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Original Article

Efficacy of pelvic floor reconstruction combined with pelvic floor rehabilitation instrument on MMP-7, TIMP-1 and pelvic floor dysfunction in women

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Abstract: Objective: To investigate the efficacy of pelvic floor reconstruction combined with pelvic floor rehabilitation instrument on pelvic floor dysfunction (PFD) in women and on the expression of matrix metalloproteinase-7 (MMP-7), tissue inhibitor of metalloproteinase-1 (TIMP-1) in ligaments. Methods: A total of 97 PFD patients who were admitted to Shouguang People's Hospital from September 2016 to October 2017 were selected and divided into control group (n=48) and study group (n=49) according to different treatment methods. Control group was treated by laparoscopic high uterosacral ligament suspension combined with pelvic floor rehabilitation instrument. Study group was treated by pelvic floor reconstruction combined with pelvic floor rehabilitation instrument. The operation-related indexes, Pelvic Organ Prolapse Quantitation (POP-Q) system, Pelvic Floor Distress Inventory (PFDI-20), pelvic floor muscle strength test results, relapse rate and the effects on the expression of MMP-7 and TIMP-1 in ligaments were compared between the two groups. Results: In study group, the operation time, exhaust time and indwelling time of urinary catheter were significantly shorter than those in control group (all P<0.001); the blood loss was significantly less than that in control group (P<0.001). Six months after treatment, the PFDI-20 score in study group was significantly lower than that in control group (P<0.001), and the relapse rate in study group (4.08%) was also lower than that in control group (16.67%) with statistical significance (χ^2=4.153, P<0.001). Three months and 6 months after treatment, the pelvic floor muscle strengths in study group were significantly better than those in control group (both P<0.001). Six months after treatment, patients with grade II POP-Q in study group (2.04%) was significantly less than that in control group (16.67%, H=6.162, P<0.001), and patients with grade 0 in study group (55.10%) was significantly more than that in control group (41.67%, H=3.792, P<0.001). In study group, the expression level of MMP-7 was lower than that in control group, but the expression level of TIMP-1 was higher than that in control group with significant differences (both P<0.001). Conclusion: Pelvic floor reconstruction operation can effectively shorten the operation time and promote postoperative recovery in PFD patients, and it has low relapse rate. Combining it with pelvic floor rehabilitation instrument can effectively improve the recovery of pelvic floor muscle strength, so the combination is worth promoting.

Keywords: Pelvic floor reconstruction surgery, pelvic floor rehabilitation instrument, pelvic floor dysfunction, pelvic floor muscle strength

Introduction

Pelvic floor dysfunction (PFD) is a common gynecological disease that occurs in middle-aged and elderly women. PDF and pelvic floor support structure damage (or defects) are caused by multiple factors such as childbirth, obesity as well as the history of gravidity and parity [1]. Clinical symptoms include pelvic organ prolapse (POP), chronic pelvic pain, stress urinary incontinence (SUI), fecal incontinence and sexual dysfunction, etc. among which POP and SUI are the most common ones [2]. Clinical study has found that the incidence of PFD is related to patients' age and delivery mode; the incidence increases with the increase of age, also, abnormal pelvic floor mechanism in initial stage after eutocia can also increase the incidence of PFD in puerperae [3]. PFD treatment aims to improve clinical symptoms through improving pelvic floor muscle function. Methods for it include surgery and non-surgical

treatment. Most of the traditional surgeries are per vagina, although its efficacy is affirmed, postoperative high relapse rate is still a restriction. Therefore, how to reduce surgical trauma and improve prognosis for recovery is still a hot research topic in clinical treatment of PFD.

In recent years, based on the integral theory and the hammock theory, pelvic floor reconstruction has gradually developed as patch repair and top suspension kit, with small trauma and significant effects through the progress of overlapping vagina repair, partial repair and total pelvic floor reconstruction [4]. It has been reported that combining with pelvic floor muscle training and electrical stimulation in follow-up rehabilitation can better improve pelvic floor function, but the effects on the expression of matrix metalloproteinase-7 (MMP-7) and tissue inhibitor of metalloproteinase-1 (TIMP-1) in ligament are not clear with very limited reports [5]. In this study, Gynecare Prolift pelvic floor repair system combined with pelvic floor rehabilitation instrument were used for the treatment of PFD in women to evaluate patients' postoperative indicators, changes of pelvic floor function and effects on the expression of MMP-7 and TIMP-1, also to provide a reference for clinical treatment.

