

Type II endoleak model creation and intraoperative aneurysmal sac embolization with *n*-butyl cyanoacrylate-lipiodol-ethanol mixture (NLE) in swine

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Background: The purpose of this study was to evaluate the feasibility of type II endoleak model creation and efficacy of intraoperative aneurysmal sac embolization using *n*-butyl-2-cyanoacrylate-lipiodol-ethanol mixture (NLE) for type II endoleak in swine.

Methods: In six swine (mean body weight 53.5 kg), abdominal aortic aneurysm (AAA) was created and then end-to-side anastomosis between the left renal artery and AAA sac was performed. And then, endovascular abdominal aortic aneurysm repair (EVAR) was performed, leading to creation of a type II endoleak model. As control group, EVAR without sac embolization was performed in two swine. In four swine, AAA sac was embolized using NLE immediately after EVAR via the microcatheter placed in AAA sac (NLE embolization group). Follow-up aortography was performed immediately and three days after the procedure, and then the aneurysms were extracted.

Results: The AAA sac and type II endoleak model were successfully created in all cases. In control group, type II endoleak persisted three days after the procedure. In NLE embolization group, endoleak disappeared immediately and three days after the procedure. In NLE embolization group, AAA sac was occupied with thrombus and embolic material. Inflammatory changes were recognized in aneurysmal sac wall in NLE embolization group.

Conclusions: This experimental study suggests that creation of a type II endoleak model in swine is feasible and that intraoperative AAA sac embolization with NLE during EVAR might reduce the occurrence of type II endoleak.

Keywords: Type II endoleak model; abdominal aortic aneurysm (AAA); intraoperative AAA sac embolization; *n*-butyl cyanoacrylate; *n*-butyl-2-cyanoacrylate-lipiodol-ethanol mixture (NLE)

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Introduction

Endovascular abdominal aortic repair (EVAR) is widely performed as a less-invasive alternative method for patients with abdominal aortic aneurysm (AAA) who are not candidates for conventional surgical open repair. EVAR has an early survival benefit compared to open repair, including

shorter hospital stay, less blood loss, shorter surgical duration, and lower morbidity and mortality rates (1-3). However, it has been recently reported that EVAR has no long-term survival benefit compared to open repair, and that there are also significantly higher risks of reintervention and aneurysm rupture after EVAR than open repair (4-6).

