant

Correlation between degree of improvement and duration of PODC according to Spearman correlation showed statistically significant negative strong correlation (P= 0.006, r_s= 0.494). Figure1

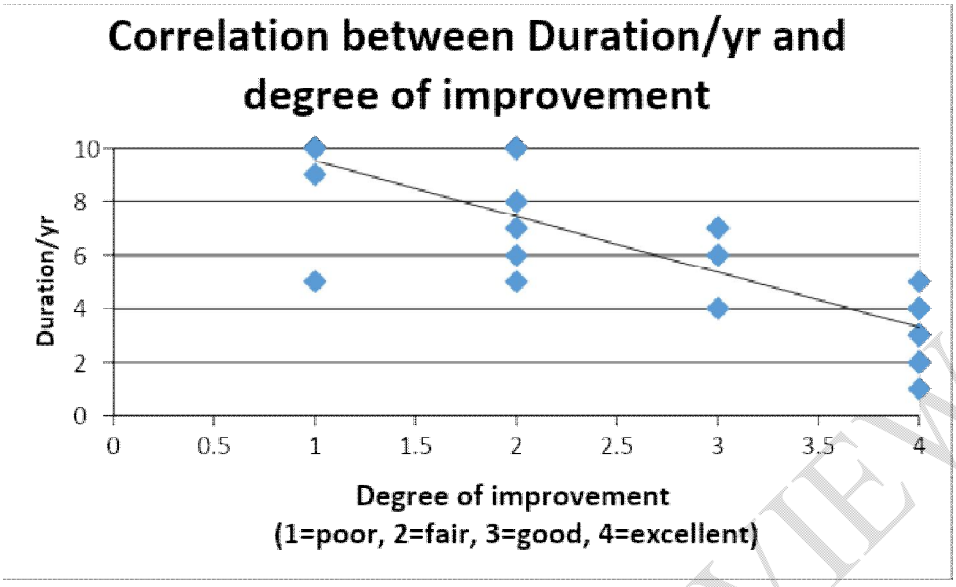


Figure 1: Correlation between duration/yr and degree of improvement in all groups.

There was a statistically insignificant relation between the three studied groups regarding recurrence rate (P= 0.531). Table 7

Table (7): Recurrence rate after the end of treatment in studied groups:

Variables	Group A (n = 10)		Group B (n=10)		Group C (n=10)		P
	N.	%	N.	%	N.	%	0.531
Recurrence							
- ve	6	60.0	7	70.0	4	40.0	
+ ve	4	40.0	3	30.0	6	60.0	

Data was presented as frequency and percentage, * $p \leq 0.05$ is statistically significant

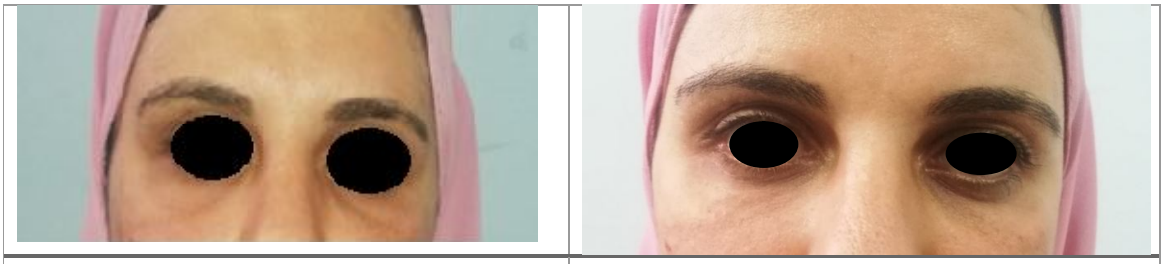
There was a statistically insignificant relation between the three studied groups regarding patient satisfaction (P= 0.895). Table 8

Table (8): Patient satisfaction in studied groups:

