

Autologous adipocyte graft in endoscopic treatment of vesico-renal reflux in children: a preliminary study

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Abstract

No bulking agent is ideal for endoscopically treating vesico-renal reflux in children. Many teams have tried to find a safe and efficient material, ideally an autologous material. We describe here a protocol for the use of autologous viable fat in the treatment of primary vesico-renal reflux in children aged from 3 to 15 years. Fat harvesting was done from the medial side of the thigh by manual aspiration. Samples were centrifuged to purify the graft from blood and lipid. Lastly fat was injected beneath the pathologic ureter by a conventional endoscopic technique. A voiding cystourethrography (VCUG) closed the procedure. Follow-up included renal ultrasonography the day after surgery, and one and three months later. A VCUG was performed systematically at three months and, in cases of acute pyelonephritis, during the survey.

Sixty-four children with 94 refluxing units were treated by autologous fat injection with a follow-up from 6 to 40 months. At the end of the procedure, we systematically obtained a very good increase in height of the pathologic meatus and VCUG was normal in all cases. None presented with an obstruction during the follow-up period. Two children presented with an acute pyelonephritis before the third month. At three months, VCUG was not realized in 14 cases (22%) because the parents refused the procedure. One of those children presented with an acute pyelonephritis five months after endoscopic treatment. VCUG was normal for 17 of 50 children (34%), and showed a real improvement for 19 other children (38%). Three children had a surgical reimplantation because of the persistence of an unchanged high-grade vesico-renal reflux; histological examination found viable adipocytes on sections of the distal pathologic ureter. Clinically, 11 children (17%) presented with an acute pyelonephritis after treatment at a mean follow-up time of 10 months.

These preliminary findings led us to modify the technique in order to improve our results. Our first concern is feasibility and

safety of this technique, regardless of the use of other synthetic bulking agents the innocuousness of which is uncertain.

Introduction

Acute pyelonephritis (APN) is a common problem in childhood and is associated with vesico-renal reflux (VRR) in 25-40% of children.¹ Persistent VRR and repeated pyelonephritis may predispose to renal scars and chronic renal failure with an end-stage renal failure in up to 3% of cases.¹ Defining a standard treatment for VRR in children is not easy because of the lack of reliable evidence. Numbers of studies prospectively or retrospectively compare medical, endoscopic, and surgical treatments.²⁻⁴ Moreover, numbers of low-grade VRR have a spontaneous resolution.

Surgical techniques are effective but invasive and not free of complications while medical treatment is dependent on close observation. The introduction of endoscopic techniques permits the prevention of new APN and renal failure by VRR elimination. Since the beginning of the use of endoscopic treatment, several bulking agents have been proposed. The ideal agent should be easy to inject, stable in time, and safe,⁵ and at least as efficient as the actual bulking agent. The use of Teflon was the first success of endoscopic treatment, despite the problem of migration.⁶⁻¹¹ Since then many substances have been proposed, autologous or exogenous, resorbable or not. However, none was ideal and no solution was found for the problem of biocompatibility or long-term stability.^{12,13} Using adipose tissue as a bulking agent is an ancient practice in plastic surgery and, in the last century, Coleman introduced a new technique called "lipostructure".¹⁴ This technique has led to a growing interest in its use in the restoration of all volume defects in plastic surgery because of the stability of the graft. We proposed to apply this technique to VRR management in children, thus combining innocuousness and efficacy.

Materials and Methods

To be included in the study, patients must be aged from 3 to 15 years and have primary VRR stage II or III. They must not have had endoscopic treatment or localized surgery in the past. We proposed this technique after failure or refusal of medical treatment. A dimercaptosuccinic acid (DMSA) scan was done preoperatively in cases of recurring APN or if renal scars were seen on ultrasonography (US). Oral and written information on

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the technique were given systematically. If the legal representative of the child refused, or if the child did not fulfill all conditions, the child was excluded. If the parent/guardian agreed with this technique and if the child met all the conditions of inclusion, consent was signed.

The first step of the procedure was fat harvesting of the patient's own adipose tissue. The donor site was the medial side of the thigh (Figure 1). The patient was in a gynecological position on the operation table and the procedure was performed under general anesthesia. We needed a cannula and a 10 mL syringe for a standard lipoaspiration. Purification of the autologous fat graft was obtained by centrifugation (Figure 2), after which we separated the intermediate layer containing viable adipocytes and their fibrovascular stroma from blood (inferior layer) and lipid (superior layer). The second step of the procedure was the endoscopic treatment of VRR. Adipocytes were transferred in several 1 mL syringes, which were placed successively in a ratchet gun. The technique for subureteral injection was standard using a needle through the endoscope (Figure 3). The graft was injected below the ureteral meatus, positioned ideally according to the STING procedure. To close the procedure, we systematically carried out an immediate voiding cystourethrography (VCUG) to complete the injection in case of persistent reflux. Patients had renal US the day after surgery to detect any obstruction, and one and three months later. Efficacy was confirmed on VCUG three months after the procedure when an adipocyte graft is considered as stable. Any APN during the follow-up period necessitated VCUG and DMSA. A DMSA was not systematically done during the follow-up period but only in cases of a new APN within two months after infection, or on the appearance of renal scars on US.

Results

Currently, 64 children with 94 refluxing units have benefited from this technique with a follow-up from 6 to 40 months. We treated 57 girls and 7 boys with a mean age of 7.8 years. Thirty of them had a bilateral VRR. All VRRs were diagnosed after one or several APNs. We decided to treat only VRR grade II or III. Three patients had a complementary injection during the procedure because of a persistent reflux on VCUG.

