Original Article
Mechanisms underlying the effects of different abdominal surgical traumas on rabbit microvascular albumin leakage

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Abstract: In this study, microvascular albumin leakage and the possible mechanisms underlying this response were investigated after different abdominal traumas in rabbits. Sixty-six rabbits were randomly divided into a control group, partial jejunum resection group (group A), and stomach-pancreas-spleen resection group (group B). The serum albumin concentration and levels of interleukin-6 (IL-6) and tumor necrosis factor α (TNF-α) were examined before surgery, as well as at various time points after surgery. Mesenteric microvascular leakage of albumin was dynamically observed, as were changes in microvascular endothelial cells. The albumin concentration in group B was lower at different postoperative time points than before surgery \( (P < 0.05) \), and no statistical difference was detected between groups at the 6 postoperative time points and before surgery \( (P > 0.05) \). Serum IL-6 concentrations in group A were higher at 24 and 48 hours postoperation than before surgery \( (P < 0.05) \); in group B, the concentration was higher at each postoperative time point than before surgery \( (P < 0.01) \). The serum TNF-α concentration at 48 hours postoperation in group A was higher than that before surgery \( (P < 0.05) \), and the concentration at each postoperative time point in group B was higher than that before surgery \( (P < 0.01) \). The mesenteric microvascular leakage rates in group B were higher at postoperative hours 12, 24, 48, and 72 than in the control group \( (P < 0.01) \). Endothelial gaps were clearly observed in group B. The serum albumin concentration decreased in the early phase after major abdominal surgery in rabbits, which may be related to the increase in serum inflammatory mediators after major surgery, resulting in increased microvascular endothelial gaps and albumin leakage.

Keywords: Surgery, albumin class, interleukin-6, tumor necrosis factor α, microvascular leakage

Introduction

A rapid decline in the serum albumin concentration is commonly observed after major surgery or severe acute trauma; the incidence of hypoalbuminemia is as high as 70% to 80% [1, 2]. Postoperative hypoalbuminemia not only delays the healing of tissues, but also reduces the anti-infection ability, increases the incidence of postoperative complications and mortality, and is closely related to patient condition and prognosis [3]. Ryan et al. [4] retrospectively analyzed the postoperative situations of 200 patients with esophageal cancer, and found that the incidence of postoperative complications in patients with an albumin concentration of < 20 g/L on the first postoperative day was two-fold greater than it was in patients with an albumin concentration of > 20 g/L, i.e., 54% and 28%, respectively, and the in-hospital mortalities were 27% and 6%, respectively. In the past, postoperative hypoalbuminemia was believed to reflect protein synthesis disorders, as well as changes in catabolism. However, the synthesis and decomposition of albumin requires certain processes, and a synthesis deficiency or catabolism-induced hypoalbuminemia might appear a few weeks or a few months after liver dysfunction or starvation. Accordingly, a reduction in serum albumin levels that appears a few hours after major surgery or severe trauma is difficult to explain by the above mechanisms. Some scholars believe that severe post-traumatic stress-related tissue and
systemic inflammatory responses can increase microvascular permeability, resulting in a redistribution of albumin inside and outside of blood vessels, which can explain an early decline in the serum albumin concentration [5, 6]. Severe trauma can cause physiological and pathological changes, such as the activation of inflammatory mediators and ischemia-reperfusion injuries, causing microvascular endothelial cell damage, enhanced adhesive abilities, widened intracellular spaces, and increased permeability. These changes reduce the barrier functions of endothelial cells and their basement membranes towards macromolecules; therefore, the “leakage” of macromolecules, such as serum albumin, into interstitial spaces increases [7-14].