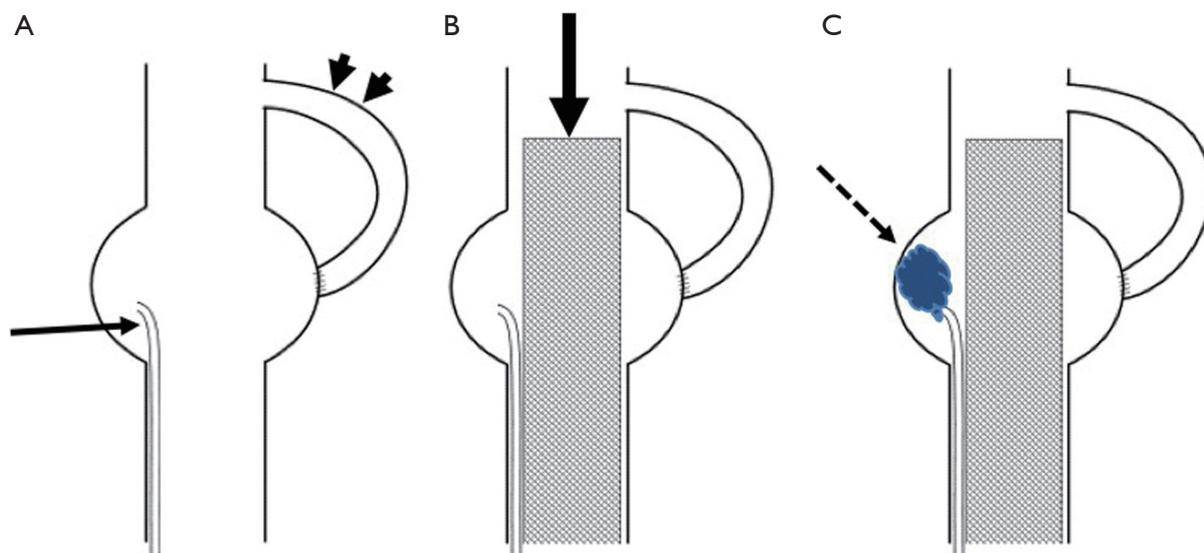


Figure 1 Schema of creation of abdominal aortic aneurysm (AAA) and type II endoleak model, and intraoperative AAA sac embolization. (A) AAA was created by using inferior vena cava. End-to-side anastomosis between the left renal artery (black arrow heads) and AAA sac was performed. A 2.7-Fr microcatheter (thin black arrow) was inserted into the AAA sac. (B) After the microcatheter was inserted into the AAA sac, the stent-graft (thick black arrow) was deployed into the abdominal aorta covering the AAA and a type II endoleak model was achieved. (C) Finally, AAA sac was embolized with NLE (dotted line arrow) via the microcatheter inserted into aneurysmal sac.

Patients who had EVAR need lifelong surveillance after EVAR. Endoleak and aneurysm sac expansion after EVAR are significant problems that need to be overcome. Type II endoleak often spontaneously disappears, but 10–25% remain (7–10). Persistent type II endoleak is strongly associated with aneurysm sac enlargement (11–13). Re-intervention is mandatory for endoleak with sac expansion.

Intraoperative aneurysmal sac embolization during EVAR can be one of the options to prevent type II endoleak (14–16). N-butyl cyanoacrylate (NBCA, Histoacryl, B. Braun, Melsungen, Germany) is one of liquid embolic materials. A mixture of NBCA with Lipiodol (Guerbet, Villepinte, France) and ethanol (Fuso Pharmaceutical Industries Ltd, Osaka, Japan) (NBCA-lipiodol-ethanol; NLE) is less adhesive embolic material than NBCA (17). The aim of this study was to evaluate the feasibility of type II endoleak model creation in swine and efficacy of intraoperative AAA sac embolization during EVAR using NLE for type II endoleak.

Methods

The approval of the Institutional Committee on Research-Animal Care was obtained before initiation of the study.

Six healthy female swine weighing 48–55 kg (mean body weight 53.5 kg) were prepared. Two swine were used for a control group, and 4 swine for NLE embolization group. Pre-anesthesia comprised 5 mg/kg ketamine and 80 µg/kg medetomidine, and general anesthesia was maintained with isoflurane gas via tracheal intubation. Cardiac and pulmonary parameters were monitored throughout the procedures. The technique of type II endoleak model creation consisted of three steps. That is, the first step is creation of an abdominal aneurysm using inferior vena cava (IVC). The second step is anastomosis between renal artery and aneurysm, and the final step is stent-graft placement in the abdominal aorta. First, laparotomy was performed and abdominal aorta was surgically exposed. Abdominal aneurysm was created by using IVC. A length of IVC of approximately 3 cm was removed and then sutured to an incision in the infrarenal abdominal aorta, under interruption of abdominal aortic blood flow. We created a total of 6 AAAs in 6 swine. The long × short diameters ranged from 42 mm × 22 mm to 56 mm × 26 mm (mean, 47.5 mm × 26.2 mm). Next, the left renal artery was selected as an artery causing type II endoleak. Left nephrectomy was performed and then end-to-side anastomosis between the left renal artery and AAA was created (*Figure 1A*).

A 6-Fr sheath (Long sheath, Terumo, Gifu, Japan) was placed in both surgically exposed femoral arteries after 0.035-inch guidewire (Terumo) insertion. A 4-Fr pig tail catheter (Medikit, Tokyo, Japan) was advanced into abdominal aorta via right femoral artery and aortography was performed. After that, a 2.7-Fr microcatheter (Carnelian; Tokai Medical, Aichi, Japan) was inserted into the AAA using a 0.014-inch guidewire (GT wire; Terumo). Finally, after 0.035-inch stiff guidewire (Cook Inc., Bloomington, IN, USA) was inserted via the left femoral artery, a 12-Fr sheath with the stent-graft (Zenith[®] iliac leg extension 8 mm diameter, 37 mm long) (Cook, USA) was inserted and deployed into the abdominal aorta, covering the AAA (*Figure 1B*). Aortography was performed to confirm the type II endoleak from the anastomosed left renal artery.

