Comparitive Study between Autologous Platelet-Rich Fibrin Membrane as A Second Layer Versus Dartos Flap in Complicated Hypospadias Cases in Pediatrics

Original Article

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ABSTRACT

Background: Hypospadias, a common congenital anomaly, requires surgical intervention to enhance functional and aesthetic outcomes. Despite advancements, postoperative complications such as urethra-cutaneous fistula (UCF) and glans dehiscence remain prevalent.

Objectives: This study compares its results with those of the conventional Dartos flap technique to assess the effectiveness of platelet-rich fibrin (PRF) membrane as a second layer covering in the healing of complicated hypospadias in pediatric male patients.

Patients and methods: A total of 56 male patients, ranging in age from 6 months to 16 years old, were recruited and equally divided into two groups for a randomized controlled trial. Group A had a traditional Dartos flap covering, whereas group B used a PRF membrane.

Results: Median age was 5.25 years (group A) and 3.17 years (group B). Group B showed significantly faster edema resolution (3 vs. 4 days, *P*= 0.029) and reduced UCF incidence (3.6 vs. 21.4%, *P*= 0.043) and infection rates (3.6 vs. 28.6%, *P*= 0.024). Operative time was shorter in group B (93.64 vs. 99.86min, *P*= 0.037).

Conclusion: PRF membrane significantly enhances surgical outcomes in hypospadias repair by reducing the occurrence of UCF and infection while minimizing operative time. PRF offers a promising alternative to conventional techniques, particularly in complicated cases.

Key Words: Dartos flap, Hypospadias, Pediatric surgery, Platelet-rich fibrin, Urethro-cutaneous fistula.

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INTRODUCTION

Hypospadias is a common congenital abnormality that frequently affects boys, which suggests an abnormal opening of the meatus urethrae on the ventral side of the penis[1]. In severe cases, penile chordee is present and is probably caused by environmental and endocrinal factors, which led to an increase in its prevalence over the past three decades worldwide[2]. It is generally estimated to occur around 1: 250 for males[3]. Besides lowering postoperative complications and reoperation rates, hypospadias repair seeks to improve both functional and cosmetic outcomes[4].

Despite advances, postsurgical complications still affect nearly half of hypospadias repairs worldwide, prompting interest in materials that enhance wound healing and are widely available[5].

It has been demonstrated that the fistula rate can be decreased by covering the neourethral suture line with an extra waterproofing layer of vascularized tissue. The most popular sources of this intermediate layer are dartos (DF) and tunica vaginalis flaps because of their accessibility and good vascularity[6].

Patients who undergo repeated surgeries for hypospadias end up with a scarred, hypo-vascular, and shorter penis. Different levels of scarring are caused by numerous surgical operations, and poor healing may be the outcome of tissue ischemia[7].

Platelet-derived concentrates evolved as a result of the search for novel biomaterials that improve wound healing.

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The qualities of its previous generations have been improved by the creation of a second-generation platelet concentrate, which eliminates the need for challenging biochemical reagents. Growth factors can be derived autologously from platelet-rich fibrin (PRF). The patients' sera are used to prepare it. The most often found growth factors in the PRF sample are endothelial growth factor, vascular endothelial growth factor, and transforming growth factor β. PRF can be employed as an intermediate layer between the neourethra and the skin to decrease the risk of fistula development since it promotes collagen production, tissue regeneration, wound healing, and angiogenesis[8].

This study's objective is to evaluate the autologous PRF membrane outcomes as a second layer urethroplasty in pediatric complicated hypospadias repairs, focusing on primary outcomes such as postoperative complications [e.g. glanular dehiscence, neourethral disruption, wound infection, urethro-cutaneous fistula (UCA), meatal stenosis] and secondary outcomes including voiding difficulties and operative time, comparing its efficacy to the DF technique.

