Clinical Investigation

Effect of Electrocautery on Endothelial Integrity of the Internal Thoracic Artery:

Ultrastructural Analysis with Transmission Electron Microscopy

Burak Onan, MD
Mehmet Yeniterzi, MD
Ismihan Selen Onan, MD
Burak Ersoy, MD
Suheyla Gonca, MD
Elif Gelenli, MD
Seyhun Solakoglu, MD
Ihsan Bakir, MD

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From: *Departments of Cardiovascular Surgery (Drs. Bakir, Ersoy, B. Onan, I.S. Onan, and Yeniterzi), Istanbul Mehmet Akif Ersoy Thoracic and Cardiovascular Surgery Training Hospital, 34303 Istanbul; Histology and Embryology (Drs. Gelenli and Gonca), Faculty of Medicine, Kocaeli University, 41380 Kocaeli; and Histology and Embryology (Dr. Solakoglu), Istanbul University, Faculty of Medicine, 34093 Istanbul; Turkey*

Address for reprints: *Ihsan Bakir, MD, Department of Cardiovascular Surgery, Istanbul Mehmet Akif Ersoy Thoracic and Cardiovascular Surgery Training Hospital, Istanbul Caddesi, Bezirganbahce mevkii, Kucukcekmece, 34303 Istanbul, Turkey*

E-mail: *ihsanbak@yahoo.com*

The internal thoracic artery (ITA) is typically harvested from the chest wall by means of conventional electrocautery. We investigated the effects of electrocautery on endothelial-cell and vessel-wall morphology at the ultrastructural level during ITA harvesting.

Internal thoracic artery specimens from 20 patients who underwent elective coronary artery bypass grafting were investigated in 2 groups. The ITA grafts were sharply dissected with use of a scalpel and clips in the control group (n=10) and were harvested by means of electrocautery in the study group (n=10). Each sample was evaluated for intimal, elastic-tissue, muscular-layer, and adventitial changes. Free flow was measured intraoperatively. Light microscopic examinations were performed after hematoxylin-eosin and Masson's trichrome staining. Transmission electron microscopy was used to evaluate ultrastructural changes in the endothelial cells and vessel walls of each ITA.

In the sharp-dissection group, the endothelial surfaces were lined with normal amounts of original endothelium, endothelial cells were distinctly attached to the basal lamina, cytoplasmic organelles were evident, and intercellular junctional complexes were intact. Conversely, in the electrocautery group, the morphologic integrity of endothelial cells was distorted, with some cell separations and splits, contracted cells, numerous large cytoplasmic vacuoles, and no visible cytoplasmic organelles. The subendothelial layer exhibited disintegration. Free ITA flow was higher in the sharp-dissection group (P=0.04).

The integrity of endothelial cells can be better preserved when the ITA is mobilized by means of sharp dissection, rather than solely by electrocautery; we recommend a combined approach. ***(Tex Heart Inst J 2014;41(5):484-90)***

The internal thoracic artery (ITA) is the best conduit for coronary artery bypass grafting (CABG). Use of the ITA provides the longest-term patency for all grafts, better survival rates, and greater freedom from cardiac events.[1-4] The ITA graft can be harvested by means of mechanical dissection with a scalpel and clips, or through the use of electrocautery, ultrasonic energy, or radiofrequency energy.[5-9] When electrocautery is used to harvest the ITA from the chest wall, heat and electric current can be conducted through the surrounding tissues or clips and cause thermal injury or vasospasm. Suboptimal flow due to vasospasm increases the risk of early postoperative sequelae in patients who have poor left ventricular function and a recent myocardial infarction.[10]

Surgeons typically dissect the ITA by means of electrocautery, with hemoclip reinforcement. Decreased free ITA flow, a clinical sign of vasospasm, can be detected during surgery. Conversely, thermal injury to the smooth muscle and endothelial cells (ECs) is not a macroscopic finding, and it can go undetected intraoperatively. In the medical literature, there are few morphologic data about the effects of electrocautery on the ITA graft, despite surgeons' awareness of heat injury. Ultrastructural changes in the graft after electrocautery harvesting have been noted in a few studies.[5-7] In those investigations, scanning electron microscopy was used to reveal histologic features on the flow surface of the ITA after mobilization.[5,6]

The aim of this study was to evaluate ultrastructural changes at the cellular level when the ITA is mobilized from the chest wall by means of electrocautery. We used transmission electron microscopy to evaluate the effect of electrocautery on EC integrity and vessel-wall morphology.