Materials and methods

General data

This study was approved by the Ethics Committee of Shouguang People's Hospital and the informed consents were obtained from all the subjects. A total of 97 patients with PFD admitted to Shouguang People's Hospital from September 2016 to October 2017 were included in this study.

Inclusion criteria: Patients with age greater than 50 years old; patients with reproductive history; patients who diagnosed as PFD by pelvic floor B-ultrasonography; patients who signed informed consent [6].

Exclusion criteria: Patients who suffered from bladder, uterine or cervical malignant tumor; patients with other serious gynecological diseases, such as cervical ulcers or vaginal irregular bleeding; patients with the history of chemoradiotherapy; patients who did not meet the surgical treatment standards; patients who suffered from diseases of nervous system or urinary system.

Methods

In control group, laparoscopic high uterosacral ligament suspension was used for hysterectomy or uterine retention [7]. Incision of pelvic peritoneum was performed along the lateral uterosacral ligament, on a horizontal line with ischial spine, followed by folding suture with 1-0 absorbable suture material, then it was fixed on the top of ipsilateral vagina and the complex site of cardinal ligament of uterus and uterosacral ligament. According to the actual damaged situation of patients' posterior vaginal wall, vaginal posterior wall repair could be added. One month after surgery, patients received rehabilitation.

In study group, pelvic floor reconstruction was performed by using the Gynecare Prolift pelvic floor repair system from Ethicon SARL, Switzerland [8]. ROLENE polypropylene mesh and a set of handy placement tools for mesh implant (including stainless steel guide rod, cannulas, PROLENE polypropylene retractor and sterilization packaging) were used. Firstly, anterior pelvic floor reconstruction surgery: puncture cones entered the obturators through the skin incisions, which were 4 cm from external urethral orifices at both sides, then through the direction of the ischial spine, near both inner sides of the inferior ramus of pubis, it was coming out through the vaginal bladder space. Puncture cones then entered at the site 2 cm below the above skin incisions and punctured at the bottom of ischial spine through the lower margins of obturators; afterwards, it was coming out through the vaginal bladder space. The Prolift mesh was tiled on vaginal anterior wall under the bladder. Secondly, posterior pelvic reconstructive surgery: puncture cones entered from the skin incisions 3 cm below both outer sides of the anus, away from rectum, and through the sacrospinous ligament, it was coming out through the bottom of ischial spine and the rectovaginal space (5-6 cm away from the vaginal orifice). The Prolift mesh was tiled on vaginal posterior wall above the rectum. If removal of uterus was performed, the Prolift mesh belt would not need to be cut apart at the middle area and only need to be entirely affixed to the vaginal anterior and posterior walls. One

Table 1. Comparison of general data between two groups

Group		Control group (n=48)	Study group (n=49)	$t/X^2/H$	P
Average age (year)		59.78±13.45	60.12±14.01	0.122	0.903
Average parity (times)		2.40±1.50	2.51±1.30	0.386	0.701
Average body mass index (kg/m^2)		25.56±2.78	25.34±2.81	0.388	0.699
Menopause (n, %)		36 (75.00)	38 (77.55)	0.087	0.768
Urinary incontinence (n, %)		15 (31.25)	14 (28.57)	0.083	0.773
Hypertension (n, %)		5 (10.42)	5 (10.20)	0.001	0.973
Diabetes (n, %)		2 (4.17)	4 (8.16)	0.667	0.414
Prolapse degree (n, %)	III	29 (60.42)	30 (61.22)	0.007	0.935
	IV	19 (39.58)	19 (38.78)	0.007	0.935

Table 2. Comparison of surgery-related indexes between two groups

Group	Operation time (min)	Bleeding volume (mL)	Exhaust time (day)	Indwelling time of urinary catheter (day)
Control group (n=48)	109.29±14.56	168.56±41.65	5.02±0.98	6.19±1.48
Study group (n=49)	85.67±16.91	121.35±34.26	3.31±0.59	4.08±1.56
t	7.377	6.090	10.385	6.835
P	0.000	0.000	0.000	0.000

month after surgery, patients received rehabilitation.