PATIENTS AND METHODS

Design and population

This randomized controlled clinical trial, Conducted at the pediatric surgery unit in the General Surgery Department of Kafr Elsheikh University Hospital from March 2023 to June 2024, encompassed 56 patients. The study received ethical approval from the Faculty of Medicine's Research Ethics Committee, Kafr Elsheikh University (Approval Code: KFSIRB200-14), and informed consent was acquired from all parents after discussing the risks and benefits in detail. Names were not mentioned. The results of the investigations were used for scientific purposes only.

Patients were divided into two equal groups each one is 28 patients: group (A) included conventional DF covering patients, and group (B) included the PRF membrane as a second layer covering patients.

Randomization

All patients who fulfilled the inclusion criteria and their parents gave consent for participation were randomized by online software (Research Randomizer Version 4.0 at https://www.randomizer.org), stratified blocks ensured balanced numbers in each group. Each patient took an unlabeled blue or black sealed envelope. The code of each color was known to the medical team. The blue color referred to the group (A) conventional DF covering, and the black color referred to the group (B) PRF membrane as a second layer covering.

Inclusion criteria

a. pediatric male patients.

b. Patients aged between 6 months to 16 years old.

c. Patients who had undergone previous hypospadias repair, including glanular, coronal, subcoronal, mid-penile, and proximal repairs.

Exclusion criteria

a. Patients with primary hypospadias.

b. Patients who were diagnosed with Disorders of Sexual Differentiation.

c. Cases that were presented with severe chordee were excluded from the study.

Preoperative assessment

Included a detailed clinical history and a comprehensive examination, covering both general and local assessments critical for operative planning. Key aspects examined in hypospadias cases were the meatus location, proximal spongiosal hypoplasia degree, presence of ventral curvature severity, the urethral plate quality and width, glans width, ventral skin deficiency degree, scrotal abnormalities (such as penoscrotal transposition and bifid scrotum), foreskin availability, and any residual torsion.

Preoperative investigations comprised laboratory tests (complete blood count, PT, PTT, INR, and urine analysis) and pelvi-abdominal ultrasound (if needed) to rule out associated conditions (e.g. solitary kidney or hydronephrosis). Preparation included stopping local androgen at least 1 week and systemic hormones like testosterone or HCG 2 weeks before surgery in patients who were receiving it outside our center, and then referred to us, but we did not use any hormonal therapy as a routine. And reviewing anesthesia fitness the night before the operation.

Intraoperative technique

Prophylactic intravenous antibiotic (4^{th}-generation cephalosporin "Cefepime" was administered half an hour before the procedure. An energy vessel sealing device (bipolar diathermy) was prepared and tested before the procedure. Additionally, a centrifuge and two sterile vacutainers were set up for PRF preparation after hypospadias repair. All patients were positioned supine with both upper limbs extended.

All participants were subjected to Snodgrass tubularized incised plate urethroplasty technique by the same operator under general anesthesia. After complete penile degloving, any residual chordee or torsion was assessed. Mild chordee ($<30^{\circ}$) was corrected through dorsal plication with 6/0 PDS suture.

Over a suitable nelaton catheter (8–12fr), a two-layer urethroplasty was performed, beginning with a running inverting 6/0 vicryl suture, followed by a second imbricating layer using the same suture.

A vascularized dartos fascia layer from the dorsal shaft skin was used to cover the neourethra in group A, while an autologous PRF membrane prepared intraoperatively provided coverage in group B (Figure 1), secured with 6/0 Vicryl sutures. The catheter was left as a stent for urinary drainage for 5–7 days.

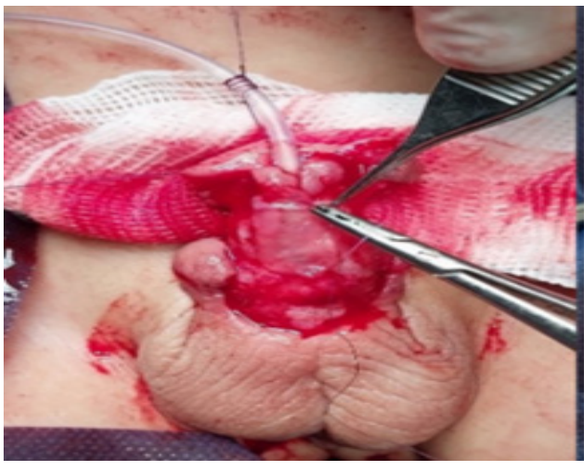

Figure 1: Autologous platelet-rich fibrin membrane fixation as a second layer coverage urethroplasty in TIP repair.