Patients and Methods

The study protocol was approved by the ethics committee of Istanbul Mehmet Akif Ersoy Thoracic and Cardiovascular Surgery Training Hospital, and all patients gave written informed consent before surgery. From September through October 2012, ITA specimens were obtained from 20 consecutive patients with ischemic heart disease who underwent elective CABG (13 men and 7 women). The patients were randomly assigned to 2 groups. In the control group (n=10), the ITA was harvested by means of mechanical dissection with use of a scalpel and clips. In the study group (n=10), conventional electrocautery was used to harvest the ITA grafts. The characteristics of the patients were similar (Table I).

The exclusionary criteria were associated valvular heart disease, chronic heart failure, diabetes mellitus, chronic renal disease, peripheral arterial disease, inflammatory vascular disease, previous cardiothoracic surgery, and a history of radiotherapy to the chest wall. Patients who underwent urgent or emergent surgery were excluded.

Internal Thoracic Artery Harvesting Technique. All ITAs were harvested from the left anterior chest wall by the same surgeon, with surgical optical magnification ×3.5. After midline sternotomy, each ITA graft was removed as a pedicle with the internal thoracic veins, fat, muscle, and parietal pleura. Each graft was harvested between the left subclavian artery and a point distal to its bifurcation into the rectus sheath. The intercostal arteries were identified by using either the tip of the electrocautery blade (without current) or the tip of fine scissors. All collateral branches, including the first intercostal artery and mediastinal branches, were secured with use of SLS-Clip™ System titanium hemostatic clips (Vitalitec International Inc.; Plymouth, Mass). Side branches were always divided at a point 2 mm distal from the origin of the vessels from the ITA. The ITA was not grasped with instruments.

TABLE I. Characteristics of the Patients

Variable	Control Group (n=10)	Study Group (n=10)
Age, yr	57 (44–68)	58 (45–67)
Male/female	7/3	6/4
Height, cm	162 (155–170)	166 (158–179)
Weight, kg	78 (68–83)	77 (65–80)
Body mass index, kg/m^2	28 (26–30)	26 (24–29)
Left ventricular ejection fraction	0.54 (0.50–0.59)	0.58 (0.50–0.65)
Smoking history	4	3
Diabetes mellitus	3	4
Hypertension	3	2
Hypercholesterolemia	2	2
Obstructive pulmonary disease	1	2

Data are presented as number and range.

In the control group, all ITA grafts were mechanically dissected with use of a scalpel and clips. The side branches of the ITA were clipped at both ends and divided with scissors. In the study group, all ITA grafts were harvested with use of a Valleylab™ Force FX™ monopolar electrocautery blade (Covidien; Mansfield, Mass). The output of the electrocautery device for tissue dissection was adjusted to be as low as possible (<25 W).

After the grafts were mobilized, the patients were given 400 IU/kg of heparin intravenously. The ITA grafts were divided at the level of their bifurcation in the 6th or 7th intercostal space, and free flow was measured. The distal end was ligated while the ITA grafts were controlled proximally with a bulldog clamp. This clamp was placed on the middle segment of the graft, because the distal end of the ITA was preserved for histologic examination. The 1.5-cm specimen was sampled from the distal end of the left ITA graft. It was then dissected from all surrounding structures, including the accompanying veins, fascia, and lymphatic and adipose tissue.

The prepared ITA specimens were divided into 2 equal pieces in the operating theater: one piece for light microscopic examination and the other for electron microscopic examination. The tissue samples were immediately fixed in formalin or glutaraldehyde for histologic analysis at room temperature.

Preparation of Specimens for Light Microscopy. The ITA specimens were immersed in 10% neutral buffered formalin and processed for embedding in paraffin in transverse orientation. Sections 5 μm in thickness were applied to charged slides, deparaffinized, and hydrated with distilled water. The sections were then stained with hematoxylin-eosin and Masson's trichrome stain, which illustrates the relative proportion of collagen to stromal smooth muscle as previously described.[11,12] The Leica DM6000 B microscope (Leica Microsystems Inc.; Buffalo Grove, Ill) and Leica application suite (imaging software) were used to examine the specimens.

