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Revascularization of the testis using a vascular induction technique: a potential approach for staged orchiopexy in high-undescended testis

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Abstract The present study was designed to determine whether a fasciovascular flap as a vascular carrier could be used to revascularize the undescended testis for avoiding the hazardous effects of the Fowler-Stephens procedure, high division of the spermatic vessels, and for bringing high-undescended testes into the scrotum. A total of 25 Wistar rats were divided into five groups of five rats each. In each group, surgical procedures were performed bilaterally, i.e. ten testes in each group, as follows: sham-operated controls (group 1), undescended testes (group 2), high division of the spermatic vessels (group 3), vascular induction with immediate division of spermatic vessels (group 4), and with delayed division of spermatic vessels (group 5). Evaluations were done by measuring the testicular weight and volume, testicular blood flow, and testicular biopsy scores and by microangiography. A moderate to severe decrease in testicular weight and volume in all experimental groups was observed compared with the sham-operated controls (group 1), but this was significantly less in groups 2 and 5. High division of the spermatic vessels in groups 3 and 4 resulted in a significantly greater decrease in the

Microangiographically, an impaired vascular supply from the deferential artery in group 3 and insufficient revascularization from the fasciovascular carrier in group 4 were observed. However, efficient revascularization stemming from the superficial epigastric artery of the fasciovascular flap was found in group 5. The testicular biopsy scores of groups 2 and 5 were significantly greater than those of groups 3 and 4. The results of the present study demonstrate that the fasciovascular flap as a vascular carrier revascularizes the testis through spermatic vessels after delayed division and provides an adjuvant treatment modality or first-stage procedure in a salvage operation for high-undescended testis during staged orchiopexy.

testicular blood flow, but this did not occur in group 5.

Keywords Undescended testis · Vascular induction · Fasciovascular flap · Revascularization · Staged orchiopexy

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Introduction

Undescended testis, which is generally synonymous with cryptorchidism, is the most frequent congenital disorder of an endocrine organ. Some of the patients with undescended testis (8–14%) have a testis that is too high for conventional orchiopexy techniques [20]. When high retroperitoneal dissection does not provide adequate spermatic vessel length to allow for descent without tension, orchiopexy by high [17] or low [27] spermatic vessel division techniques, staged orchiopexy [8, 15, 34], orchiectomy [21], and microvascular autotransplantation [23, 31, 41, 42] have been suggested for bringing high-undescended testes into the scrotum; however, the most popular method of surgical treatment remains orchiopexy using a high spermatic vessel division technique (Fowler-Stephens procedure).

A compilation of published data indicates that when patients are selected carefully, the procedure is successful in a high percentage of cases. Authors who report success with the Fowler-Stephens procedure have utilized gross appearance (size and consistency) of the testis as the criterion of success or failure [4, 7, 19, 28], and biopsy has been performed only in a very few cases. The reported long-term success rates with the standard Fowler-Stephens procedure are never more than 62.6–68.5% [9]. Ligation of the spermatic vessels aids in placing these high testes in the scrotum, but has been attended by testicular atrophy in 20–50% of cases [2, 26]. The 2-stage Fowler-Stephens orchiopexy offers a theoretical advantage over the 1-stage procedure. In the first stage, the testis and spermatic cord are mobilized as much as possible so that the spermatic vessels can be ligated. At a later date, preferably after 1-2 years, a staged Fowler-Stephens orchiopexy can be performed [35]. Nowadays, most surgeons perform the first stage laparoscopically, with clipping of the spermatic vessels intraperitoneally [29]. It seems more logical that this approach, with minimal manipulation of the testis at the first intervention, has the best chance of allowing testicular growth to proceed undisturbed until the second intervention. It has been hypothesized that following ligation of the spermatic vessels, the collateral blood supply to the testis will compensate for the loss sustained from ligation of the spermatic vessels and a time efficient blood supply will be provided to the gonad to preserve its viability and function. While this hypothesis on initial vessel ligation seems reasonable from a theoretical viewpoint, there are few clinical and experimental data to substantiate it [29, 35]. When compared with the 1-stage Fowler-Stephens procedure, the 2-stage operation yields slightly better results, with an overall success rate of 76.8% [9]

The time-honored observation of the "delay procedure" [5] combined with a greater understanding of the biology of "angiogenesis" [16] has led to the concept of tissue revascularization and prefabrication from a transferred vascular carrier. When a tissue is not situated on a convenient vascular pedicle, it is sometimes possible to revascularize it by juxtaposing an appropriate vascular carrier to provide an efficient blood supply (Fig. 1). The first step is the surgical juxtaposition of a vascular carrier at a suitable donor site with the target tissue. After a period in which revascularization of the target by the carrier occurs, the original blood supply to the target can be ligated, and the target can be carried as a vascularized tissue by the vascular carrier. The prerequisites for an ideal vascular carrier to revascularize other tissues are high vascularity, a pedicle of adequate length and caliber, limited bulk, acceptable donor site morbidity, and predictable vascularization ability [25]. Although the mesenteric vascular pedicle [12, 13], vascular bundle [10, 11, 22], and musculovascular pedicle [14, 39] have been demonstrated as potential vascular carriers for revascularization, a deep fascia or a septal fascia with a fine vascular network, thin architecture, and a long vascular pedicle is usually an excellent carrier [3, 6, 45].