In our pre-clinical studies, patients with middle and major abdominal surgeries exhibited decreases in plasma albumin at early time points and inflammatory mediators increased to varying degrees. The variation in the degree to which inflammatory mediators increase might reflect the degree of surgical trauma, whereby greater surgical trauma is associated with more obvious increases in inflammatory mediators. Owing to the limitations of testing methods and medical ethics considerations, there is still controversy regarding the causes of decreased postoperative serum albumin concentrations at early time points. In this study, the plasma albumin concentration, serum interleukin-6 (IL-6), and tumor necrosis factor α (TNF-α) were examined in the early phase after different abdominal traumatic surgeries. Additionally, the leakage of mesenteric microvascular albumin, microvascular permeability rate, and spatial changes in lung, mesentry, and intestinal microvascular endothelial cells were characterized to explore whether microvascular albumin leakage occurred after abdominal traumas in rabbits and the possible mechanisms underlying this response.

Methods

Animals

The experimental animals were provided by the Experimental Animal Center, Kunming Medical University [license no. SYXK (Dian) 2011-0004]. The rabbits were fed with full-price nutrients in separate cages, anesthetized with 3% sodium pentobarbital (30 mg/kg) before surgery according to body weight, and killed by the air embolism method after the experiment. Animal carcasses were uniformly disposed of by the Experimental Animal Center. This study was carried out in strict accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health. The animal use protocol has been reviewed and approved by the Institutional Animal Care and Use Committee (IACUC) of Kunming Medical University.

Experimental methods

Sixty-six male Japanese rabbits were randomly divided into six groups, with 30 rabbits in the partial jejunum resection group (group A) and the stomach-pancreas-spleen resection group (group B) (6 rabbits each for observations at 6, 12, 24, 48, and 72 hours). Rabbits in the control group were not subjected to surgery, those in group A were subjected to post-jejunectomy anastomosis, and those in group B were subjected to major gastrectomy plus pancreatectomy and splenectomy. All rabbits were provided nutritional support with isocalories and isonitrogen; the 150 kcal included glucose and fat, which provided 70% and 30% of the total calories, respectively. The nitrogen-calorie ratio was 1:130. The serum albumin concentration, serum IL-6, and TNF-α levels in rabbits of the experimental groups were tested before surgery, as well as at different time points after surgery (postoperative hours 6, 12, 24, 48, and 72). Rhodamine B isothiocyanate (RBITC)-labeled bovine serum albumin (BSA) (Sigma-Aldrich, St. Louis, MO, USA) was injected intravenously, and mesenteric microvascular leakage of albumin was dynamically observed using a laser scanning confocal microscope. Microvascular albumin permeability was then calculated. Changes in the lung and mesentery, as well as spatial changes in intestinal microvascular endothelial cells were observed by transmission electron microscopy (TEM).

Surgical procedures and laser scanning confocal microscopy

All rabbits were adaptively fed for one week in the laboratory before surgery, and the surgical group was fasted for 12 hours before surgery, but provided free access to water. In group A, 3% sodium pentobarbital (30 mg/kg) was injected intravenously, an ~10-cm incision was
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made along the median abdominal line, the abdomen was opened layer by layer, the stomach was freed, the gastric body was transected for the major gastrectomy, the full-thickness of duodenal stump was sutured and closed. The jejunum was transected approximately 15 cm under the flexor ligament and stomach-jejunum Roux-Y anastomosis was performed. The end of pancreatic body was freed, cut off, and the splenic artery and vein were ligated in the corresponding positions, the pancreatic duct was ligated, and the pancreatic stump was sutured. After careful ligation and hemostasis, the peritoneal drainage tube was indwelled, the surgical equipment was checked, and the abdomen was closed layer by layer. Aseptic techniques were strictly used for the surgery. The operations were performed by specific staffs, and the operation time was strictly controlled to within 1.5 hours. For group B, an incision was made along the median abdominal line of approximately 5 cm, the abdomen was opened layer by layer, approximately 10 cm of the jejunum was cut from about 15 cm below the flexor ligament, and stump anastomosis was performed. After careful ligation and hemostasis, the peritoneal drainage tube was indwelled, surgical equipment was checked, and the abdomen was closed layer by layer, ensuring a strictly aseptic surgery. The operations were performed by specific staff, and the operation time was strictly controlled to within 0.5 hours.