NLE was prepared by mixing NBCA, Lipiodol, and ethanol at a ratio of 1:5:1 by use of a three-way stopcock. The components were mixed thoroughly before injection.

As a control group, stent-graft deployment was performed without AAA sac embolization in two swine. For NLE embolization group, after stent-graft deployment, AAA sac was embolized with NLE in four swine. Before injection of NLE, we estimated the volume of the AAA sac by test injection of contrast medium. NLE was slowly injected via the microcatheter inserted into AAA sac under fluoroscopic control (*Figure 1C*). NLE was injected until the AAA sac was completely filled in with NLE. After the AAA sac was filled in with NLE, the microcatheter was removed. And then aortography was performed to confirm the presence or absence of flow into the aneurysm and the type II endoleak after embolization. Three days later, aortography was performed again to depict type II endoleak.

After the final aortography, the swine are euthanized to extract the aneurysms. After the formalin-fixation, Hematoxylin-Eosin (HE) staining was performed to confirm the thrombosis of the aneurysm, and observe the extent of the histopathological inflammatory changes of vessel wall.

Results

The AAA sac and type II endoleak model were successfully created in all cases.

In the NLE embolization group, AAA sac was completely filled in with NLE (*Figure 2*). Endoleak disappeared immediately after the procedure and no recurrence was observed three days after the procedure (*Table 1*). AAA sac

was occupied with organized thrombus, degenerate blood cells, and embolic material (*Figure 3*). Inflammatory changes were recognized in aneurysmal sac wall (*Figure 4A*).

In the control group, type II endoleak persisted three days after the procedure (*Table 1*). Inflammatory changes were almost not recognized in aneurysmal sac wall (*Figure 4B*). There was much more infiltration of inflammatory leukocytes into the vessel wall in the NLE embolization group than in the control group as shown with HE stain specimens.

Discussion

Endoleak after EVAR are classified into four types (18). Type I endoleak is characterized by arterial blood flow into the aneurysmal sac from the proximal or distal attachment site of the stent-graft as a result of inadequate fixation. Type II endoleak, the most common type, is characterized by blood flow in a retrograde manner into the aneurysmal sac through patent branch vessels of the abdominal aorta, such as lumbar artery (LA) or inferior mesenteric artery (IMA). Type III endoleak is characterized by blood leak from a modular disconnection or graft fabric tear. Type IV endoleak is characterized by sac perfusion from blood permeation through the graft material, which results from excessive graft porosity. The mechanism of type II endoleak is thought to be that backflow from the aortic branch vessel occurs by the sac pressure reduction by covering the aneurysm with the stent-graft. We succeeded in creating a type II endoleak model by anastomosing the renal artery with the aneurysm. After stent-graft placement into the abdominal aorta covering the AAA sac, a type II endoleak model was achieved.

NBCA is a liquid adhesive composed of monomers, and polymerizes into a solid material following contact with anions within blood. To achieve radio-opacity, NBCA is usually mixed with iodized oil (Lipiodol). Polymerization time depends on the dilution ratio of these two components (19-22). NBCA is increasingly being used to treat acute arterial hemorrhage and pseudoaneurysms because its rapid polymerization in blood results in complete and instantaneous occlusion of the bleeding vessel (23-25). However, because of the strong adhesive properties of NBCA, NBCA has some concerns such as adhesion to the catheter and vessel wall, and early polymerization of NBCA within the catheter and proximal portion of the target vessel, leading to incomplete embolization.