In the present study, we aimed to determine whether an arteriovenous bundle and its surrounding fascia (i.e., the epigastric fasciovascular flap) could be used as a

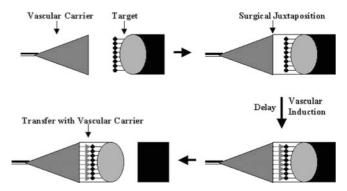


Fig. 1 Schematic depiction of the vascular induction technique. The vascular carrier is surgically juxtaposed to the target. After a delay procedure with resultant angiogenesis, the target can survive based on the blood supply derived from the vascular carrier

vascular carrier to revascularize the undescended testis as a salvage procedure and the effect of immediate and delayed division of the spermatic vessels on the revascularization rate from the transferred exogenous vascular carrier.

Materials and methods

Animals

Adult, male Wistar rats weighing 350–400 g were used for the experiments because their testicular vascularization is similar to that of humans [1, 30, 46]. The Japanese National Research Council guide for the care and use of laboratory animals was followed during the experimental study. Anesthesia was obtained with sodium pentobarbital (50 mg/kg) injected intraperitoneally. The animal was placed in a supine position. The abdominal, inguinal, and scrotal areas were depilated bilaterally using a commercial depilatory cream and cleansed with povidone-iodine solution.

Design of the experiment

A total of 25 animals were randomly separated into five groups of five rats each. In each group, surgical procedures were performed bilaterally, i.e. ten testes in each group. An operating microscope (magnification 10×) was used during the surgical procedures.

Group 1: sham-operated controls

The animals were anesthetized and the abdominal, inguinal, and scrotal areas were depilated bilaterally. The testis, the vas deferens, and the spermatic, deferential, and cremasteric vessels were exteriorized through a horizontal lower abdominal transperitoneal incision of about 1.5 cm on the inguinal canal and placed into their original position, but the testes had no surgical intervention and served as controls. The skin incision was sutured with 4–0 nylon.

Group 2: undescended testis model

An intra-abdominal undescended testis model in rats was created to simulate high-undescended testis. The testis, the vas deferens, and the spermatic, deferential, and gubernacular vessels were exteriorized, and the testis was released by ligation and division of the vessels from the cremasteric muscle. The testis, which is supplied by spermatic and deferential vessels, was withdrawn into the abdomen via the inguinal canal. The external ring of the inguinal canal was closed with 5–0 polyglactin and the skin incision was sutured with 4–0 nylon.

Group 3: the Fowler-Stephens procedure

The undescended testis model was performed as in the group 2. Additionally, the spermatic vessels were exposed retroperitoneally. High ligation and division of the spermatic vessels (the Fowler-Stephens procedure), above the iliac vessels and away from the deferential vessels, was carried out, and then the testis was withdrawn into the abdominal cavity. The external ring of the inguinal canal was closed with 5–0 polyglactin and the skin incision was sutured with 4–0 nylon.

Group 4: vascular carrier and immediate division of the spermatic vessels

The undescended testis model was performed as in the group 2. After immediate high ligation and division of the spermatic vessels, (the Fowler-Stephens procedure) as in group 3, the epigastric fasciovascular pedicled flap was harvested and the spermatic vessels were wrapped by the fasciovascular flap as follows (Fig. 2A–F): the epigastric fasciovascular pedicled flap based on the superficial epigastric vessels, which was described by Tark et al. [45], was used as a vascular carrier for vascular induction (Fig. 2A). Rats were examined through bilateral inguinal incisions. The superficial epigastric vascular bundle and surrounding 2.5×4-cm patch of fascia were dissected free from the underlying groin muscles. The superficial epigastric pedicle was ligated distally as it entered the lateral abdominal wall. The epigastric fasciovascular pedicled flap was elevated based on the superficial epigastric vessels to revascularize the testis (Fig. 2B). The pampiniform plexus, which is the convoluted part of the distal spermatic vessels, proximally 2 cm away from the testis, was placed on the center of the flap. Both free edges of the flap were sutured each to the other longitudinally and also fixed onto the perivascular adipose tissue surrounding the pampiniform plexus with 6-0 nylon stitches laterally (Fig. 2C), and then the testis was withdrawn into the abdominal cavity (Fig. 2D).