Variables	Group A (n = 10)	Group B (n=10)	Group C (n=10)	P
Patient satisfaction Unsatisfied	5 (50%)	4 (40%)	3 (30%)	0.895
Satisfied	5 (50%)	6(60%)	7 (70%)	

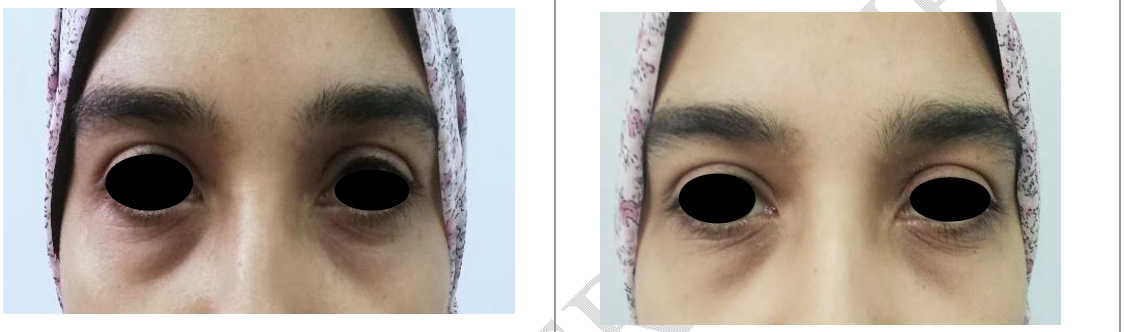
Data was presented as frequency and percentage, * $p \leq 0.05$ is statistically significant

Case (1)



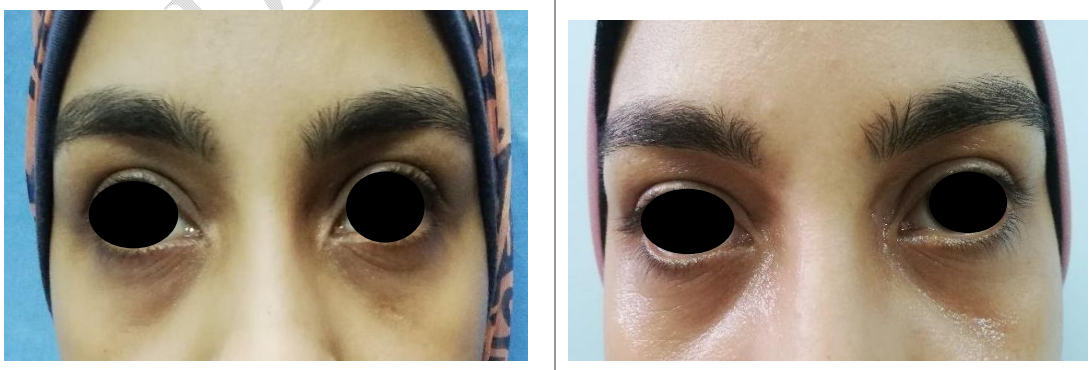
(A) (B)

Figure 2: A 44-year old female patient with bilateral PODC, pigmented structural type (grade III). (A): Before treatment with PRP gel injection. (B): After treatment, showing excellent improvement (grade 0).



(A) (B)

Figure 3: A 30-year old female with bilateral PODC, pigmented vascular type (grade III). (A): Before treatment with carboxytherapy. (B): After treatments, showing excellent improvement (grade I).



(A)	(B)

Figure 4: A 30-year old female with bilateral PODC, pigmented type (grade III). (A): Before treatment with vitamin C mesotherapy. (B): After treatment, showing excellent improvement (grade I).

Discussion

Periorbital dark circles (PODC) are characterised as bilateral, round, homogeneously pigmented macules on the periorbital areas; this condition is rather prevalent and affects people of all ages, sexes, and races. This condition develops with age and can be a serious aesthetic problem for female patients. PODC alter the facial appearance, giving the patient a fatigued, unhappy, or bloated appearance[6].