Preparation of Specimens for Transmission Electron Microscopy. The fixation process was performed for 24 hours on 0.5-mm tissue samples, within 2.5% phosphate-buffered glutaraldehyde solution at 4 °C. Postfixation was performed within 1% phosphate-buffered osmium tetroxide for an hour. The samples were dehydrated by passing them through ethyl alcohol, and they

were embedded in Epon® 812. The samples were kept at 70 °C in an incubator for one night for polymerization. Thin sections (400–600 Å) were cut with use of an ultramicrotome and were stained with uranyl acetate and lead citrate. Finally, the sections were evaluated with use of a JEM-1011 transmission electron microscope (Jeol; Tokyo, Japan).

Histopathologic Examination

Two independent histologists, blinded to the study protocol, used light and electron microscopy to perform all histopathologic examinations. Changes in the intima, elastic tissue, muscular layers, and adventitia were evaluated visually. Intimal changes included gross endothelial injury, endothelial surface disruption, disorganization, and denudation. Elastic-tissue changes were evaluated for disorganization of elastic fibers and for elastic tissue damage or disruption. Changes in the muscular and adventitial layers were also studied. Photomicrographs were taken in high-resolution mode (1,024 × 768 pixels).

The histologists used a scoring system to describe endothelial damage: 1, completely confluent endothelium; 2, partially confluent endothelium; 3, loosely netted endothelium; 4, islands of endothelium; and 5, no endothelium.[13]

Statistical Analysis

Data are expressed as mean ± SD. The Mann-Whitney test was used to analyze differences between groups. The statistical software package SPSS 16.0 (IBM Corporation; Armonk, NY) was used for data analyses. A *P* value of less than 0.05 was considered statistically significant.

Results

Intraoperative Macroscopic Findings

The ITA grafts were macroscopically evaluated with optical magnification ×3. Findings in both groups were similar: no perivascular hematoma or subadventitial bleeding, dissection, intimal injury, or change in the color of the vessels. The mean free ITA flow was 57 ± 11.4 mL/min in the control group (range, 42–75 mL/min) and 40.1 ± 15.7 mL/min in the study group (range, 30–70 mL/min) (P=0.04).

Light-Microscopic Findings

As shown in Figures 1 and 2, light-microscopic examination revealed vessel injury in most study-group sections, including denudation of the endothelium and disorganization of the subintimal collagen and elastic fibers. Tissue detachments were evident between the tunica media and tunica adventitia. There were morphologic disruptions between the endothelial and subendothelial layers. The luminal surfaces of some ITA tissues were not lined with ECs. The morphologic integrity of the ECs was distorted. There were some cell separations and splits from the basal lamina.

In the control group, the morphologic integrity of the vessel wall was better preserved. The ECs were morphologically preserved, and the luminal surfaces of the ITA were lined with ECs in all the specimens.

Ultrastructural Findings with Transmission Electron Microscopy

Figures 3 and 4 show the ultrastructural changes detected upon transmission electron microscopy. In the study group, the morphologic integrity of the ECs was distorted. There were some cell separations on the endothelial surface. Contracted endothelial cells were evident. The ECs showed numerous large cytoplasmic vacuoles. No cytoplasmic organelles were visible. There were fragmentations in the subendothelial layer.

In the control group, the luminal surfaces were lined with normal amounts of the original endothelium. The ECs were distinctly attached to the basal lamina, and cytoplasmic organelles were evident. The intercellular junctional complexes were intact. In accordance with the histologic scoring system, endothelial morphology was normal in all control-group specimens (1 ± 0.5), whereas the study-group specimens displayed considerable EC changes (3.5 ± 0.5) (P <0.01).

Postoperative Follow-Up. Postoperatively, no patient had major cardiovascular morbidity. Each was discharged from the hospital with a favorable outcome within one week of surgery.

