Supplementary Materials

Supplementary Methods

Intraoperative heparin administration. Twice during surgery, the activated clotting time (ACT) was measured to titrate heparin administration. The two points included first, before ligation of the right proximal CCA and during administration of heparin (Heparin Bichsel AG, Switzerland) bolus 500 international units (IU)/kg, and second after reopening all arterial clamps.

Arterial wall degradation protocols. Arterial pouches from a donor animal (allografts from both CCAs) were decellularized in 1% SDS for 6 hours at 37° C. Until ready for use, decellularized donor grafts were stored in phosphate-based saline buffer (0.1 molar) at a potential hydrogenii from 7.4 at -70° C. For elastase degradation, the arterial pouch (autograft from the right CCA) was meticulously cleaned from soft tissue intraoperatively, incubation with 100 Units porcine elastase (Sigma Aldrich, Nr. 45124/45125, Switzerland) dissolved in 5 milliliter (ml) TRIS-Buffer (Sigma Aldrich, Nr. 93302, Switzerland) followed on the day of the experiment for 20 minutes with gentle shaking at room temperature.

Anesthesia protocol. Rabbits were sedated with ketamine (Narketan 100 mg/ml, Vétoquinol, Germany) 20 mg/kg sc, dexmedetomidine 100 mcg/kg (Dexdomitor 0.5 mg/ml, Orion Pharma, Finland) and methadone 0.3 mg/kg (Methadon Streuli, 10 mg/ml, Streuli pharma, Switzerland), and left undisturbed for 10-15 minutes. Once sedation was adequate, oxygen supplementation via face-mask was started and continued until tracheal intubation was performed. A venous catheter and arterial catheter were placed, respectively, in the auricular vein and in the

contralateral auricular artery. For anesthesia induction and tracheal intubation, propofol (Propofol 1% MCT Fresenius, Fresenius Kabi, Germany) 1-3 mg/kg intravenously (iv) was administered to effect. Tracheal intubation was performed with a silicone endotracheal tube (internal diameter 3 mm), which was then connected to a low-resistance pediatric rebreathing system and ventilation mode (spontaneous, not mechanical) was individually selected to avoid uncontrolled hypercapnia ($PaCO_2 \geq 55$ mmHg). Unawareness was maintained with isoflurane (Isofluran Baxter AG, Switzerland) in 100% oxygen targeting an end tidal of 1.8 %. A forced-air warming device was used to ensure normothermia. Intraoperative monitoring consisted of the following: rectal temperature, ECG (II lead), pulse oxymetry, inspired and expired gas, spirometry, invasive blood pressure, and indexed EEG (bispectral index). Balanced analgesia was provided using peri-incisional infiltration of ropivacaine (Ropivacain 2 mg/ml, Fresenius Kabi, Switzerland) 0.75% 2-3 mg/kg before surgery. Intraoperative analgesia consisted of continuous rate infusion (CRI) of lidocaine (Lidocaine 1% HCl, Streuli Pharma, Switzerland) 50 mcg/kg/min and fentanyl (Fentanyl 50 mg/ml, Jannsen, Belgium) (3-10 mcg/kg/h). Fluid therapy was administered from induction of general anesthesia until rabbits recovered sternal recumbency and consisted of Ringer lactate 3-5 ml/kg/h.

Postoperative care. When the rabbit regained spontaneous swallowing, tracheal extubation was performed. This was followed by administration of meloxicam (Metacam 5 mg/ml, Boheringer Ingelheim, Switzerland) 0.5 milligram (mg)/kilogram (kg) intravenously (iv), ASA 10 mg/kg iv, vitamin B_{12} (Vitarubin, Streuli Pharma, Switzerland) 100 mcg subcutaneously (sc), and amoxicillin and clavulanic acid (Clamoxyl 250 mg, Glaxo Smith AG, Switzerland) 20 mg/kg iv. Supplemental

oxygen via face mask was delivered until the rabbits regained spontaneously sternal recumbency. Normothermia was ensured by a forced-air warming device.