Group 5: vascular carrier and delayed division of the spermatic vessels

The undescended testis model as in the group 2 and wrap-around procedure of the pampiniform plexus by the epigastric fasciovas-cular pedicled flap as in the group 4 were carried out (Fig. 2A–F). After a 2-week delay, the spermatic vessels were exposed retroperitoneally under general anesthesia in a second stage and high ligation and division of the spermatic vessels, above the iliac vessels and away from the deferential vessels, was performed.

Evaluation

Testis volume and weight

At the end of the fourth postoperative week, the weight and volume of the each testis was measured after dissection and removal of the attached tissues from the testis.

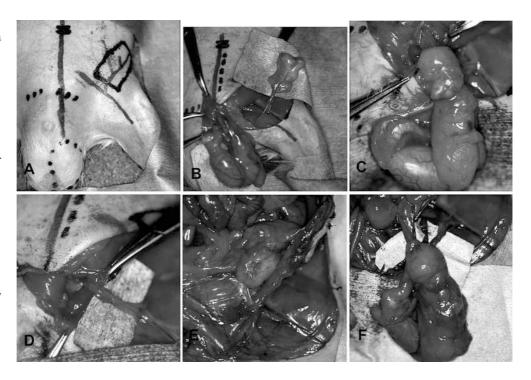
Testicular blood flow

A laser Doppler flowmeter (ALF 21 N, Advance Laser Flowmeter, Tokyo, Japan) was used to record the testicular blood flow for 10 min in all groups just before the surgical procedure (baseline), 1 h after the surgical procedure, and finally at the end of fourth week during exploration. Measurements were obtained by placing a laser Doppler probe on the exposed aspect of the testis where no visible large arteries or veins were present.

Microangiography

A microangiography technique was used to visualize the vasculature and revascularization of the testis. After the animals were killed with an overdose of sodium pentobarbital, the abdomen and thorax were opened in the midline, and a polyethylene catheter (20 G) was inserted into both the thoracic aorta and the vena cava inferior and secured by a ligature to avoid leakage. Before injection of the radiopaque, 200 ml warm saline (30–40°C) including 1 ml

Fig. 2 The epigastric fasciovascular pedicled flap as a carrier for vascular induction: A outlining of the superficial epigastric vascular bundle and surrounding 2.5×4-cm patch of fascia. B The fasciovascular carrier harvested to wrap the distal part of the spermatic vessels, the plexus pampiniformis. C Wrapping procedure by the fasciovascular carrier completed, and then **D** withdrawing the testis into the abdomen via the inguinal canal. E After immediate or delayed division of the spermatic vessels and consequent delay procedure, exploration laparatomy of the intra-abdominal testis. F Appearance of the revascularized testis supplied by both the superficial epigastric and deferential vessels



heparin (5,000 UI) was used to perfuse the area of investigation, infusing via the thoracic aorta just below the diaphragm. A mixture consisting of lead oxide (50 g), gelatin powder (1 ml), and water (25 ml) heated to 50°C in the saline bath, was stirred vigorously in a plastic disposable bowl, and then injected in a pulsatile manner into the aorta, achieved by gently pressing and then releasing the plunger of the syringe to simulate the arterial pulse. When the injection was completed, the testes with related tissues and vessels were dissected and removed from the area and then fixed on a paperboard (Fig. 3A). The material was stored in the refrigerator at 4°C for 1 h. Radiographs were taken with an exposure of 50 mA and 64 kV at 140 cm between the film and X-ray source.

Histology

The testes were fixed in 10% buffered formalin, embedded in paraffin, sectioned at 3 µm, and stained with hematoxylin and eosin. All specimens were coded to guarantee blinded determination of the histological features. An independent pathologist evaluated and compared histological changes after randomly examining many specimens and achieving standardization of grading. The maturity of the germinal epithelium of the seminiferous tubules was graded according to the Johnsen testicular biopsy score (TBS) [24]. In each testicular biopsy section under a $25\times$ objective, 25-50 tubules were evaluated, and each tubule was given a score from 1-10 according to the criteria shown in Table 1. The mean TBS for each testis was calculated.

Statistical analysis

All values were expressed as mean \pm SEM. A one-way analysis of variance was used to determine significant differences between groups, and when a difference was found a Tukey's test (all pairwise multiple-comparison test) was used to identify the differences. SigmaStat statistical software was used.