The rabbits in each group were fixed in a fixation box at each detection time point and injected with 3% pentobarbital sodium (30 mg/kg) through the ear vein. After they were anesthetized, rabbits were removed from the fixation box, fixed supinely on the operating table, disinfected, covered with sterile towels, and the abdomen was reopened along the original incision. The rabbit was moved under a confocal laser microscope (FV1000; Olympus, Tokyo, Japan), and a small part of the small intestine and intestinal loop, approximately 15 cm from the cecum, was lifted and the distribution of mesenteric vessels was visually observed. An intestinal part with small blood vessels and less fat was selected as the observation area. The extruded mesentery was stretched onto the transparent plexiglass perfusion box, lights and curtains were closed, the excitation wavelength of the laser confocal microscope was set at 543 nm, and a 20× objective lens was used. RBITC-labeled BSA albumin solution (1 mL/kg) was injected through the ear vein, and a microvessel was selected with a diameter of 20-35 μm to dynamically record mesenteric microvascular albumin leakage. Digital images were obtained simultaneously.

Leakage rate calculation

FV10-ASW 2.1 Viewer was used to evaluate the digital images. Two regions, inside and outside of the vessel, were selected to measure fluorescence intensities before and 2 min after the RBITC-BSA injection. Average values were then calculated, and actual fluorescence intensity = fluorescence intensity after injection-fluorescence intensity before injection. (The extravascular relative fluorescence intensity was estimated in the region 20 μm from the blood vessel at the vascular leakage site).

The relative changes in intra- and extravascular mesenteric fluorescence intensities were used to determine mesenteric microvascular albumin permeability as follows [15]: ΔI = 1-(II-IO)/II (ΔI, relative change in light intensity; II, intravascular light intensity; IO, extravascular light intensity per unit area).

Detection of inflammatory mediators

Venous blood (3 mL) was sampled from the ear vein of rabbits in all experimental groups before surgery and at different time points after surgery (6, 12, 24, 48, and 72 hours). Samples were maintained for 4 hours at room temperature and centrifuged at 3,000 rpm for 10 minutes to carefully and rapidly separate serum and red blood cells. The upper serum was then extracted and stored at -80°C for future tests. Before tests, the kit (Sigma-Aldrich) was removed from the refrigerator (4°C) 20 minutes in advance, and the serum samples were removed from the refrigerator (-80°C) and equilibrated to room temperature before tests. The double-antibody one-step ELISA method was used, absorbance (OD) was determined at 450 nm, and the standard curve was drawn to calculate sample concentrations.

TEM

The dual-blade sawing method was used to sample lung, mesentery, and intestinal tissues,
from which 2-mm slices were prepared. The samples were then placed in 2.5% glutaraldehyde (prepared using 0.2 mol/L sodium cacodylate buffer) at 4°C for 2 hours for fixation, followed by rinsing, fixed, rinsed, dehydrated, embedded, trimmed, sliced (Leica EM UC6 Ultra-thin Slicer, Buffalo Grove, IL, USA), stained, rinsed, and dried. A Hitachi transmission electron microscope (H-7650 (80 kV); Tokyo, Japan) was used to observe the specimens.

**Statistical analysis**

Microsoft Office Excel 2007 was used to record data. The data are expressed as means ± standard deviation (X ± s). Data were examined using repeated measures analysis of variance (ANOVA), and the least significant difference (LSD) method was used to further estimate differences for pairwise comparisons. Differences in indexes between groups before and after the surgery were assessed using paired t-tests. All statistical analyses were implemented in SPSS 17.0, and P < 0.05 was considered significant.

**Results**

**Albumin concentrations**

In group A, there was no significant difference in albumin concentration between different postoperative time points and before surgery (P > 0.05), but the concentrations at different postoperative time points in group B were lower than that before surgery (P < 0.05, Table 1). No significant difference in serum albumin was detected between the 6 postoperative time points and before surgery (P > 0.05).

**Serum IL-6 concentrations**

The serum IL-6 concentrations were higher at postoperative hours 24 and 48 in group A than before surgery (P < 0.05, Table 1), and the concentrations at each postoperative time point in group B were higher than that before surgery (P < 0.01, Table 1).