The rehabilitation included two stages. One month after surgery, the two groups of patients were treated with Phenix USB4 pelvic floor rehabilitation instrument (Beijing Yuyan Medical Devices Co., Ltd.), with mode of muscle strength recovery. After electrical diagnosis, vaginal contraction was practiced, also, electrical stimulation and biological feedback for 20 min were performed. Parameters were as follows: frequency 30 Hz, pulse width 500 μs, time 20 min. After 4 weeks of treatment, the above mode was converted to primary training for type II muscle fibers. Parameters were as follows: frequency 50 Hz, pulse width 250 μs, time 20 min. Patients were treated for a treatment course, 10-15 times, determined by pelvic floor myoelectric activity and muscle strength [9].

Observation index

There were 3 main observation indexes. Firstly, Pelvic Floor Distress Inventory (PFDI-20) and pelvic floor muscle strength of two groups before treatment, 3 and 6 months after treatment were compared; pelvic floor muscle strength was recorded by pelvic floor rehabilitation instrument; the PFDI-20 score was collected by questionnaire survey in the two groups [10]. Secondly, Pelvic Organ Prolapse Quantitation (POP-Q) system of the two groups before treatment, 3 and 6 months after treatment were compared; POP-Q System of American College of Obstetricians and Gynecologists was used for graduation, index measurement and evaluation of pelvic floor function indexes [11]. Lastly, the expression of MMP-7 and TIMP-1 in the ligaments of the two groups was observed. After HE staining of cytoplasmic cells in two groups, they were made into slices, then immunohistochemical streptavidin-peroxidase method was used for staining of anti-MMP-7 monoclonal antibody and anti-TIMP-1 monoclonal antibody respectively. The staining was observed under a microscope. The yellow-brown granules in the cytoplasm showed that MMP-7 and TIMP-1 were positive. Moreover, 10 high power fields were selected for microscope images of the granules, which were taken by a dedicated digital camera and transmitted to a computer. The number of yellow-brown granules in the cytoplasm of the uterosacral ligaments in both groups could be used to determine the content of MMP-7 and TIMP-1. Granule image processing analysis software Image J was used for image processing and analysis. More granules and deeper staining indicated smaller gray value, so that we could get average gray values of MMP-7 and TIMP-1 in the positive areas of each group.

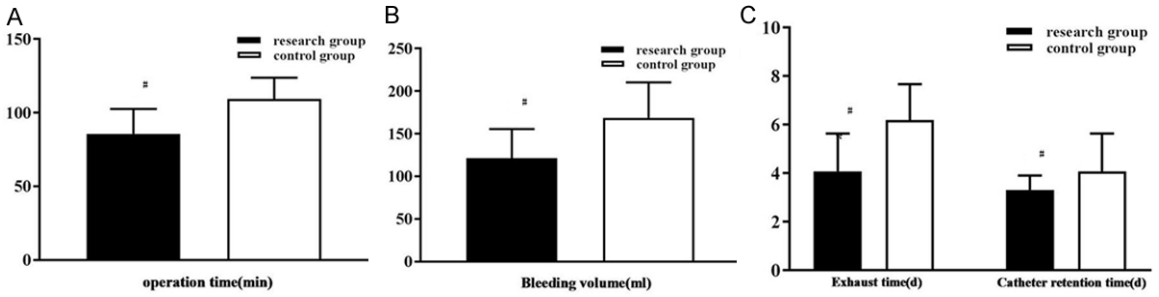

Figure 1. Comparison of surgery-related indexes between two groups. Compared with the control group, #P<0.001.

There were 2 secondary observation indexes. Firstly, the operation-related indexes of two groups were compared, including operation time, exhaust time, intraoperative blood loss and indwelling time of urinary catheter. Secondly, the relapse rates at 3 and 6 months after operation between the two groups were compared. Questionnaire survey was used to collect the relapse rate of two groups of patients.

Statistical methods

Data analysis was performed by SPSS21.0 professional statistical software. Measurement data were expressed as mean ± standard deviation, and tested by independent samples t-test, denoted as t; rank variables were tested by rank sum test, denoted by H. Enumeration data were expressed as number and rate (n, %), tested by X^2 and Fisher exact test, denoted by X^2. The difference was statistically significant when P<0.05.