The characteristics of NLE have been shown to be

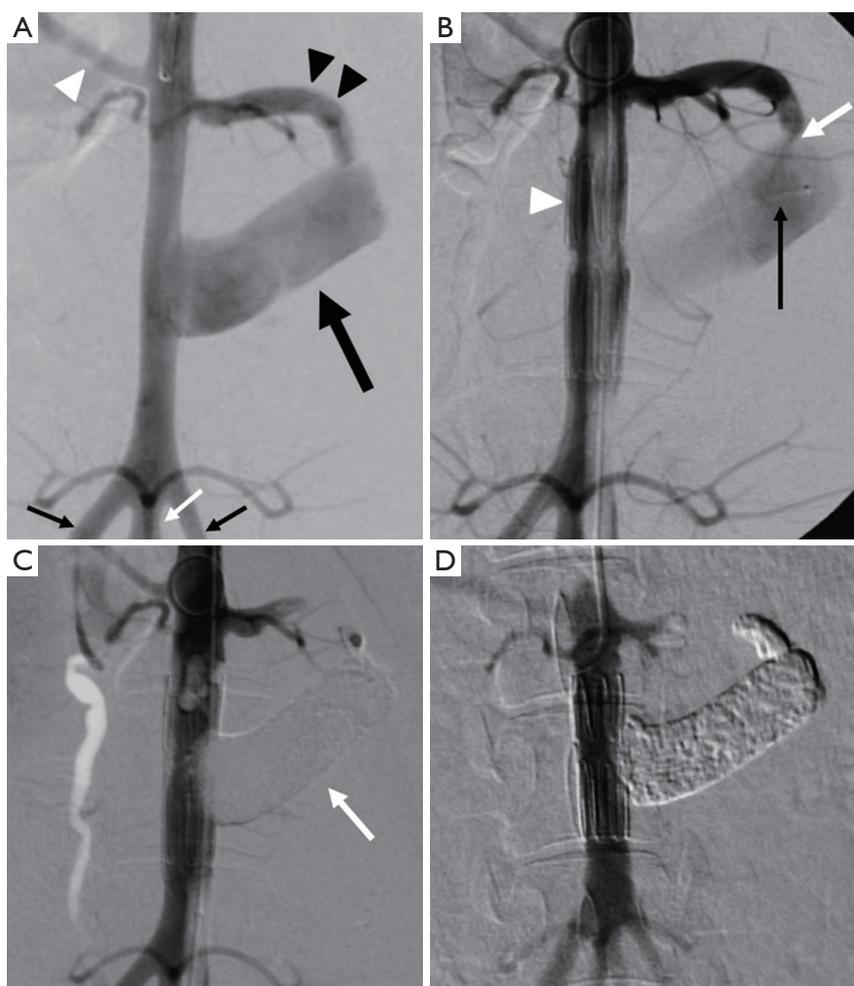


Figure 2 Angiography of NLE embolization group. (A) Aortography after abdominal aortic aneurysm (AAA) creation and anastomosis with the left renal artery and AAA sac. AAA was created by using inferior vena cava (thick black arrow). End-to-side anastomosis between the left renal artery (black arrow heads) and AAA sac was performed (white arrow head: right renal artery; thin black arrow: external iliac artery; thin white arrow: internal iliac artery). (B) Aortography after stent-graft deployment. After a microcatheter (thin black arrow) was inserted into the AAA sac, the stent-graft (Zenith® iliac leg extension 8 mm diameter, 37 mm long) (white arrow head) was deployed into the abdominal aorta with covering the AAA. Type II endoleak from the left renal artery was recognized (thin white arrow). (C) Aortography immediately after sac embolization with NLE. AAA sac was embolized and completely filled with NLE (white arrow). (D) Aortography after 3 days. Type II endoleak disappeared.

different from those of the NBCA-lipiodol mixture (NL) (17,26-28). NLE may have multiple advantages over NL for embolization, such as less adhesive properties to the catheter and less damage features to the vascular wall (26,27). NLE doesn't adhere to the balloon catheter and showed the feasibility of packing a wide-neck aneurysm (26,27). Ishikawa *et al.* reported that NLE showed consistent and reproducible complete embolization with flow control compared to NL and was stable after balloon deflation (28).

In this study, NLE with a ratio of 1:5:1 was used as the embolic material. We determined the ratio of the components of the embolic material based on our clinical experiences (29). Our mixing ratio enabled us to achieve complete packing of the AAA sac. We believe that this mixing ratio was appropriate for AAA sac embolization, considering adhesion of NLE glue and safety, although the optimum ratio of NLE may still be controversial. The optimum ratio of NLE might need to be adjusted depending

Table 1 Post-embolization angiographic results in swine

No.	Group	Sac diameter		NLE (mL)	Endoleak	
		Long axis	Short axis		Immediately after stent-graft deployment	3 days after embolization
1	Control	42	22	–	Type II	Type II
2	Control	48	26	–	Type II	Type II
3	NLE	56	26	4.5	Type II	Disappear
4	NLE	45	28	3	Type II	Disappear
5	NLE	50	28	3.5	Type II	Disappear
6	NLE	44	27	3	Type II	Disappear

NLE, *n*-butyl cyanoacrylate-lipiodol-ethanol mixture.