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### Table 1. Comparison of albumin, serum IL-6 and TNF-α concentrations at different detection time points

<table>
<thead>
<tr>
<th></th>
<th>6th-h subgroup</th>
<th>12th-h subgroup</th>
<th>24th-h subgroup</th>
<th>48th-h subgroup</th>
<th>72th-h subgroup</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Group B</strong></td>
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</tr>
<tr>
<td>Albumin (g/L)</td>
<td>41.98±6.04</td>
<td>39.57±3.63</td>
<td>40.37±4.56</td>
<td>42.23±4.94</td>
<td>40.82±3.69</td>
</tr>
<tr>
<td>After</td>
<td>38.17±5.03</td>
<td></td>
<td>31.93±3.74</td>
<td>29.97±3.30</td>
<td>32.52±2.89</td>
</tr>
<tr>
<td><strong>P</strong></td>
<td>0.315</td>
<td>0.047</td>
<td>0.009</td>
<td>0.004</td>
<td>0.013</td>
</tr>
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<td><strong>Group A</strong></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>40.85±7.44</td>
<td>39.55±6.58</td>
<td>40.97±7.98</td>
<td>40.97±6.47</td>
<td>39.62±6.61</td>
</tr>
<tr>
<td>After</td>
<td>38.37±6.23</td>
<td>37.72±6.22</td>
<td>37.01±7.12</td>
<td>37.30±5.24</td>
<td>37.30±5.96</td>
</tr>
<tr>
<td><strong>P</strong></td>
<td>0.277</td>
<td>0.492</td>
<td>0.173</td>
<td>0.214</td>
<td>0.324</td>
</tr>
<tr>
<td><strong>Group B</strong></td>
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<td></td>
</tr>
<tr>
<td>IL-6 (pg/ml)</td>
<td>12.30±1.84</td>
<td>11.85±1.65</td>
<td>12.08±1.11</td>
<td>12.16±1.55</td>
<td>12.28±1.57</td>
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<tr>
<td>After</td>
<td>22.35±2.08</td>
<td>24.28±3.50</td>
<td>28.37±4.70</td>
<td>23.42±2.03</td>
<td>20.76±5.77</td>
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<td><strong>P</strong></td>
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<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.006</td>
</tr>
<tr>
<td><strong>Group A</strong></td>
<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-6 (pg/ml)</td>
<td>12.00±1.15</td>
<td>12.31±1.60</td>
<td>11.51±1.47</td>
<td>11.41±1.54</td>
<td>12.67±1.15</td>
</tr>
<tr>
<td>After</td>
<td>13.07±1.53</td>
<td>13.96±1.66</td>
<td>15.38±1.79</td>
<td>13.68±1.77</td>
<td>13.40±1.11</td>
</tr>
<tr>
<td><strong>P</strong></td>
<td>0.289</td>
<td>0.075</td>
<td>0.007</td>
<td>0.017</td>
<td>0.367</td>
</tr>
<tr>
<td><strong>Group B</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TNF-α (pg/ml)</td>
<td>49.98±4.57</td>
<td>52.33±6.32</td>
<td>52.70±5.00</td>
<td>50.25±8.76</td>
<td>51.62±6.38</td>
</tr>
<tr>
<td>After</td>
<td>62.30±6.27</td>
<td>72.00±7.20</td>
<td>76.25±5.94</td>
<td>81.61±6.04</td>
<td>73.50±4.25</td>
</tr>
<tr>
<td><strong>P</strong></td>
<td>0.003</td>
<td>0.000</td>
<td>0.000</td>
<td>0.003</td>
<td>0.002</td>
</tr>
<tr>
<td><strong>Group A</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TNF-α (pg/ml)</td>
<td>54.25±10.83</td>
<td>54.97±12.55</td>
<td>51.19±8.89</td>
<td>53.33±9.44</td>
<td>51.44±9.70</td>
</tr>
<tr>
<td>After</td>
<td>57.35±9.71</td>
<td>61.43±11.80</td>
<td>66.15±6.95</td>
<td>62.67±10.49</td>
<td>62.16±10.28</td>
</tr>
<tr>
<td><strong>P</strong></td>
<td>0.372</td>
<td>0.061</td>
<td>0.040</td>
<td>0.257</td>
<td>0.070</td>
</tr>
</tbody>
</table>