Fig. 3 A The testis with related tissues and vessels fixed onto a paperboard after injection of the radiopaque. B Microangiograph showing the shamoperated control testis (group 1) and its original vascular supply. C Well vascularized testis by the spermatic and deferential vessels in group 2. **D** Impaired vascular supply and collateral support to the testis from the deferential artery in group 3. E Insufficient revascularization from the fasciovascular flap in group 4. F Efficient revascularization (asterisk) stemming from the superficial epigastric artery of the fasciovascular flap in group 5. The testis (t), the deferential (da), spermatic (sa), superior and inferior epidydimal (ea), the gubernacular-cremasteric (ga), femoral (fa), and superficial epigastric (sea) arteries

Table 1 Testicular biopsy score. If the germinal epithelium is disorganized with sloughing or obliteration of the lumen, a lower score is given

Score	Description
10	Complete spermatogenesis with many spermatozoa
9	Only few spermatozoa present (five per tubule)
8	No mature spermatozoa but late spermatids present
7	Many spermatids without any sign of differentiation
6	Only few spermatids present (five per tubule)
5	Many spermatocytes present
4	Only few spermatocytes present (five per tubule)
3	Spermatogonia are only germ cells present
2	No germ cells but Sertoli's cells present
1	No cells in tubular section

Results

All rats survived without any complications and were evaluated.

Testicular weight and volume

A moderate to severe atrophy in the testes of all experimental groups (groups 2–5) was observed when compared to sham-operated controls (group 1), but it was significantly less in groups 2 and 5 (Table 2). Intra-abdominal transposition of the testes (group 2) resulted in a significant decrease in mean testicular weight and volume

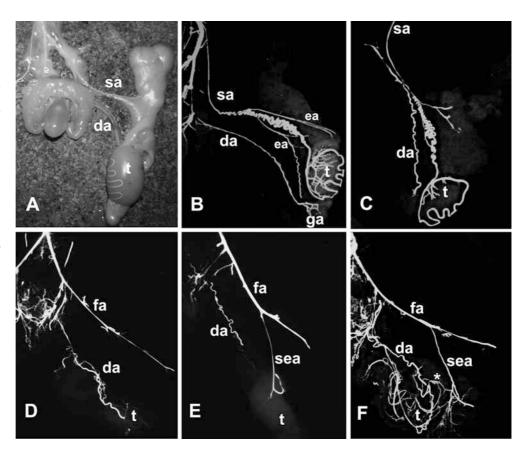


Table 2 Testicular weight and volume. Results are given as mean \pm SEM

Groups	Weight (mg)	Volume (mm ³)
1 2 3	$1,728 \pm 59.9$ $1,466 \pm 63.6$ 871 ± 42.8	$1,970 \pm 65.8$ $1,615 \pm 55.6$ $1,150 \pm 63.6$
4 5	$902 \pm 37.9 \\ 1,264 \pm 65.8$	$1,120 \pm 69.4 \\ 1,440 \pm 87.6$

(P < 0.05 and P < 0.005, respectively). There was no significant differences between groups 2 and 5 or between groups 3 and 4 (P > 0.05); however, group 5 had a significantly greater testicular weight and volume than both groups 3 and 4 (P < 0.005 and P < 0.05, respectively).

Testicular blood flow

Laser Doppler flowmeter results revealed no significant differences in baseline testicular blood flow between the groups (P > 0.05) (Table 3). Only division of the gubernacular-cremasteric vessels caused a significant decrease in testicular blood flow at the first postoperative hour in both groups 2 and 5 when compared with the baseline values (P < 0.005 and P < 0.05, respectively), despite no significant difference between groups 2 and 5 (P > 0.05). Comparisons of mean testicular blood flow within groups demonstrated that immediate ligation and division of both the spermatic vessels and the gubernacular-cremasteric vessels in groups 3 and 4 resulted in a significantly greater decrease (an average of 80%) in blood flow at the first postoperative hour, when compared with the baseline values (P < 0.0001). At the end of the fourth postoperative week, the testicular blood flow in group 2 increased from the low levels found at the first postoperative hour to above baseline values. However, low blood flow values in groups 3 and 4 were persistently lower than the baseline values (P < 0.0001), even though there was a slight increase in blood flow during the 4-week period. The testicular blood flow in group 5 was decreased to 46.57% of the baseline value after delayed division of the spermatic vessels; however, it was still significantly greater than those found in groups 3 and 4 (P < 0.005) (Table 3).