Discussion

The endothelium plays a major role in regulating membrane permeability, lipid transport, vasomotor tone, coagulation, and inflammation. Preserved endothelial integrity protects vascular modulatory properties through the release of prostacyclin, endothelin, and endothelium-derived relaxing factors.[3,4] These crucial EC functions are affected by hypoxia, by exposure to cytokines, endotoxin, cholesterol, and nicotine, and by manipulations during surgery. In addition, investigators who have examined the role of ITA-harvesting techniques have noted that the structural integrity of the vessel wall, including the endothelium, is important in maintaining graft patency and in the long-term outcome of CABG.[3,4]

Various techniques are used when ITA grafts are harvested, including skeletonization, semi-skeletonization, and pedicled mobilization.[14] In addition, electrocautery, radiofrequency energy, and ultrasonic energy can serve as less traumatic ways of removing the ITA from the anterior chest wall. The aim of each approach is to maintain the structural integrity of the ITA graft in optimal fashion. Trauma during ITA harvesting can

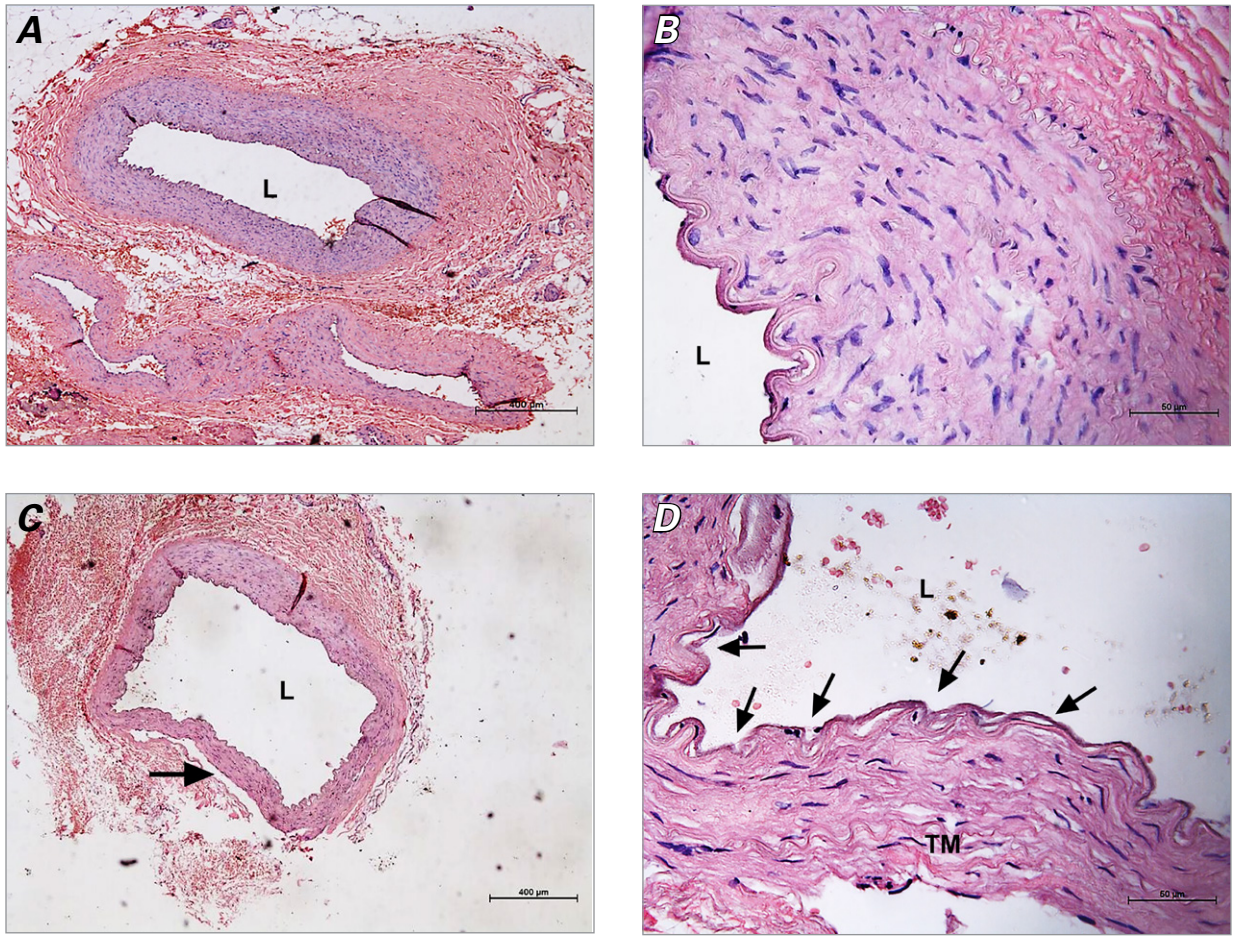

Fig. 1 *Photomicrographs from the control group show* ***A****) an internal thoracic artery (H & E, orig. ×5) and* ***B****) intact endothelial and subendothelial layers (H & E, orig. ×40). Photomicrographs from the study group show* ***C****) detachment (arrow) of the subendothelial layer of an internal thoracic artery (H & E, orig. ×5) and* ***D****) denudation of the endothelial layer (arrows) and disorganization of subintimal collagen and elastic fibers (H & E, orig. ×40).*

L = lumen; TM = tunica media

lead to graft dysfunction and detrimentally affect the postoperative course of CABG patients.

Conventional electrocautery typically enables easy, rapid, and inexpensive mobilization of the ITA. However, the heat that is transmitted to the artery can occasionally injure the endothelium.[5-7] This iatrogenic sequela presents with regional vascular spasm and associated decreased ITA flow. Adventitial hematoma or intimal dissection might also develop. In addition to noting such macroscopic findings caused by thermal injury, Lehtola and associates[5] reported endothelial-flow surface damage caused by cautery current, either through direct contact with the wall or via conduction through metal hemoclips. Yoshida and colleagues[6] used scanning electron microscopy to show partial loss of ECs on the flow surface of the ITA. In accordance with these findings, it is preferable to deliver lower coagulation energy during tissue dissection and pedicled harvesting of the graft, to avoid endothelial damage and associated graft failure or vasospasm. Nevertheless, thermal injury to the vessel wall and ECs might go undetected during surgery despite all these maneuvers.