Figure 3 Macroscopic findings in NLE embolization group. AAA sac was occupied with organized thrombus, degenerate blood cells, and embolic material (black arrow). AAA, abdominal aortic aneurysm; NLE, *n*-butyl cyanoacrylate-lipiodol-ethanol mixture.

on the size of the AAA sac and its flow. The behavior of NLE was safely monitored under fluoroscopic guidance. NLE can be slowly injected in a controlled manner, and the procedure does not require rapid removal of the catheter, unlike with standard NL mixture. NLE induced thrombosis of the aneurysmal sac, and this procedure eliminated the type II endoleak.

There are some reports on embolization for the sac and endoleak using other liquid embolic materials such as thrombin or Onyx (LES, Covidien, Plymouth, MN, USA) (14,30). However, the difficulty of monitoring thrombin progression may be at risk of non-target vessel embolization. Furthermore, the injected thrombin is usually completely reabsorbed within several days as a result of fibrinolysis and tissue plasminogen reactions (31), possibly leading to less good occlusive properties. Onyx needs more time for preparation and is more painful at the time of injection. Additionally, it has more cytotoxic

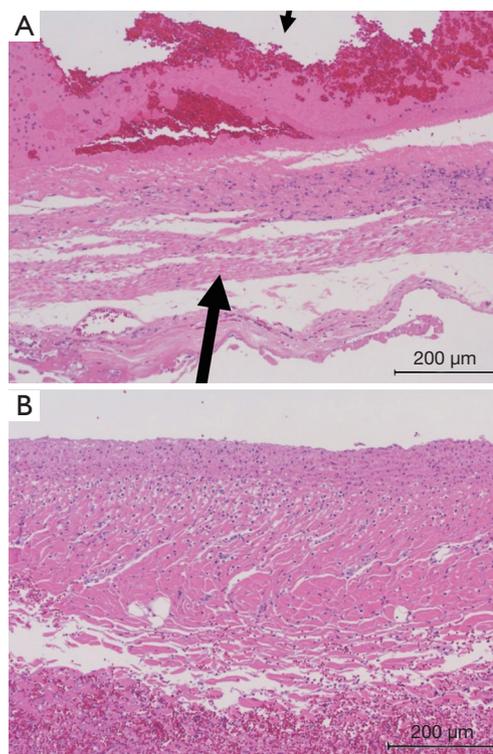


Figure 4 Microscopic findings in the aneurysm wall (Hematoxylin and Eosin stain). (A) NLE embolization group. There was much infiltration of inflammatory leukocytes into the vessel wall (thick black arrow) (thin black arrow: thrombus in the lumen of aneurysm). (B) Control group. There was few infiltration of inflammatory leukocytes into the vessel wall.

effect than NBCA due to dimethyl sulfoxide (DMSO) (32). Intraoperative aneurysmal sac embolization needs too much liquid embolic material, so a large amount of onyx has concern about toxicity. Therefore, we think the use of NLE

is quite easier and better.

The limitation of our study is to have used the left renal artery to create a type II endoleak model. The renal artery provides more blood flow into the sac than thin aortic branches such as LA and IMA which commonly cause type II endoleak. That is, the hemodynamic of this endoleak model differs slightly from that of the naturally occurring endoleak. However, this study showed that even endoleak caused by such thick artery could be embolized by NLE. Another limitation is the relatively short follow-up period after endovascular procedure. We could not evaluate about the recurrence of endoleak after long term follow-up from the viewpoint of animal welfare.

In conclusion, this experimental study suggests that creation of a type II endoleak model in swine is feasible and that intraoperative AAA sac embolization with NLE during EVAR might reduce the occurrence of type II endoleak.

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Footnote

Conflicts of Interest: The authors have no conflicts of interest to declare.

Ethical Statement: This experimental study was performed in compliance with the requirements of the institutional review board and approved by the institution ethical committee on research animal care.

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