Microangiography

The original vascular supply of the testis through the spermatic, deferential, and cremasteric arteriesis shown for group 1 (sham-operated controls) (Fig. 3B). In group 2, the testis was well vascularized by the spermatic and deferential arteries (Fig. 3C); however, an impaired vascular supply to the testis from only the deferential artery was observed in group 3, and no anastomosis between the deferential and the pampiniform plexus providing collateral blood supply was found (Fig. 3D). In group 4, revascularization of the testis from the fasciovascular carrier, shown by microangiography, was found to be insufficient (Fig. 3E). In group 5, efficient

Table 3 Testicular blood flow. Results are given as mean \pm SEM

Group	s Baseline (ml/min/100 g)	1	Postoperative fourth week (ml/min/100 g)
1	20.3 ± 1.1	18.7 ± 0.9	21.2 ± 1.4
2	18.9 ± 1.4	12.0 ± 0.7	19.6 ± 1.3
3	19.5 ± 1.2	3.9 ± 0.3	7.5 ± 0.8
4	20.1 ± 1.4	4.3 ± 0.4	7.8 ± 0.7
5	20.4 ± 1.3	12.7 ± 0.8	10.9 ± 0.8

revascularization stemmed from the superficial epigastric artery of the fasciovascular flap, and a retrograde filling of the testicular vascular network was observed (Fig. 3F). However, anastomoses between the very fine collaterals were sometimes impossible to demonstrate, because the thicker medium would not reach the very fine radicles of the arterial tree that appears as a filling defect.

Histology

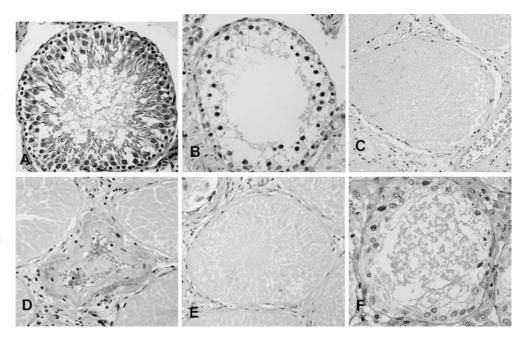
Varying degrees of histologic change were observed in the testes of experimental groups when compared with the sham-operated controls (Fig. 4A–F). In group 2, there was a normal testicular architecture in two testes; but less orderly, non-cohesive germinal cells, and spermatogenic arrest in seven testes, while in one testis disordered, sloughed germinal cells with closely packed seminiferous tubules were found (Fig. 4B). In group 3, variable percentages of coagulative necrosis (less than 50% in four testes and more than 70% in six testes) in the central portion of the testicular parenchyma were observed (Fig. 4C), and the peripheral compartment had fibrosis in the interstitium and less distinct seminiferous tubule borders without spermatogenesis. Despite obvious infarction of the seminiferous tubules, blood vessels could have contained blood cells even if the central portion of the parenchyma and interstitial cells were intact. Thickening of the arterial wall with concomitant hyalinization was another typical feature (Fig. 4C, D). In group 4, similar findings to group 3 were observed (Fig. 4E). In group 5, most of the testes showed spermatogenic arrest and tubular damage without infarction, although some focal necrotic areas were present (Fig. 4F).

The difference in mean TBS between the sham-operated control and experimental groups was significant (P < 0.0001) (Fig. 5). There was no significant difference between either groups 2 and 5 or groups 3 and 4 (P > 0.05); however, the mean TBS of groups 2 and 5 were significantly higher than those of groups 3 and 4 (P < 0.0005).

Discussion

The concept of orchiopexy by high spermatic vessel division was popularized by Fowler and Stephens in 1959 when they demonstrated collateral arterial blood

Fig. 4 A Sham-operated control testis (group 1) demonstrating normal spermatogenesis with many mature spermatozoa. B Spermatogenic arrest with the presence of a few spermatocytes in group 2. C, D Infarction of germinal epithelium even though some blood continued to circulate into interstitium. with thickening of the vessel wall in group 3. E Severe ischemia and necrosis of the seminiferous tubules in group 4. F Regardless of spermatogenic arrest and tubular damage, preserved germinal epithelium from ischemic necrosis in group 5 (original magnification ×540; H and E)



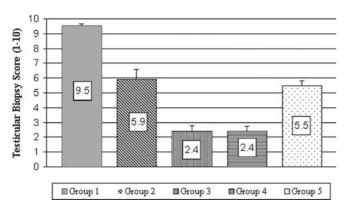


Fig. 5 Mean testicular biopsy scores of each group, showing significant differences between the sham-operated control and experimental groups (P < 0.0001), as well as groups 2 and 5 and groups 3 and 4 (P < 0.0005)

supply from the deferential artery by intraoperative arteriography. They also emphasized the value of testing the adequacy of the collateral blood supply by temporarily occluding the spermatic artery (the Fowler-Stephens test) [17]. Therefore, the viability of the testis after Fowler-Stephens orchiopexy depends on the collateral deferential blood supply. Patients who have failed the 10 min Fowler-Stephens test are the candidates for 2-stage orchiopexy or orchiopexy by microvascular revascularization. Several published and numerous anecdotal reports have indicated the success of microvascular autotransplantation [18, 23, 31, 41, 42]. On the one hand, the lack of general training in microsurgery and the lack of a readily available set up in these circumstances make it unlikely that the majority of highundescended testes will be treated by this technique. However, the critical testicular ischemia time has been