In reality, periorbital pigmentation can be caused by a number of reasons. It may be an indication of a systemic illness, skin issue, allergic response, nutritional deficit, or sleep disorder. On the other hand, it has been indicated that cutaneous hyperchromia of the orbital area is not always connected with systemic illness; it has also been observed in healthy individuals. Bleaching creams, topical retinoic acid, chemical peels, laser therapy, autologous fat transplantation, injectable fillers, microneedling, and carboxytherapy are available therapies for PODC[7].

Platelet-rich plasma (PRP) and carboxytherapy may hold promise for face rejuvenation and treatment of PODC in aesthetic dermatology.[8].

To the best of our knowledge, there is no previous study evaluating the effect of PRR gel treatment on PODC. In this study, ~~there were~~ 50% of the studied cases in thePRP gel group had an excellent improvement.As a point of interest, there is a study by **Al-Shami, [2]**in which three ~~months of~~ monthly platelet-rich plasma injections were administered to patients. Six months after the initial session, two blinded dermatologists utilising standardised digital photography and the patient's self-assessment evaluated the final outcome. According to the dermatologists' judgement, the outcome was~~highly~~ excellent, as about 60% of patients had moderate to considerable improvement.

In group B,there were 1 (10%) patient with pigmented type, 2 (20%) patients with vascular type, 1(10%) patient with vascular structural type, 2 (20%) patients with pigmented structural type and 4 (40%) patients with pigmented vascular type.

As regard the degree of improvement in group B who weretreated with carboxytherapy, 5 (50%) subjects cases had excellent improvement, 2 (20%) of them were vascular type and 3 (30%) of them were pigmented vascular. Good improvement occurred in 1 (10%) of patients with pigmented vascular type. The other 4 (40%) of patients showed fair improvement, 1(10%) of them was pigmented type, 1(10%) was vascular structural type and 2 (20%) were pigmented structural type. Carboxytherapy showed excellent and good results mainly with patients with PODC vascular and mixed vascular types, however it showed fair results with structural and pigmented types, but the degree of improvement between the different types of PODC in this group was statistically insignificant (P= 0.081).

This was in agreement with the study done by **Hassan et al., 2016[8],** who stated that the degree of improvement of PODC for the group treated by carboxytherapy was excellent in 46.7% of the patients, while showing good and fair results in 40% and 13.3% of them, respectively.

In group C,there were 5 (50%) patients with pigmented type, 1 (10%) patients with vascular type, 2 (20%) patients with pigmented structural type and 2 (20%) patients with pigmented vascular type.

Regarding the degree of improvement in group C, 4 patients (40%) had excellent improvement who had mainly pigmented type. Good improvement occurred in 3 patients (30%), one of them was pigmented type, one was pigmented vascular, and the last one was pigmented structural type. Two (20%) patients showed fair improvement, one of them were pigmented vascular and the other one was pigmented structural type. 10% of patients showed poor improvement who had mainly vascular type. Treatment with vitamin C mesotherapy

showed excellent results mainly with patients with PODC pigmented type. This finding suggests that the main cause of improvement might be due to the anti-pigmentary and antioxidant effect of vitamin C, so most patients noticed lightening of pigmentation after the first two sessions. However, it showed fair and poor results with mixed and vascular types. The degree of improvement between the different types of PODC in group C was statistically significant (P= 0.024).

In accordance with our study, **Ohshima et al. [9],** used two types of 10% vitamin C lotion, sodium ascorbate and ascorbic acid glucoside for six months in a split-faced manner for dark circles and found that there was lightening of pigmentation owing to concealment of dark discoloration due to increase in dermal thickness.

In our study there was alteration in PODC grading before and after the treatment. **In group A,** before treatment, 6 (60%) patients had grade III and 4 (40%) patients had grade II. After treatment they became 3 (30%) patients with grade 0, 2 (20%) patients with grade I and 5 (50%) patients with grade II and there was a statistically significant improvement between patients of group A regarding grading of PODC before and after treatment (P= 0.036).