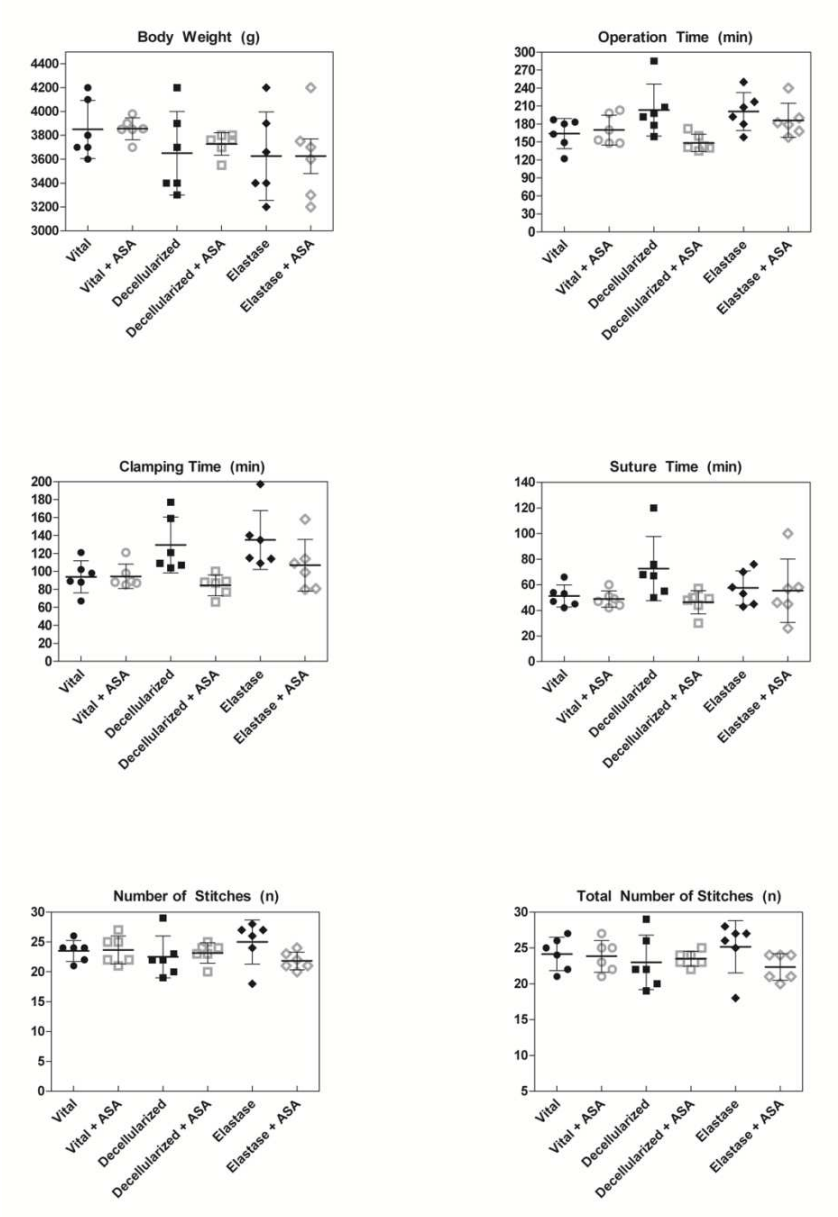
Pain scoring and postoperative follow-up care were performed for 3 days in accordance with the guidelines for the assessment and management of pain in rodents and rabbits. Postoperative analgesia consisted of meloxicam for 3 days sc and a fentanyl patch (Durogesic 12 mcg/h, Janssen, Belgium) placed on the outer ear for 72 hours. As rescue analgesia, an injection of methadone (Methadon Streuli, 10 mg/ml, Streuli Pharma, Switzerland) 0.2 mg/kg was planned. For the ASA group, 500 mg powder (Sanofi-Aventis, Switzerland) was dissolved in the drinking water until the day of the final MRI. Low molecular weight heparin (250 IU/kg) for all groups was administered for 3 days.

MRI protocol. Prior to MRI, 2 peripheral venous accesses (marginal auricular veins) were established in each rabbit. Following premedication (see Methods in paper) and transport, ketamine 15 mg/kg and medetomidine (Domitor, 1 mg/ml, Vétoquinol, Germany) 0.1 mg/kg were injected intramuscularly or subcutaneously prior to MRI, according to the individual awareness after premedication. Ten to 20 minutes after injection, ketamine CRI (5mg/kg/h) was started: dosage was adapted to guarantee immobility and unawareness with progressive increases up to 15 mg/kg. The CRI was preceded by an intravenous bolus of ketamine 1-3 mg/kg if unawareness was not achieved with the combination of ketamine-medotimidine. Imaging sequences acquired on a 3 T Magnetom Skyra (Siemens) included T2 space sagittal, T2 dark fluid fs sagittal, transversal DWI, transversal T1 fs, TOF angiography, neck angiography with contrast enhancement, T1 mprage sagittal fs with contrast enhancement, and transversal T1 fs.