estimated as 1.6 h [18] and the duration of ischemia directly affects the degree of testicular hazard. A period of 1-h ischemia leads to moderate germinal epithelial destruction, whereas a 2-h ischemia period during microvascular revascularization of the testis results in a much more generalized and severe damage [44]. This procedure is also not feasible in the 1–2-year age group, when orchiopexy should ideally be performed. Initial reports using 2-stage Fowler-Stephens orchiopexy have indicated a high success rate which seems to offer an alternative in borderline circumstances [29, 35]. It has been proposed that this approach allows for enhanced collateral vessel development and for additional spermatic cord lengthening [35]. While this hypothesis for initial vessel ligation seems reasonable from a theoretical viewpoint, its scientific basis, with a time interval (1– 2 years after first operation) that allows for some additional lengthening of the spermatic cord and that increases collateral blood flow, is inadequate for substantiation. In addition, the 2-stage Fowler-Stephens procedure cannot be used in patients with a very short vas deferens.

Clinical studies to evaluate testicular viability and function after the spermatic vessel division have also been sparse and inconclusive because claims of positive results are generally based on gross the appearance of the testis [9]. However, our results and previous experimental studies [32, 36, 37, 38] show that testicular injury caused by ligation and division of the spermatic vessels is much more evident histologically than one would expect from gross examination of the testis. The testis is sustainable at low blood flow for a long time, until a sufficient collateral blood supply from the deferential vessels can be established [33]. Because of the avascularity of the seminiferous tubules, nutrition of the germ cells is provided by diffusion, which depends on

sufficient circulation to the interstitial space and on capillary permeability. It is likely, therefore, that after division of the spermatic vessels, the risk of damage to the seminiferous tubules may be higher than to the interstitium and tunica albuginea. Germ cells may not be perfused sufficiently, even though some blood continues to circulate into the interstitium after spermatic vessel division (Fig. 4C, D), or bleeding is evident after incising the tunica albuginea [38]. It therefore appears that the presence of bleeding from incised testes (the Fowlers-Stephens test) and gross appearance are not adequate to presume function and do not guarantee the viability of the testes.

To prevent the failure of the Fowler-Stephens procedure due to either the loss or inadequacy of the collateral blood supply, and to reorganize vascular supply and improve testis viability, it seems reasonable to revascularize the testis from a transferred intra- or extraabdomimal vascular carrier before division of the spermatic vessels during staged orchiopexy. Recently, Sönmez et al. investigated whether neovascularization of the testes through the spermatic vessels by an omental pedicle flap could be achieved. They reported that the testes in which Fowler-Stephens plus omentopexy were performed had much better intratesticular blood flow rates and testicular biopsy scores than the Fowler-Stephens procedure alone [43]. Shoshany et al. also investigated the effect of "omentotesticulopexy" in terms of angiogenesis to improve the vascularization of the cryptorchid rat testis. Despite the fact that they did not provide any histological or physiological data, they claimed that an improvement of the vascularization of high abdominal testes as well as conservation of testicular weight. They used angiography and testicular weight measurement to support their argument [40]. In the present study, the epigastric fasciovascular flap was chosen as a vascular carrier because of its high vascularity and large contact surface area, as well as its high ability for angiogenesis. It has been demonstrated that 93% of the target tissue is adequately perfused by the fasciovascular carrier by day 5 [45]. The epigastric fasciovascular pedicled flap has a greater arc of rotation because of its long pedicle and it can easily reach both the abdominal cavity and the scrotum. In the model used in our study, the pampiniform plexus was wrapped by the fasciovascular flap to increase the contact surface on a 2.5-cm portion of the vessels (Fig. 2A-F). However, Shoshany et al. [40] abraded a 1-cm² area of the tunica albuginea of the testis with sand paper and punched five to seven times with a needle to fix the omentum on the testis, hence the name "omentotesticulopexy". This procedure will disrupt the delicate vascular consistency of the testis and the surface cooling effect [1, 30, 46].

The present study shows that revascularization from the fasciovascular carrier after delayed division of the spermatic vessels can compensate and provide efficient blood supply loss sustained from the spermatic vessel division. Many investigators have demonstrated that a delay of between 7 and 21 days provides sufficient time

for angiogenesis from a vascular carrier [3, 5, 12, 13, 25, 45]. Based on this evidence, a 2-week period of delay was chosen for angiogenesis to develop following the creation of the epigastric fasciovascular flap prior to division of the spermatic vessels. Microangiography showed clear evidence of revascularization stemming from the level of contact surface between the fasciovascular flap and spermatic vessels and the vascular supply to the testis was preserved. Contrary to delayed division of the spermatic vessels, immediate division of the spermatic vessels does not have any supportive effect on the blood flow of the testes after the wrapping procedure of the spermatic vessels by the fasciovascular flap, and an impaired revascularization from the vascular carrier was observed. Therefore, we think that severe ischemia of the testis to be revascularized interferes with neoangiogenesis from the transferred exogenous vascular carrier.