To prepare PRF, 10ml of venous blood was collected intraoperatively using butterfly needles and Vacutainer tubes. Blood samples underwent immediate centrifugation at 3000rpm for a period of 10m. After centrifugation, the middle layer containing the fibrin clot was isolated 2mm below the lower dividing line. The PRF layer was carefully extracted with forceps, with the red thrombus (red blood cells) section removed along its border with scissors. The PRF clot was then compressed between two sterile glass slides (Figures 2,3).

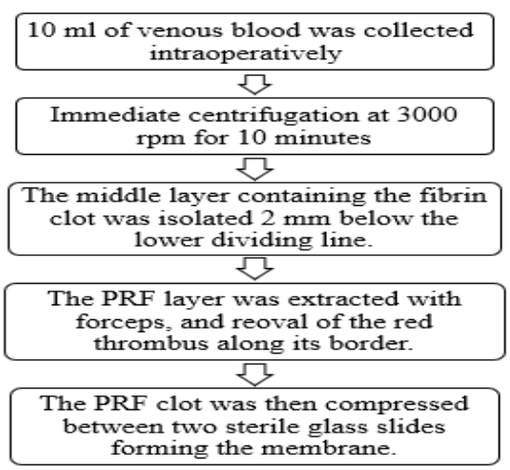

Figure 2: Schematic diagram showing steps for the autologous platelet-rich fibrin membrane preparation.

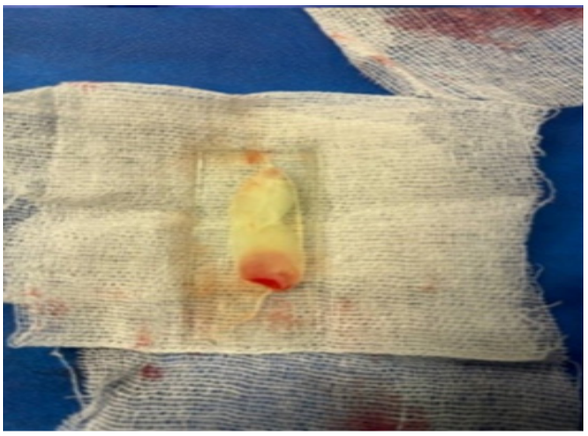

Figure 3: The platelet-rich fibrin clot is compressed between sterile glass slides, resulting in the formation of the platelet-rich fibrin membrane.

Postoperative follow-up

Nursing patients in a supine position. Strong analgesics, including IV paracetamol and nonsteroidal anti-inflammatory drugs suppositories, were provided as needed. Patients received IV 4th-generation cephalosporin antibiotics during their hospital stay and continued on oral antibiotics for a total of 10 days postdischarge. Dressings were removed three days postoperation, and local antibiotics, such as fusidic acid cream and amikacin topical spray, were applied. The urinary catheter was typically removed on the 57th day postoperatively. Follow-up involved weekly outpatient clinic visits for the first month and monthly visits for 6 months, with evaluations at 6 weeks and 6 months to check for meatal stenosis or neourethral stricture through calibration of the new meatus.

Sample size

Based on the incidence of 6 months postoperative wound infection in 5% and 35% PRF hypospadias and normal hypospadias, respectively, Eryilmaz *et al*., the sample size is calculated to be 28 in each group using open Epi software at a confidence level of 95% and power of the study 80%.

Statistical analysis

SPSS version 28 was utilized to analyze the data. The Shapiro–Wilk test was used to determine if quantitative data was normal, while the χ^2, Fisher's exact, and Monte Carlo tests were used to analyze categorical variables. Multivariant analysis examined inter-factor correlations and predictive values, and both parametric and nonparametric techniques (*t*-tests, Mann–Whitney tests, and Spearman's correlation) were used, with statistical significance set at *P* less than 0.05.