After our harvesting of the ITA grafts by means of low-voltage electrocautery, we observed no subadventitial hematoma or bleeding, vasospasm, dissection, intimal injury, or color change in the vessels. However, upon histologic examination, some study-group sections showed evidence of thermal injury, including denudation of the endothelium and disorganization of the subintimal collagen and elastic fibers. Light microscopic examinations revealed tissue detachments between the tunica media and tunica adventitia, and morphologic disruptions between the endothelial and subendothelial layers. In the tunica intima, the luminal surfaces of some ITA tissues were not lined with ECs. The morphologic integrity of the arterial wall was therefore distorted after the tissue had been dissected via electrocautery. In the control group, the morphology of the ECs was preserved, and the luminal surfaces of the ITAs were lined with ECs in all specimens.

Electron-microscopic studies about the effect of cautery on endothelial layers are sparse.[5,6] The few available studies reported features of the luminal surface of ITA endothelium after electrocautery harvesting. Yoshida and associates[6] observed accumulations of red blood cells and platelets, with almost a complete loss of ECs, in skeletonized ITAs harvested by means of monopolar electrocautery. Lehtola and colleagues[5] showed that the flow surface of the ITA was well preserved after harvesting by means of low-voltage electrocautery. They noted that caution is needed, because contact of the cautery blade with the artery or the hemostatic clips on the side branches might cause tissue damage. However, these 2 studies clarified neither the ultrastructural features nor the effects of electrocautery on the vessel wall. Conversely, in our study, analysis with transmission electron microscopy yielded distinct evidence of ultrastructural and vessel-wall damage in specimens in the study group. We observed no such damage in the control group.

Alternative energy sources have been used to reduce injury to the ITA. Keeley and colleagues[15] noted that bipolar electrocautery enables precise control of current and avoids random spraying of heat, whereas monopolar electrocautery does not. Yoshida and associates[6] concluded that monopolar electrocautery is more often associated with intimal corrugation than is bipolar electrocautery, and they suggested that bipolar mode might be preferable to monopolar, especially in skeletonized dissection of the ITA. Ultrasonic coagulation is another alternative to electrocautery, especially when the artery is harvested by means of a skeletonizing technique during CABG.[8] In monopolar or bipolar mode, radiofrequency energy yields preserved endothelial function and anatomic integrity of the ITA graft.[9]

Because of variability in surgical techniques, it is difficult to recommend electrocautery over other techniques or the converse. Electrocautery is typically used, and the resultant long-term patency of ITA grafts is excellent