Of the patients, 50 had VCUG at three months (78%), of which 17 (34%) were completely cured on VCUG, 19 (38%) were improved with the persistence of a minor radiological reflux but no recurrent infection during the follow-up period. Thirteen patients (26%) refused VCUG at three months. Among them, one patient presented with APN five months after the procedure. In three cases (6%), VCUG was exactly the same as before the treatment. As these patients had severe pyelonephritis with renal scars before treatment, we decided to do a surgical reimplantation. Viable adipocytes without any necrosis were found microscopically. For the 11 other children with VCUG at three months, improvement was considered as minor (22%). Of the refluxing units, 41% were cured, 24% were improved, 18% were unchanged, and 17% were not explored by VCUG at three months. US was done at one and three months and has enabled confirmation of the persistence of the bulking agent even if its volume seemed to be lower than on the day after surgery.

Clinically, among the patients without VCUG at three months, one had APN at five months and refused any exploration or treatment of a possible reflux. Among the patients cured on VCUG at three months, three (17.6%) had APN during the follow-up (two at one year and one at two years). Only one patient accepted VCUG, which was normal. Since then, follow-up was uneventful. Among the patients who improved by this treatment, three (15.7%) had APN six to 12 months later and two of them had a second endoscopic treatment with another bulking agent. Finally, among the failures of this technique (14 patients), four (28%) had APN, three had a reimplantation because of renal scars before treatment, and two had a second endoscopic treatment with another bulking agent. None of the children had an obstruction or new renal scar on renal US or on DMSA scans when done. No complication, particularly no pain or bleeding, concerning the donor site of fat was reported. The majority of the children and their legal representatives were satisfied with the technique and result.



Figure 1. Donor site on the medial side of the thigh.



Figure 2. Purification by centrifugation.



Figure 3. Endoscopic injection.

Discussion

The introduction of fat as a bulking agent in surgery dates from 1889 when Van Der Meulen¹⁵ treated a diaphragmatic hernia with epiploon. Using autologous fat in surgery became common from 1910 to 1977.¹⁶⁻¹⁹ Illouz

introduced lipoaspiration in 1977, and great progress was the introduction by Coleman of the notion of atraumatic manipulation of fat. In 1997 he published his technique called lipostructure: the harvesting, purification by centrifugation, and reinjection of fat.¹⁴ After this, it was evident that autologous fat grafts need gentle manipulation of the tissue to pre-

serve the viability of the adipocytes. Since then, many more surgeons have tried this technique in their fields of competence.²⁰ The main argument against fat grafting was its lack of stability, but MacRae *et al.* have demonstrated that centrifugation and gentle manipulation of fat had no significant effect on fat viability.^{21,22}

Since the beginning of endoscopic treatment of VRR, many bulking agents have been tried. Several studies have compared different agents to determine the ideal one but none combined all the conditions to become the standard. Simultaneously, Santiago *et al.* and Matthews *et al.* used autologous fat for the treatment of urinary incontinence.^{23,24} Results were not satisfying owing to instability issues, but they did not apply Coleman's technique. Furthermore, no study mentions this technique of autologous adipocyte grafts in endoscopic treatment of VRR.

Our unit has applied the endoscopic treatment of VRR since 1983. Until 2000, we used Teflon; since 2001, we have preferred polydimethylsiloxane. Our clinically good results are around 85%, as reported in the literature. After the problems of Teflon migration we examined the stability of this resorbable agent. For this reason we considered another bulking agent which could defer biocompatibility, stability, efficiency and cost-effectiveness. Concerning biocompatibility, this technique is guaranteed because we use autologous tissue. As regards stability, studies have proved a radiological stabilization of about 50% of the initial volume three months after injection.²⁵ Histological analysis of grafts has shown persistence of mature adipocytes^{26,28} several years after surgery. Lastly, autologous adipocyte grafts allowed money-saving by a reduction of disposables.

This preliminary study shows 34% of good results but 64% of persistent reflux (improved or unchanged) with a few cases of new APN but no new renal scars. This is satisfactory clinically but we have to improve our radiological results. No complication has been diagnosed on US and clinical follow-up. We agree with the fact that radiological classification is not satisfactory with regard to VRR but, for the moment, it is the only objective criterion to evaluate the efficiency of a new bulking agent. We decided to treat VRR staged II or III because of the large evidence base for endoscopic treatment in the advanced stages of VRR. For this study, we injected a mean of 0.4 mL of fat in one site, which seems to be insufficient. Because of volume loss we have to inject at least 1 mL of fat in several sites under each pathologic ureter. Indeed, viability of the adipocyte graft is compromised in the case of too voluminous a graft, when the large area of vascularization of the middle of a voluminous graft leads to apoptosis of the cells.¹⁸ Moreover,

we decided to give up the STING procedure for intraureteral injection after hydrodistension of the ureter. In this way we began a second cohort of patients and we improved our clinical and radiological results. However, the follow-up period is too short to conclude that the improvement is statistically significant.

Conclusion

Autologous adipocyte grafts for endoscopic treatment of VRR in children is a potentially satisfactory approach. It is feasible, safe, and provides good results on the mid-term follow-up. The biocompatibility of the autologous adipocyte graft and the theoretical long-term stability of this viable graft lead us to publish our research findings. We recently improved our technique by injecting more autologous adipocytes in several points in the ureter after its hydrodistension. We are waiting for results from this second cohort. Although it is too early to draw conclusions, our first results do not lead us to suggest that the autologous adipocyte graft is any closer to the ideal bulking agent.

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