Results

Comparison of the general data between two groups

There was no significant difference in general data of two groups of patients, including the average age, parity, body mass index, menopause rate, urinary incontinence rate, associated diseases and prolapse indexing (all P>0.05). See **Table 1**.

Comparison of surgery-related indexes between two groups

In study group, the operation time, exhaust time and indwelling time of urinary catheter were significantly shorter than those in control group (all P<0.001); the blood loss was significantly less than that in control group (P<0.001). See **Table 2** and **Figure 1**.

Comparison of PFDI-20 score and pelvic floor muscle strength between two groups

Before treatment, there was no significant difference in PFDI-20 score and pelvic floor muscle strength between the two groups (all P>0.05). Three and 6 months after treatment, the PFDI-20 scores in both groups were significantly lower than those of before treatment (all P<0.001). Six months after treatment, the PFDI-20 score in study group was significantly lower than that in control group (P<0.001). Three and 6 months after treatment, the pelvic floor muscle strength in both groups were significantly increased (all P<0.001), and the increase in study group was more significant than that in control group (all P<0.001). See **Table 3** and **Figure 2**.

Comparison of relapse rate between two groups

Three months after treatment, 1 patient relapsed in both groups respectively, with no statistical significance (X^2=0.001, P=0.985). Six months after treatment, 2 cases relapsed in study group (4.08%), and 8 cases relapsed in control group (16.67%). The relapse rate in study group was significantly lower than that in control group (X^2=4.153, P<0.001).

Comparison of POP-Q between two groups in different time

There was no significant difference in POP-Q system between the two groups before treatment (P>0.05). Three and six months after treatment, patients with stage II, III and IV were

Table 3. Comparison of PFDI-20 score and pelvic floor muscle strength between two groups

	Time	Control group (n=48)	Study group (n=49)	t	P
PFDI-20	Before treatment	168.96±47.66	168.01±48.68	0.097	0.933
	3 months after treatment	27.41±4.16*	28.41±5.16*	1.051	0.296
	6 months after treatment	22.64±5.73*	16.32±3.08*,#	6.746	0.000
Muscle voltage of pelvic floor muscle fiber type I (UV)	Before treatment	3.11±0.63	3.27±0.64	1.241	0.218
	3 months after treatment	4.43±0.98*	6.40±1.13*,#	9.178	0.000
	6 months after treatment	5.33±1.17*	8.92±1.53*,#	12.997	0.000
Muscle voltage of pelvic floor muscle fiber type II (UV)	Before treatment	4.88±0.75	4.73±0.74	0.991	0.324
	3 months after treatment	5.02±0.61*	7.09±1.32*,#	9.947	0.000
	6 months after treatment	5.97±1.19*	9.16±2.21*,#	8.876	0.000

Note: Compared with before treatment, *P<0.001; study group compared with control group in the same period, #P<0.001.

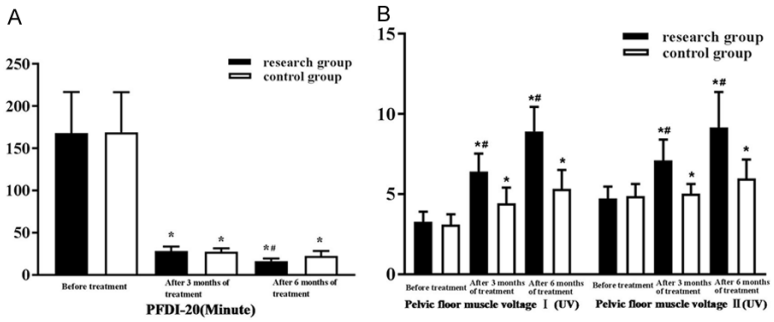

Figure 2. Comparison of PFDI-20 score and pelvic floor muscle strength in two groups. Compared with before treatment, *P<0.001; study group compared with control group in the same period, #P<0.001.

decreased, and patients with stage 0 and I increased significantly in both groups (all P<0.001). Six months after treatment, the number of patients with degree II in study group (2.04%) was significantly less than that in control group (16.67%, H=6.162, P<0.001), and the number of patients with degree 0 in study group (55.10%) was significantly more than that in control group (41.67%, H=3.792, P<0.001). See **Table 4**.