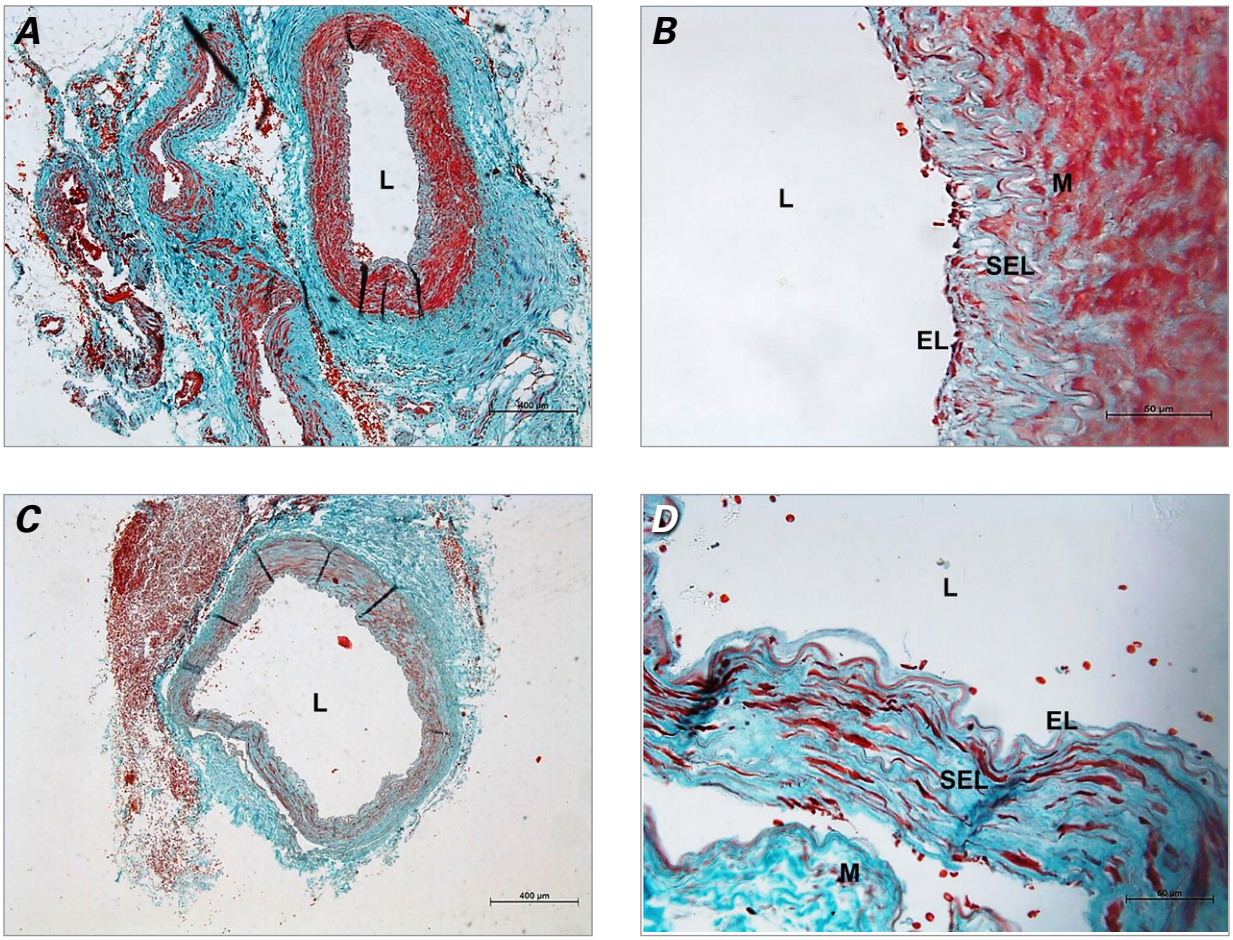

Fig. 2 *Photomicrographs with Masson's trichrome stain. In the control group,* ***A****) the vessel wall is intact (orig. ×5) and* ***B****) the integrity of the endothelial cells and subendothelial elastic fibers is preserved (orig. ×40). Note the homogeneity of the muscular fibers. In the study group,* ***C****) tissue is detached within the subendothelial layer (orig. ×5), with less intense staining than in the images from the control group; and* ***D****) injury to the endothelial layer and subendothelial elastic fibers is evident (orig. ×40). Note the irregularity of the endothelial layer and separation of the subendothelial layers.*