Note: ANOVA analysis result: Albumin in the A group: before operation, F=0.058, P=0.998; after operation, F=0.139, P=0.982; TNF-α in the B group: before operation, F=0.267, P=0.928; after operation, F=4.436, P=0.004. IL-6 in the A group: before operation, F=0.723, P=0.612; after operation, F=1.999, P=0.107; TNF-α in the B group: before operation, F=0.069, P=0.996; after operation, F=13.357, P < 0.0001. TNF-α in the A group: before operation, F=0.133, P=0.984; after operation, F=1.418, P=0.246; TNF-α in the B group: before operation, F=0.158, P=0.976; after operation, F=14.488, P < 0.0001. The P values presented in the Table are obtained using LSD method through comparing with the control group.
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Table 2. Comparisons of microvascular albumin leakages among different detection time points

<table>
<thead>
<tr>
<th>Groups</th>
<th>6th-h subgroup</th>
<th>12th-h subgroup</th>
<th>24th-h subgroup</th>
<th>48th-h subgroup</th>
<th>72th-h subgroup</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.54±0.08</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>0.79±0.22</td>
<td>1.17±0.19</td>
<td>1.63±0.27</td>
<td>2.14±0.39</td>
<td>1.76±0.25</td>
</tr>
<tr>
<td>P</td>
<td>0.09</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>A</td>
<td>0.56±0.07</td>
<td>0.61±0.07</td>
<td>0.62±0.06</td>
<td>0.58±0.09</td>
<td>0.56±0.07</td>
</tr>
<tr>
<td>P</td>
<td>0.635</td>
<td>0.12</td>
<td>0.082</td>
<td>0.365</td>
<td>0.635</td>
</tr>
</tbody>
</table>

Note: ANOVA analysis result: A group: F=0.982, P=0.445; B group: F=36.321, P < 0.0001. The P values presented in the Table are obtained using LSD method through comparing with the control group.

Serum TNF-α concentration

The TNF-α concentration at postoperative hour 48 in group A was higher than that before surgery (P < 0.05, Table 1), and the concentration at each postoperative time point in group B was higher than that before surgery (P < 0.01, Table 1).

Mesenteric microvascular leakage rate

There was no significant increase in the mesenteric microvascular leakage rate at different postoperative time points in group A compared with the control group (P > 0.05), but group B exhibited increased leakage rates at postoperative hours 12, 24, 48, and 72 compared with the control group (P < 0.01, Table 2). There was almost no extravascular fluorescence in the control group, but after 6 hours post-surgical trauma, a small amount of extravascular leakage could be observed. Leakage increased at postoperative hours 12, 24, 48, and 72, and was most obvious at 48 hours (Figure 1).

Correlation analysis

The microvascular albumin leakage rate was positively correlated with the concentrations of IL-6 and TNF-α (r=0.653, P < 0.01).

Spatial changes in microvascular endothelial cells (Figure 2)

The control group and group A showed integral lung, mesenteric, and intestinal microvascular endothelial cellular membranes, and the connections were tight. Group B showed obvious swelling in lung, mesenteric and intestinal microvascular endothelial cellular membranes; 0.2-0.5-μm cracks appeared in the microvascular endothelial cellular junctions (which originally exhibited tight connections), and some cellular membranes exhibited vacuolization.

Discussion

Clinical studies have shown that an acute decline in the serum albumin concentration typically occurs in the early phase after surgery, severe trauma, and infection, and the incidence rate of hypoalbuminemia is up to 70% to 80% [1, 2]. This seriously affects patient prognosis and has become an important area of research in the fields of surgery and critical medicine. However, the specific causes and mechanisms underlying this pattern are not fully understood. Studies have shown that in the early phase of severe stress, such as infective toxic shock and severe pancreatitis, albumin levels might be reduced; this reduction might be related to increased microvascular permeability and the redistribution of albumin inside and outside of blood vessels.