Expression of MMP-7 and TIMP-1 in ligament of two groups

The positive expression of MMP-7 and TIMP-1 in the ligaments was showed in both groups. The expression of MMP-7 in study group was lower than that in control group, but the expression of TIMP-1 in study group was higher than that in control group with significant differences (both P<0.001). See **Table 5** and **Figure 3**.

Discussion

PFD is a syndrome caused by multifactorial muscle strength decrease of pelvic floor supporting tissues, leading to pelvic organs displacement and dysfunction [12]. The pelvic floor supporting tissues mainly consists of cells and intercellular substance. Its main component is the extracellular matrix (ECM), whose synthesis and decomposition maintain dynamic equilibrium to keep the stability of the tissue structure and function [13]. The proteolytic system of ECM degradation is composed of matrix metalloproteinases (MMPs). In recent years, Bozkurt et al. have found that tissue inhibitor of metalloproteinases (TIMPs) are a family of multifunctional factors that can specifically inhibit the activity of MMPs; the interaction between TIMPs and MMPs as well as the content variation can affect the degradation of ECM, therefore, they are inseparable from the development of PFD [14]. The current operation methods for PFD are tram vaginal hysterectomy, vaginal anterior & posterior wall repair and high uterosacral ligament suspension, etc. However, study has found that after conventional surgery, there are still 30% to 40% PFD patients need a secondary surgery for failure in repair [15]. In particular, the risk of reoperation in elderly patients with weakened immune function is higher. In recent years, many pelvic floor repair and reconstruction surgeries were gradually applied in clinic with the rapid development of obstetrics and gynecology, the research of pelvic floor anatomy theory, the innovation of surgical instruments and the upgrading of pelvic floor material. Pelvic reconstruction can roundly cor-

Table 4. Comparison of POP-Q between two groups in different time

Group	Time	POP-Q				
		0	I	II	III	IV
Control group (n=48)	Before treatment	0 (0.00)	0 (0.00)	14 (29.17)	22 (45.83)	12 (25.00)
	3 months after treatment	16 (33.33)*	18 (37.50)*	11 (22.92)*	3 (6.25)*	0 (0.00)*
	6 months after treatment	20 (41.67)*	19 (39.58)*	8(16.67)*	1 (2.08)*	0 (0.00)*
Study group (n=49)	Before treatment	0 (0.00)	0 (0.00)	14 (28.57)	23 (46.94)	12 (24.49)
	3 months after treatment	18 (36.73)*	20 (40.82)*	7 (14.29)*	1 (2.04)*	0 (0.00)*
	6 months after treatment	27 (55.10)*,#	21 (42.86)*	1 (2.04)*,#	0 (0.00)*	0 (0.00)*

Note: Compared with before treatment, *P<0.001; compared with control group during the same period, #P<0.001. Six months after treatment, the number of patients with degree 0 and degree II between the two groups were compared (H=3.792, P<0.001), and the number of patients with degree II between the two groups were also compared (H=6.162, both P<0.001).

Table 5. Expression of MMP-7 and TIMP-1 in ligament of two groups

Group	Average gray value of MMP-7	Average gray value of TIMP-1
Control group (n=48)	87.76±3.74	84.63±3.29
Study group (n=49)	72.44±3.15	99.21±4.16
t	21.799	19.166
P	0.000	0.000

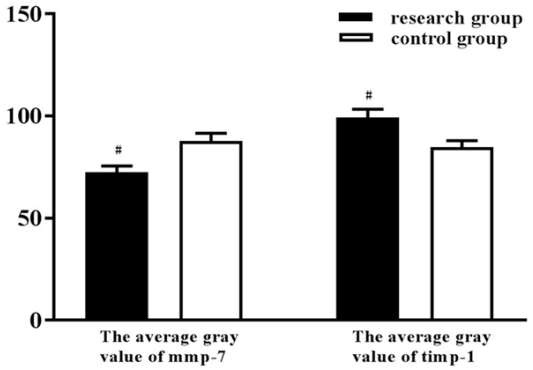

Figure 3. Expression of MMP-7 and TIMP-1 in ligament of two groups. Compared with control group, #P<0.001.

rect pelvic floor defects, then rebuild pelvic floor, also, it avoids obvious bulging of the anterior vaginal wall by strengthening the cardinal ligament of uterus and bladder cervical ligament. Comparing with the shortcomings of traditional surgery, such as vaginal distortions and anatomic displacement, the postoperative complications are significantly reduced. It was also reported that the use of Prolift system for pelvic floor reconstruction of PFD patients with postoperative follow-up more than 12 months could reach a high cure rate of 90-95% [16]. Study showed that Prolift pelvic floor repair system implanting transvaginal mesh for the treatment of PFD, although the surgical procedure could be simplified, could result in serious complications such as mesh exposure, dysuresia and infections in later period due to inadequate knowledge about indications, inadequate training of surgeons or improper operation [17]. But mesh implant has good effects on elderly patients with PFD.