EL = endothelial layer; L = lumen; M = muscular fibers; SEL = subendothelial layer

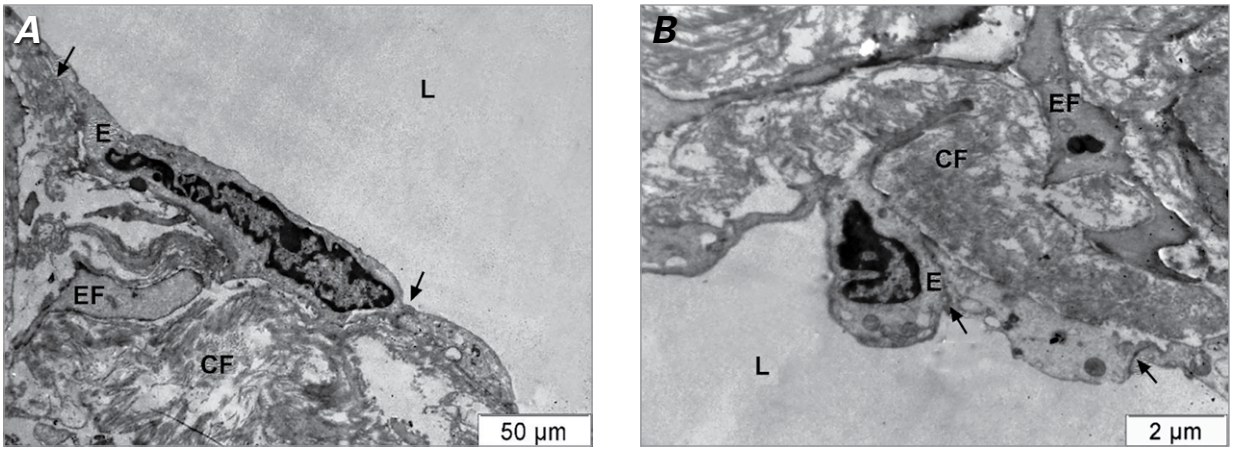

Fig. 3 *Transmission electron microscopic images. In the control group,* ***A****) the endothelial surface of the artery is regular, endothelial cells are distinctly attached to the basal lamina, cytoplasmic organelles are clearly seen, and intercellular junctional complexes (arrows) are evident.* ***B****) The luminal surface of the endothelium is regular and lined with normal endothelial cells, cytoplasmic organelles are apparent, and intercellular junctional complexes (arrows) are evident.*