RESULTS

In comparing group A and group B, the participants' median age was 5.25 years (IQR: 2.92–8.5) for group

A and 3.17 years (IQR: 2–6.75) for group B, with a Z value of -1.582 and a P value of 0.114, indicating no statistically significant difference. In terms of preterm outcomes, invitro fertilization, type of prior surgery, family history, hormonal therapy received preoperatively, and preoperative evaluations such as meatus, chordee, glans width, torsion, or urethral plate width, the study found no statistically significant differences between the groups; notably, none of the patients in either group experienced scrotal transposition (Table 1).

Table 1: Comparison between the studied groups regarding demographic data

	Group A	**Group B**		
	Median (IQR)	**Median (IQR)**	***Z***	***P***
Age (year)	5.25(2.92–8.5)	3.17(2–6.75)	-1.582	0.114
	N= 28(%)	*N*= 28(%)	$\chi 2$	*P*
IVF				
No	24(85.7)	25(89.3)	Fisher	>0.999
Yes	4(14.3)	3 10.7)		
Postnatal history				
Preterm	3(10.7)	4(14.3)	Fisher	>0.999
Term	25(89.3)	24(85.7)		
Type of previous surgery				
TIP	24(85.7)	25(83.3)	Fisher	>0.999
Modified koyanagi	4(14.3)	3(16.7)		
Family history				
No	27(96.4)	26(92.9)	Fisher	>0.999
Yes	1(3.6)	2(7.1)		
Hormonal therapy				
No	25(89.3)	24(85.7)	MC	>0.999
Local	2(7.1)	2(7.1)		
Systemic (IM injection)	0	1 (3.6)		
Local and systemic	1 (3.6)	1 (3.6)		
Meatus				
Glanular	2(7.1)	0	MC	0.125
Coronal	10(35.7)	4(14.3)		
Subcoronal	9(32.1)	12(42.9)		
Mid-penile	5(17.9)	10(35.7)		
Proximal shaft	2(7.1)	2(7.1)		
Residual chordee				
No	27(96.4)	23(82.1)	Fisher	0.193
Yes	2(3.6)	5(17.9)		
Residual torsion				
No	28(100)	26(92.9)	Fisher	0.491
Yes	0	2 (7.1)		
UCF				
No	22(78.6)	27(96.4)	MC	0.043*
Yes	6(21.4)	1(3.6)		
Infection				
No	20 (71.4)	27 (96.4)	Fisher	0.024*
Yes	8 (28.6)	1 (3.6)		
Glans dehiscence				
No	25 (89.3)	26 (92.9)	Fisher	>0.999
Yes	3 (10.7)	2 (7.1)		
Complete dehiscence				
No	25 (89.3)	27 (96.4)	Fisher	0.611
Yes	3 (10.7)	1 (3.6)		
Stream				
Normal	14 (50)	21 (75)	MC	0.143
Deviated	0	1 (3.6)		
Double	3 (10.7)	2 (7.1)		
Spray	9 (32.1)	2 (7.1)		
Stenotic	2 (7.1)	2 (7.1)		
	Median (IQR)	**Median (IQR)**	*Z*	*P*
Resolution of edema (day)	4(3–5)	3(3–4)	-2.184	0.029*
	Mean±SD	**Mean±SD**	***T***	***P***
Urethral plate (mm)	9.11±1.15	9.02±1.47	0.253	0.801
Glans width	13.21±1.99	12.57±1.57	1.342	0.185
Operative time (min)	99.86±9.49	93.64±12.11	2.137	0.037*

Previous operation, family and drug history, Pre and postoperative examination, and operative time; *: *P* less than 0.05 is statistically significant; IM: Intramuscular; IQR: Interquartile Range; IVF: Invitro fertilization; MC: Monte Carlo test; min: minute; SD: Standard Deviation; *t*: Independent sample *t*-test; TIP: Tubularized Incised Plate; UCF: Urethro-Cutaneous Fistula; *Z*: Mann–Whitney test; χ^2: Chi square test.