Reduced plasma albumin levels are common after severe trauma, infection, burns, and major surgery. In this experiment, the serum albumin concentration was not significantly reduced after partial jejunectomy, but it was significantly lower after stomach-pancreas-spleen resection than before surgery. The reduction in the serum albumin concentration was most obvious at 48 hours postoperation, indicating that not all patients have a reduction in serum albumin, and the changes are related to the magnitude of the surgical stress, to some extent.

Studies have shown that the reduction in albumin levels in the early post-trauma period is primarily associated with inflammation-related increases in microvascular permeability, resulting in the redistribution of albumin inside and outside of blood vessels [1]. Estimating the microvascular albumin leakage rate is an important method to assess microvascular permeability and albumin leakage. Childs et al.
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Figure 1. Mesenteric microvascular leakage. A: Mesenteric microvascular albumin leakage of each group after stomach-pancreas-spleen resection. B: Mesenteric microvascular albumin leakage of each group after partial jejunectomy.
Figure 2. Spatial changes of microvascular endothelial cells. A: Rabbit pulmonary microvascular endothelial cells. Al: Alveolar space; BM: basal membrane; TJ (black thin arrow): cells were tightly connected; Cap: capillary cavity; Ed: endothelial cells; RBC: red blood cells; N: nuclei; EP: type II alveolar epithelial cells; thick black arrow: spaces among...
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endothelial cells; *: vacuolized membranes. B: Rabbit mesenteric microvascular endothelial cells. IS: intercellular space; BM: basal membrane; TJ (black thin arrow): cells were tightly connected; Cap: capillary cavity; Ed: endothelial cells; RBC: red blood cells; N: nuclei; thick black arrow: spaces among endothelial cells. C: Rabbit intestinal microvascular endothelial cells. BM: basal membrane; TJ (black thin arrow); cells were tightly connected; Cap: capillary cavity; Ed: endothelial cells; RBC: red blood cells; thick black arrow: spaces among endothelial cells.

[15] injected fluorescein isothiocyanate-labeled BSA into a rat model of hemorrhagic shock, and examined the relative changes in fluorescent intensity to study vascular permeability. They found that albumin leakage of the post-small mesenteric capillary microvein increased significantly and that permeability had increased as well. These previous studies provide evidence for the occurrence of microvascular albumin leakage, but studies of microvascular albumin leakage after surgical trauma are rare. In this study, mesenteric microvascular albumin leakage was similar after partial jejunectomy and in the control group at each time point. An imaging analysis revealed leakage, which appeared as fluorescence-labeled albumin that was uniformly spread outside the blood vessels, but no notable punctate exudation was observed. Microvascular albumin leakage was higher at each time point after stomach-pancreas-spleen resection than in the control group. The maximal leakage rate was observed after 48 hours, at which point leakage was most obvious, indicating that microvascular albumin leakage occurred after major surgery. The reduction in the serum albumin concentration was related to extravascular albumin leakage, and this might result from trauma-induced stress responses, which causes increased systemic capillary permeability, facilitating extravascular leakage of albumin along a concentration gradient. The extent of leakage was also associated with the degree of surgical trauma.