CF = collagen fiber; E = endothelial cell; EF = elastin fiber; L = lumen

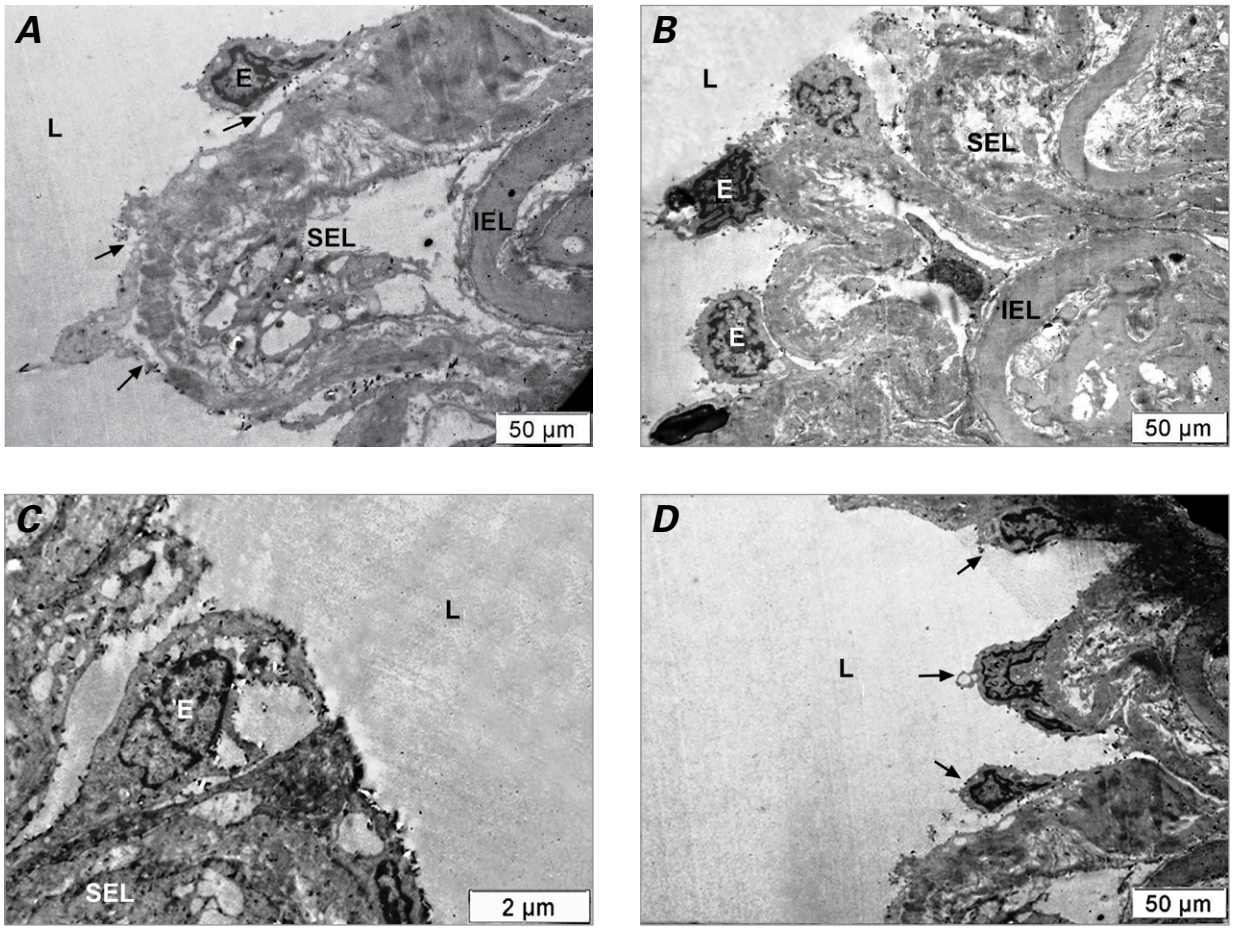

Fig. 4 *Transmission electron microscopic images. In the study group,* ***A****) endothelial integrity is distorted, with separations of endothelial cells (arrows). Endothelial cells are contracted and have numerous large cytoplasmic vacuoles. No cytoplasmic organelles are visible. There are fragmentations in the subendothelial layer.* ***B****) Endothelial integrity is distorted, with separations and contracted endothelial cells.* ***C****) Numerous large cytoplasmic vacuoles are evident, no cytoplasmic organelles are visible, and contracted endothelial cells are seen.* ***D****) Arrows point to contracted endothelial cells.*

E = endothelial cell; IEL = internal elastic lamina; L = lumen; SEL = subendothelial layer

in CABG.[1,2,16] Some authors might argue in behalf of sharp dissection over electrocautery. Our ultrastructural findings suggest that using a scalpel and clips for ITA harvesting might have a positive impact on graft patency; however, this has not been proved. In addition, free ITA flow in our control group was significantly higher than that in our study group. The results of our study confirm the value of a combined surgical approach for ITA harvesting: incising the pleura and muscle on either side of the ITA and accompanying veins by using low-power cautery as an initial maneuver, mobilizing the ITA pedicle from the chest wall without cautery, clipping the proximal and distal sites of the side branches, and finally dividing the branches between the clips with scissors. Cautery should be used only for incising the pleura—to avoid heat injury, it should not be used to divide the side branches.

The first limitation of our study is the small sample size; regardless, the ultrastructural findings from transmission electron microscopy were significantly different between the groups. Clinically, it would be valuable to see whether the histologic changes translate into graft patency.

Despite the small number of specimens, our study indicates that ITA grafts harvested by means of mechanical dissection have better vessel-wall morphology than do ITA grafts harvested solely by means of electrocautery. Extreme care is needed to avoid endothelial injury when electrocautery is used to harvest the ITA.

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