In addition to surgical treatment of PFD, ancillary pelvic floor muscle rehabilitation training is equally important. In recent years, electrical stimulation as a biomimicry tool has become more and more popular in gynecology; current at different frequencies was used to stimulate the pelvic floor muscles and to dominate nerves, aiming to enhance pelvic floor muscle contraction and nerve function recovery [18]. Study of Gigliobianco et al. has shown that biofeedback + electrical stimulation + rehabilitative instrument training has advantages like easy operation, painless and small trauma for pelvic floor muscle rehabilitation exercise and has exact effects on the prevention and treatment of PFD [19]. Pelvic floor rehabilitation instrument has been widely used in clinic. It prevents damage of pelvic floor support structure, reduces pelvic pain, restores pelvic floor muscle function, and consolidates the efficacy of surgery. In this study, the pelvic floor rehabilitation instrument Phenix USB4 was used, through the gradually treatment of biofeedback + electrical stimulation, voltage of pelvic floor muscle fiber type I and II was increased, and pelvic floor muscle tension was gradually

restored. The results of this study showed that the operation time, exhaust time, indwelling time of urinary catheter and blood loss in study group were significantly less than those in control group; 6 months after treatment, the PFDI-20 score and relapse rate in study group were significantly lower than those in control group. Three and 6 months after treatment, pelvic floor muscle strength of study group was significantly better than that of control group, and the improvement of POP-Q in study group was also significantly better than that in control group. Our results indicated that pelvic floor reconstruction combined with pelvic floor rehabilitation instrument for the treatment of PFD could promote postoperative recovery and significantly improve pelvic floor muscle strength of patients, which was consistent with the study results of Gigliobianco et al. [19].

The results of this study also showed that in study group, the expression of MMP-7 was significantly lower than that in control group, but TIMP-1 expression was significantly higher than that in control group, indicating that the combination therapy could increase the content of TIMP-1, inhibit the increase of MMP-7, which was inconsistent with the findings of Bozkurt et al. [14]. The decrease of MMP-7 content makes it less effective, so that ECM synthesis is greater than decomposition, leading to normal structure and function recovery of pelvic floor connective tissues, and promoting the recovery of pelvic floor muscle function [20]. However, it is noteworthy that the high incidence of PFD has not been valued by many women; the obsolete concept coupled with inadequate understanding of the disease cause patients fail to seek timely medical treatment, although doctors advise pregnant women to use pelvic floor rehabilitation instrument, the actual use ratio is unfavorable; meanwhile, compared with more developed countries and regions, the diagnosis and treatment of this disease in some developing countries are still lagging behind; conventional clinical examination only knows about the external shape changes, but cannot get accurate information of pelvic floor anatomy and functional changes in deep tissues, which is also the bottleneck of scientific treatment guidance for patients [21]. For the first time, this study explored the treatment effect of pelvic floor reconstruction combined with pelvic floor rehabilitation instrument for PFD on the MMP-7, TIMP-1 expression in uterosacral ligament tissues. The interaction of the two enhanced the degradation of ECM, which is also an important cause of PFD. The study provides a new way of thinking and direction for the prevention and treatment of PFD in molecular perspective. But this study did not make statistical analysis on postoperative complications between the two groups, such as mesh exposure, dysuresia and infections. This may result in a slight insufficiency of surgical indications, which deserves further study.

In summary, pelvic floor reconstruction combined with rehabilitation instrument for the treatment of PFD has shorter operation time, less blood loss and lower relapse rate, and it can effectively restore pelvic floor muscle strength. But clinical surgery needs to strictly follow the indications, and clinical choice need to make comprehensively based on patient's age, degree of prolapse, economic status, pregnancy requirements and other actual conditions.

Disclosure of conflict of interest

None.

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