A statistically significant association was observed between infection incidence and operative time, age, UCF, and time till edema resolution (Table 2).

Table 2: Relation between infection and the studied parameters

	No infection Median (IQR)	**Infection Median (IQR)**	***Z***	***P***
Age (year)	5.25(2.92–8.5)	2.46(1.81–4.81)	-2.681	0.007*
Edema	4(3–5)	3(3–4)	-2.184	0.029*
	Mean±SD	**Mean±SD**	***T***	***P***
Glans width	12.94±1.79	12.67±2.0	0.407	0.686
Operative time (min)	95.13±10.05	105.22±13.76	-2.597	0.012*
UCF, n (%)				0.002*
No	44(93.6)	5(55.6)	χ^2	
yes	3(6.4)	4(44.4)	MC	

*: *P* less than 0.05 is statistically significant; IQR: Interquartile Range; t: Independent sample t-test; UCF: Urethro-Cutaneous Fistula; Z: Mann–Whitney test; χ^2: Chi square test.

The binary regression analysis indicates that factors such as age and edema resolution time do not significantly increase the risk of infection, with odds ratios of 1.207 and 1.062, respectively. However, the presence of UCF significantly increases the risk of infection by 8.629 times.

Additionally, increasing age is associated with a decreased risk. Statistically significant results are marked by *P< 0.05 (Table 3).

Table 3: Binary regression analysis of factors related to infection

				95% CI	
	B	***P***	**AOR**	**Lower**	**Upper**
Age	-0.184	0.225	0.832	0.618	1.12
Edema resolution (days)	0.143	0.595	1.153	0.682	1.949
Operative time (min)	0.064	0.362	1.067	0.929	1.225
UCF (yes)	2.136	0.015*	8.629	3.547	19.634

AOR, adjusted odds ratio; CI, confidence interval; UCF, urethro-cutaneous fistula, min: minutes. *P less than 0.05 is statistically significant.

The rate of UCF (21.4% in group A vs. 3.6% in group B, P= 0.043), the incidence of infection (28.6% in group A vs. 3.6% in group B, P=0.024), the time it took for edema to resolve (median of 4 days in group A vs. 3 days in group B, P= 0.029), and the time needed for surgery (mean of 99.86 min for group A vs. 93.64 min for group B).the study found statistically significant difference regarding these items (Table 4).

Table 4: Comparison between two groups as regards parameters that showed statistical significance

	Group A N=28 (%)	**Group B N=28 (%)**	***χ2***	***P***
UCF				
No	22 (78.6)	27 (96.4)	MC	0.043*
Yes	6 (21.4)	1 (3.6)		
Infection				
No	20 (71.4)	27 (96.4)	Fisher	0.024*
Yes	8 (28.6)	1 (3.6)		
	Median (IQR)	**Median (IQR)**	***Z***	***P***
Resolution of edema (day)	4(3–5)	3(3–4)	-2.184	0.029*
	Mean±SD	Mean±SD	*T*	*P*
Operative time (min)	95.13±10.05	105.22±13.76	-2.597	0.012*

*P less than 0.05 is statistically significant. IQR, interquartile range; t, Independent sample t-test; UCF, Urethro-cutaneous fistula; Z, Mann–Whitney test; χ2, Chi square test.

In group A, there is a significant positive correlation between the time taken for edema resolution and operative time (Figures 4,5).

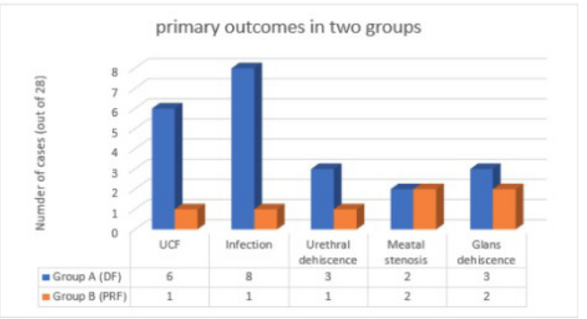

Figure 4: Multiple bar chart showing a comparison between groups regarding primary outcomes.