After stimulation by major surgery, trauma, and other stress factors, the body is capable of producing large amounts of inflammatory mediators, particularly cytokines. The greater the surgical trauma, the greater the increase in cytokine release [16]. Among the complex chain reactions of cytokines after major abdominal surgery, TNF-α plays a central role [17]. TNF-α is mainly produced by macrophages shortly after the body suffers from severe trauma and stress. TNF-α is not only involved in inflammatory reactions, but could also indirectly stimulate other inflammatory mediators involved in inflammatory reactions; accordingly, it is called a “broad-spectral inflammatory mediator” [18]. TNF-α could directly activate mononuclear phagocytes and leukocytes, leading to the release of interleukin-1β and other pro-inflammatory cytokines. This further promotes the release of de-granules and lysosomal enzymes, increases the generation of fat metabolites, and mediates cell and tissue damage. IL-6 is a component of the nerve-endocrine-immune system, and has dual effects in pro-inflammation and anti-inflammation. Therefore, it is a marker of tissue damage, and is a central regulatory of immune responses and inflammation. A high-concentration of TNF-α, IL-6, and other inflammatory mediators produced by the body could interact with inflammatory cells; however, high-concentrations of TNF-α and IL-6 could also activate the cytokine cascade. In this study, the concentrations of TNF-α and IL-6 in group B were higher than those in the control group; IL-6 and TNF-α levels peaked at 24 and 48 hours postoperation, and began to decrease after 48 and 72 hours, but were still higher than those of the control group. These results indicate that minor surgeries have little effects on inflammatory mediators, and the duration of the effects was short, but greater surgical trauma results in higher serum cytokine concentrations and longer-lasting effects. Therefore, it could be inferred that the concentrations of serum cytokines might reflect the degree of surgical trauma. The higher the concentrations of inflammatory mediators, the higher the microvascular albumin leakage rate, suggesting that greater abdominal surgical trauma is associated with more severe inflammation, greater microvascular permeability, and more obvious microvascular albumin leakage. It is possible that inflammatory mediators cause direct damage and indirectly aggravate hypoxic-ischemic damage. The aggravation of microcirculation disorders
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would lead to the activation of more endothelial cells, thus triggering the cascade effects of inflammatory responses, increasing microvascular permeability and, consequently, the microvascular albumin leakage rate.

Lum and Malik [19] demonstrated that the exchange of macromolecules within microvascular endothelial cells occurs via two mechanisms, i.e., the paracellular pathway and the transcellular pathway. In physiological conditions, small molecules, such as water and gas, can be freely exchanged through the endothelial barrier via the paracellular pathway. Owing to their high molecular weights, plasma proteins cannot be exchanged within intact endothelial cells, but are exchanged transcellularly by a complex vesicle transportation system. In this study, the control group and group A showed integrated lung, mesenteric, and intestinal microvascular endothelial cell membranes, and the connections were tight. However, group B showed obvious swelling of lung, mesenteric, and intestinal microvascular endothelial cells, cracks of 0.2-0.5 μm appeared among the microvascular endothelial cells, which were tightly connected originally, and some cellular membranes exhibited vacuolization. This might indicate that under pathological conditions, such as trauma, inflammatory mediators are released in high quantities, thus activating the constricting myosin and actin proteins. At the junction points where the extracellular matrix proteins adhere to cells, these substances might induce the activity of tissue endothelial cytoskeletal proteins and the retraction of endothelial cells, resulting in structural and functional disorders of connexin, and increased space between endothelial cells [20]. Studies have shown that vascular endothelial cellular spaces match the distributions of leakage sites within the microvascular system. After leakage forms, macromolecules, such as serum albumin, could pass the endothelial space, and do not require energy-dependent transmembrane transportation. During the acute inflammation period, the endothelial space may form rapidly, and combined with the increase in inflammatory mediators and leakage sites, the number of spaces may also increase. Therefore, the microvascular endothelial cellular spaces may represent albumin leakage sites under an inflammatory state [21].

In summary, in the early stage after stomach-pancreas-spleen resection, the rabbit serum albumin concentration was reduced, microvascular albumin leakage was detected by confocal microscopy, and microvascular endothelial cellular spaces increased based on TEM analyses. In contrast, the early serum albumin concentration in rabbits after partial jejunectomy showed no significant decrease, the serum concentrations of IL-6 and TNF-α increased slightly, no mesenteric microvascular endothelial spaces were opened, and mesenteric microvascular albumin leakage was not significantly enhanced. The microvascular leakage rate was positively correlated with the concentration of inflammatory mediators, suggesting that after major abdominal surgery, microvascular albumin leakage occurs initially, and the early postoperative reduction in serum albumin is related to microvascular albumin leakage, while the occurrence of microvascular leakage is related to inflammatory mediators. This study provides a basis for the development of clinical therapeutic strategies for hypoalbuminemia in the early stage after major surgeries.

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Disclosure of conflict of interest

None.

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References

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