It is well known that 38% of unilateral and bilateral undescended testes have completely lost their germ cells, caused by the impaired transformation of gonocytes [20]. The germinal cells are remarkably sensitive to hyperthermia resulting from body temperature. The scrotum performs an important thermoregulatory function by constantly altering the position of the testes. The complex convolutions of the spermatic artery known as pampiniform plexus also operate in conjunction with changes in position of the scrotum to either preheat or cool the blood [1, 30, 46]. In the case of highundescended testes, this compensatory mechanism does not work and/or cannot maintain the gonads at an optimal temperature, which is from 1-8°C below normal body temperature for normal function. In the present study, we found that intra-abdominal transposition of the testes (group 2) alone resulted in a significant decrease in both mean testicular biopsy scores and mean testicular weight and volume when compared with the sham-operated controls. In the literature, there is no clear-cut evidence that current treatment modalities for undescended testis improve fertility. Finally, our results affirm that revascularization of the testis by a fasciovascular carrier may be an alternative treatment modality or first-stage procedure in a salvage operation for high-undescended testis during staged orchiopexy, and that a further study is needed to evaluate the fertility—ability of male rats to impregnate female rats—at a later time (i.e., 4 or 6 weeks) after scrotal placement of the testes revascularized by the fasciovascular flap.

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References

- 1. Baker HJ, Lindsey JR, Weisbroth SH (1979) The laboratory rat. Academic Press, New York
- Bar-Maor JA, Nissan S, Lernau OZ, Oren M, Levy, E (1979) Orchidopexy in cryptorchidism assessed by clinical, histologic and sperm examinations. Surg Gynecol Obstet 148: 855

- Borud LJ, Shaw WW, Brunicardi FC, Mullen Y, Passaro EP (1996) Prefabrication of a neo-endocrine organ: a rat model. J Surg Res 61: 221
- 4. Brendler H, Wulfsohn MA (1967) Surgical treatment of the high undescended testis. Surg Gynecol Obstet 124: 605
- Callegari PR, Taylor GI, Caddy CM, Minabe T (1992) An anatomic review of the delay phenomenon: I experimental studies. Plast Reconstr Surg 89: 397
- Can Z, Apaydın İ, Erçöçen AR, Demirseren E, Sabuncuoğlu B (1998) Prefabrication of a high-density porous polyethylene implant using vascular induction technique. Ann Plast Surg 41: 264
- Clatworthy HW, Hollanbaugh RS, Grosfeld JL (1972) The "long-loop vas" orchiopexy for the high undescended testis. Am Surg 38: 69
- Corkery JJ (1975) Staged orchiopexy: a new technique. J Pediatr Surg 10: 515
- Docimo SG (1995) The results of surgical therapy for cryptorchidism: a literature review and analysis. J Urol 154: 1148
- Erk Y, Rose FA, Spira M (1983) Vascular augmentation of skin and musculo-cutaneous flaps. Ann Plast Surg 10: 341
- 11. Erol OO (1976) The transformation of a free skin graft into a vascularized pedicled flap. Plast Reconstr Surg 58: 470
- Erol OO, Spira M (1978) Omentum island skin graft flap. Surg Forum 29: 594
- Erol OO, Spira M (1980) Development and utilization of a composite island flap employing omentum: experimental investigation. Plast Reconstr Surg 64: 405
- 14. Erol OO, Spira M (1980) Secondary musculocutaneous flap: an experimental study. Plast Reconstr Surg 65: 277
- 15. Firor HV (1971) Two-stage orchiopexy. Arch Surg 102: 598
- Folkman J, Klagsburn M (1987) Angiogenic factors. Science 235: 442
- 17. Fowler R, Stephens FD (1959) The role of testicular anatomy in the salvage of high undescended testes. Aust N Z J Surg 29: 92
- Frey P, Bianchi A (1987) Microvascular orchiopexy. Eur J Pediatr 146 (Suppl): S51
- 19. Gibbons MD, Cromie WJ, Duckett JW (1979) Management of the abdominal undescended testicle. J Urol 122: 76
- Hadziselimovic F (1991) Cryptorchidism. In: Gillenwater JY, Grayhack JT, Howards SS, Duckett JW (eds) Adult and pediatric urology. Mosby-Year Book, St Louis, p 2217
- 21. Hinman F (1979) Unilateral abdominal crytorchidism. J Urol 122: 71
- Hori Y, Tamai S, Okuda H, Sakamoto H, Takita T, Masuhara K (1979) Blood vessel transplantation to bone. J Hand Surg 4A: 23
- Janecka I, Romas N (1978) Microvascular free transfer of human testes. Plast Reconstr Surg 63: 41
- 24. Johnsen GJ (1970) Testicular biopsy score count-a method for registration of spermatogenesis in human testes: normal values and results in 335 hypogonadal males. Hormones 1: 2
- Khouri RK, Upton J, Shaw WW (1992) Principles of flap prefabrication. Clin Plast Surg 19: 763
- King LR (1984) Optimal treatment of children with undescended testes. J Urol 131: 734
- 27. Koff SA, Sethi P (1996) Treatment of high undescended testes by low spermatic vessel ligation: an alternative to the Fowler-Stephens technique. J Urol 156: 799
- 28. Kogan SJ, Houman BZ, Reda EF, Levitt SB (1989) Orchiopexy of the high undescended testis by division of the spermatic