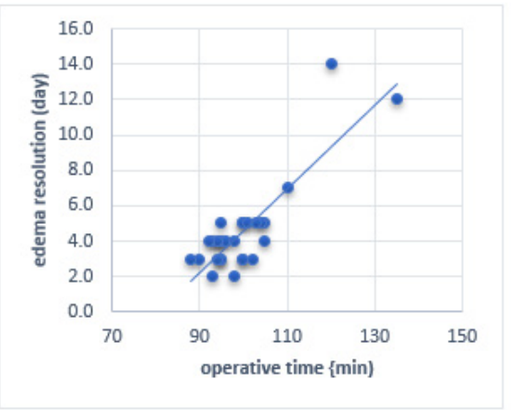

Figure 5: Scatter dot plot showing a significant positive correlation between operative time and time for edema resolution within group A.

In group B, a significant positive correlation exists between edema resolution time and both the urethral plate and operative time (Figure 6).

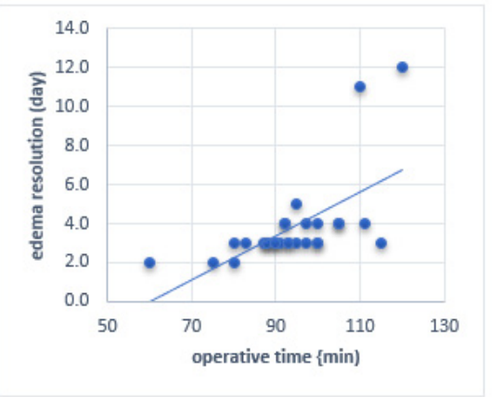

Figure 6: Scatter dot plot showing a significant positive correlation between operative time and time for edema resolution within group B.

DISCUSSION

Our study found that 28.6% of group A had a postoperative infection, while only 3.6% of group B had an infection. This is consistent with a study that was led by Eryilmaz and colleagues, who investigated postoperative outcomes in hypospadias patients, comparing those receiving PRP therapy versus those who did not. The study examined both immediate and extended postsurgical complications across two patient groups, both treated using the Snodgrass tubularized incised plate urethroplasty surgical technique. The researchers designated patients into two cohorts: group A received PRP as part of their treatment protocol, whereas group B underwent the procedure without PRP supplementation. Their analysis revealed that patients in group A, who received PRP therapy, demonstrated significantly reduced postoperative infection rates in comparison with their non-PRP counterparts[9].

The viability, safety, and effectiveness of using autologous PRF membrane for urethroplasty covering in instances of distal hypospadias were also examined in research headed by Guinot *et al*., No instances of wound infection were reported by the authors[10].

According to our research, group A experienced a 21.4% fistula incidence, whereas group B had a 3.6% incidence. According to Guinot *et al*., urethral fistula developed in 2/33 (6.06%)of the study's participants, with a median follow-up of 8 months (range: 6–18 months). For urethroplasty coverage in distal hypospadias surgery, they employed autologous PRF membrane[10].

A systematic meta-analysis study was done by Pfistermuller and colleagues, including 49 studies (4675 patients). UCF and redo surgery rates were significantly higher in complicated repairs (15.5 and 23.3%) in comparison with primary proximal (10.3 and 12.2%) and primary distal (5.7 and 4.5%) (P= 0.045 and P< 0.001, respectively)[11].

Moreover, a study conducted by Abdelazim *et al*., evaluated the therapeutic autologous PRF membrane benefits in enhancing wound healing and minimizing fistula development in primary distal hypospadias cases. The research encompassed 40 participants with primary distal hypospadias who underwent tubularized incised plate (TIP) repair, categorized into two equal groups of 20 patients each. The first group (group A) received traditional treatment utilizing preputial or local DF as a protective covering over the urethroplasty. The second group (group B) was treated with PRF membrane coverage over the urethroplasty. The outcomes revealed a notable difference in complications: group A experienced seven cases of UCF, yielding a 35% occurrence rate, whereas group B revealed only two cases of fistula formation, resulting in a substantially lower 10% incidence during the early postoperative phase[12].