In group B, before treatment with carboxytherapy, there were 2 (20%) patients with PODC grade IV, 4 (40%) patients with grade III and 4 (40%) patients with grade II. After treatment, PODC grading of this group patients have changed into 2 (20%) patients with grade 0, 4 (40%) patients with grade I and 4 (40%) patients with grade II. There was statistically insignificant difference between patients of group B regarding grading before and after treatment,which agree with the study done by **West and Alter[10],** who studied 12 female patients with PODC grades ranged from 2 to 4 before treatment and after treatment ranged from 1 to 3 with non-significant difference in between.

In group C, before treatment with vitamin C mesotherapy, 1 (10%) patient had grade IV, 8 (80%) patients had grade III and 1 (10%) patient had grade II. After treatment with vitamin C

mesotherapy, they became 1(10%) patient with grade 0, 5 (50%) patients with grade I and 4 (40%) patients with grade II. There was statistically significant difference between group C patients regarding grading before and after treatment (P= 0.021) and this in agreement of the study done by **Ahmed et al.** [5]who found that the group treated with vitamin C mesotherapy showed a higher degree of excellent improvement with statistically significant improvement between group patients.

Regarding patient satisfactionthere was no statistically significant difference in between the three treatment modalities in PODC with P= 0.895

In accordance with our study, a study done by**Ahmed et al.**[5]found that there was no statistically significant difference between carboxy therapy and mesotherapy groups regarding patient satisfaction after treatment (P > 0.05). However, mesotherapy group showed a higher degree of excellent satisfaction after treatment and that agrees with our study.

Regarding the duration of PODC, we found statistically significant negative strong correlation between degree of improvement and duration of PODC according to Spearman correlation. This means that the shorter the duration of PODC, the better the improvement with treatment. This agrees with the study done by **Hassan AM et al.**[11]who found negative moderate correlation between duration of PODC and degree of improvement.

Plasma gel, as one of the platelets formulations, works through release of several growth factors upon degranulation of their platelets` α-granules including PDGF, TGF-B, IGF, VEGF, and EGF[12]**.**

Carboxytherapy which is transcutaneous and intradermal injection of CO_2 that is recognized subcutaneously as an oxygen deficit,~~and this~~ causes an increase in blood flow and growth factors like VEGF that stimulates neo angiogenesis and so increases transcutaneous oxygen supply due to increase capillary blood flow[14]**.** Carboxytherapy is also effective in reducing

the maximum circumference of the treated area as CO_2 infiltration showed fracturing of the adipose tissue, with lysis of the adipocytes not involving the vessel structure. This lipolytic effect was observed in patients with adipose bags under the eye, so it is effective in improving skin irregularities and texture.

The vitamin C acts as an antipigmentation agent. It inhibits the function of tyrosinase, the major enzyme responsible for the conversion of tyrosine to melanin, by interacting with copper ions at tyrosinase-active sites[15]. Vitamin C also directly stimulates the transcription factors involved in collagen production and stabilises the procollagen mRNA that controls the synthesis of Type I and Type III collagen. In addition, vitamin C enhances collagen gene expression and tissue MMP-1 inhibitor production, which lowers collagen breakdown. [16]

Conclusions:

Gel injection, carboxytherapy and vitamin C mesotherapy were effective and safe therapeutic modalities in thetreatment of PODC. Improvement of PODC was shown by the decrease of its severity and improvement of skin texture of periorbital area. Improvement of PODC occurred from the first session mainly with PRP gel injection and carboxytherapy, Plasma gel was effective in all types of PODC with marked improvement in structural type, while carboxthytherapy showed marked improvement in vascular type of PODC and vitamin C mesotherapy was effective mostly with pigmented type of PODC, Minimal and temporary side effects as pain, edema, erythema and ecchymosis were noticed and well tolerated with no needed medical treatment.,

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