- vessels: a critical review of 38 selected transactions. J Urol 141: 1416
- Lindgren BW, Franco I, Blick S, Levitt SB, Brock WA, Plamer LS, Friedman SC, Reda EF (2001) Laparoscopic Fowler-Stephens orchiopexy for the high abdominal testis. J Urol 162: 990
- 30. Murakami T, Uno Y, Ohtsuka A, Taguchi T (1989) The blood vascular architecture of the rat testis: a scanning electron microscopic study of corrosion casts followed by light microscopy of tissue sections, Arch Histol Cytol 52: 151
- O'Brien BMcM, Rao VK, Macleoad AM, Morrison WA, MacMahon RA (1983) Microvascular testicular transfer. Plast Reconstr Surg 71: 87
- 32. Ortolano V, Nasrallah PF (1986) Spermatic vessel ligation (Fowler-Stephens maneuver): experimental results with regard to fertility. J Urol 136: 211
- Pascual JA, Villanueva-Meyer J, Salido E, Ehrlich RM, Mena I, Rajfer J (1989) Recovery of testicular blood following ligation of testicular vessels. J Urol 142: 549
- Persky L, Albert DJ (1971) Staged orchiopexy. Surg Gynecol Obstet 132: 43
- 35. Ransley PG, Vordermark JS, Caldamone AA, Bellinger MF (1984) Preliminary ligation of the gonadal vessels prior to orchiopexy for the intra-abdominal testicle: a staged Fowler-Stephens procedure. World J Urol 2: 266
- Romero-Maroto J, Nistal M, Gonzales-Gancedo P, Bellas C, Arana A (1983) Transection of the spermatic vessels (Bevan's technique): an experimental study. J Urol 130: 1223
- Romero-Maroto J, Verdu-Tartajo F, Garcia-Gonzales R, Gonzales-Gancedo P, Cacicedo L (1986) The functional value of collateral testicular vascularization: an experimental study. Br J Urol 58: 553
- Salman FT, Fonkalsrud EW (1990) Effects of spermatic vascular division for correction of the high undescended testis on testicular function. Am J Surg 160: 506
- 39. Shintomi Y, Ohura T (1982) The use of muscle vascularized pedicle flaps. Plast Reconstr Surg 70: 725
- Shoshany G, Shofty R, Livne E, Hayari L, Mordechovitz D (1996) Testicular neovascularization by "omentotesticulopexy": a possible adjuvant in the surgical correction of high undescended testes. J Pediatr Surg 31: 1229
- 41. Silber SJ (1981) The intra-abdominal testes: microvascular autotransplantation. J Urol 125: 329
- Silber SJ, Kelly J (1976) Successful autotransplantation of an intra-abdominal testis to the scrotum by microvascular technique. J Urol 115: 452
- 43. Sönmez K, Basaklar AC, Türkyılmaz Z, Demiroğulları B, Numanoğlu V, Konus Ö, Dursun A, Altın MA, Kale N (1995) Neovascularization of the testicle through spermatic vessels by omental pedicle flap: a new experimental model. J Pediatr Surg 30: 1654
- Stern JA, Lui RC, LaRegina MC, Herbold DR, Tolman KC, Johnson FE (1990) Long-term outcome following testicular ischemia in the rat. J Androl 11: 390
- 45. Tark KC, Khouri RK, Shin KS, Shaw WW (1991) The fasciovascular pedicle for revascularization of other tissues. Ann Plast Surg 26: 149
- 46. Weerasooriya TR, Yamamoto T (1983) Three-dimensional organization of the vasculature of the rat spermatic cord and testis: a scanning electron-microscopic study of vascular corrosion casts. Cell Tissue Res 241: 317