To evaluate the use of autologous PRF membrane as a covering layer in the repair of urethra-cutaneous fistula following hypospadias surgeries, Wishahy *et al*., conducted another research. Twenty patients received local dartos coverage, and seventeen patients received PRF coverage out of the 37 instances that were part of the study. Five out of six instances had a minor single fistula, three cases had a recurring coronal fistula, and the other three cases developed a subcoronal fistula, according to the results of the postlocal DF[13].

Glans dehiscence was found in 10.7% of group A and 7.1% of group B, according to our study. This aligns with research conducted by Mahmoud *et al*., The goal was to evaluate the results and rates of complications associated with covering primary distal hypospadias repair using platelet-rich plasma (group A) as opposed to DF (group B). Four patients in group B and one patient in group A both had partial glans dehiscence[14].

Additionally, a study headed by Elsayem *et al*., examined the potential therapeutic benefits of autologous platelet gel in improving surgical results and healing processes after hypospadias correction. Thirty children between the ages of 6 months and 12 years were included in their study and divided into two equal groups. While group B had normal therapy without platelet gel augmentation, group A underwent the procedure with autologous platelet gel application. Although it did not approach statistical significance, the comparison of postoperative complications showed a tendency towards higher complication rates in group B compared with group A. In particular, group B showed glans dehiscence at a rate of 20%, whereas group A showed a lower rate of 7%[15].

In terms of the time required to resolve edema, our research showed a statistically significant difference between the studied groups, with group B resolving edema more quickly than group A. According to Elsayem *et al*., research group B had edema insignificantly more frequently than group A (27 vs. 13%). TIP repair with the autologous platelet gel application was performed in group A, while TIP repair without the gel was performed in group B[15].

Postsurgical edema can present as significant swelling affecting both penile and scrotal regions. When the meatal area is involved, it may temporarily affect urination patterns, causing stream dispersal, though this rarely persists long-term. Literature reports indicate an edema occurrence rate of approximately 11.11%. The condition typically becomes more pronounced following compression dressings removal, which usually occurs between the second and fifth day postsurgery. The swelling extent can be exacerbated by various factors, comprising hematoma formation, urine leakage due to bladder contractions, or premature displacement of the urethral stent[16].

However, there are some limitations to our study, including the small sample size of 56 patients, the study's narrow focus at a single center, which may affect the results' generalizability to a larger population, and the 6-month follow-up period, which may not have adequately evaluated the longer-term outcomes like urethral function and cosmetic results.

CONCLUSION

Our study indicates that the use of a PRF membrane as a second layer covering in complicated hypospadias repair significantly reduces operative time, edema resolution time, and the occurrence of UCF and infection compared with conventional DF covering. Over the follow-up period, patients in the PRF group demonstrated better postoperative outcomes, particularly in terms of fistula formation and infection rates. These results suggest that the PRF membrane can be an effective intervention for improving surgical outcomes in complicated hypospadias cases, especially in reducing postoperative complications.

ABBREVIATIONS

CBC: Complete blood cunt; **CI:** Confidence interval; **DF:** Dartos flap; **DSD:** Disorders of sexual differentiation; **Fr:** French (unit of measurement for catheter size); **HCG:** Human chorionic gonadotropin; **IM:** Intramuscular; **IVF:** Invitro fertilization; **MC:** Monte Carlo test; **NSAID:** Nonsteroidal anti-inflammatory drugs; **PDS:** Polydioxanone (a type of suture material); **PRF:** Platelet-rich fibrin; **rpm:** Revolution per minute; **SD:** Standard deviation; **TIP:** Tubularized incised plate; **TVF:** Tunica vaginalis flap; **UCF:** Urethro-cutaneous fistula.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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