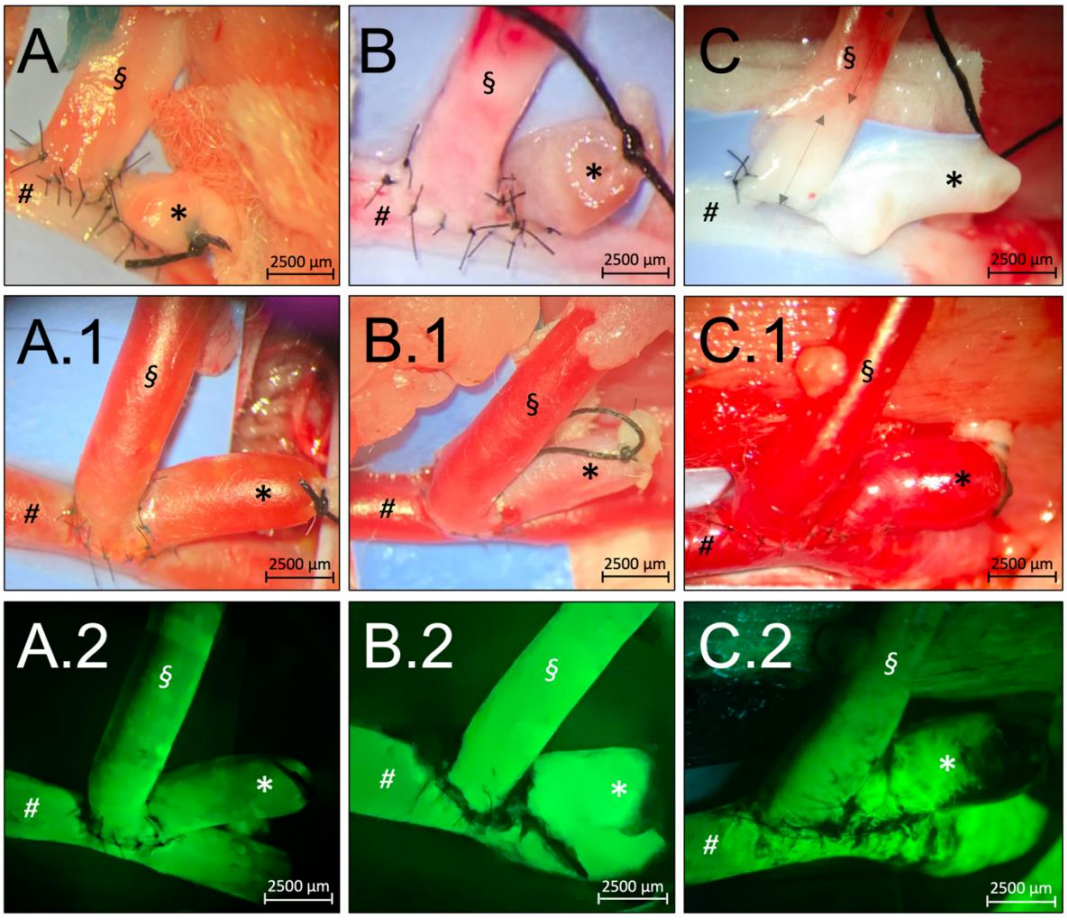
Histological Grading System. Characteristics assessed and scored included: Periadventitial inflammation (0 = none, 1 = mild, 2 = moderate, 3 = severe); periadventitial fibrosis (0 = none, 1 = mild, 2 = moderate, 3 = severe); aneurysm wall inflammation (0 = none, 1 = few (1-3) spots, 2 = many (>4) spots, 3 = ubiquitous); aneurysm wall hematoma (0 = none, 1 = few (1-3) spots, 2 = many (>4) spots, 3 = ubiquitous); aneurysm wall cellularity (0 = none, 1 = few (1-3) spots, 2 = many (>4) spots, 3 = ubiquitous); aneurysm wall dissection (0 = none, 1 = few (1-3) spots, 2 = many (>4) spots, 3 = ubiquitous); endothelial cellularity (0 = none, 1 = few (1-3) spots, 2 = many (>4) spots, 3 = ubiquitous); luminal thrombus (0 = absent, 1 = present); neutrophils in the thrombus (0 = none, 1 = mild, 2 = moderate, 3 = severe); and neointima formation (0 = none, 1 = organizing thrombus, 2 = organizing thrombus and neointima formation, 3 = mature neointima). Scores were dichotomized as (1) (none/mild) and moderate/severe; (2) no/few cells and focal hypo-cellularity/normal cell count; and (3) no neointima/organizing thrombus and organizing neointima/mature neointima.

Supplementary Figures

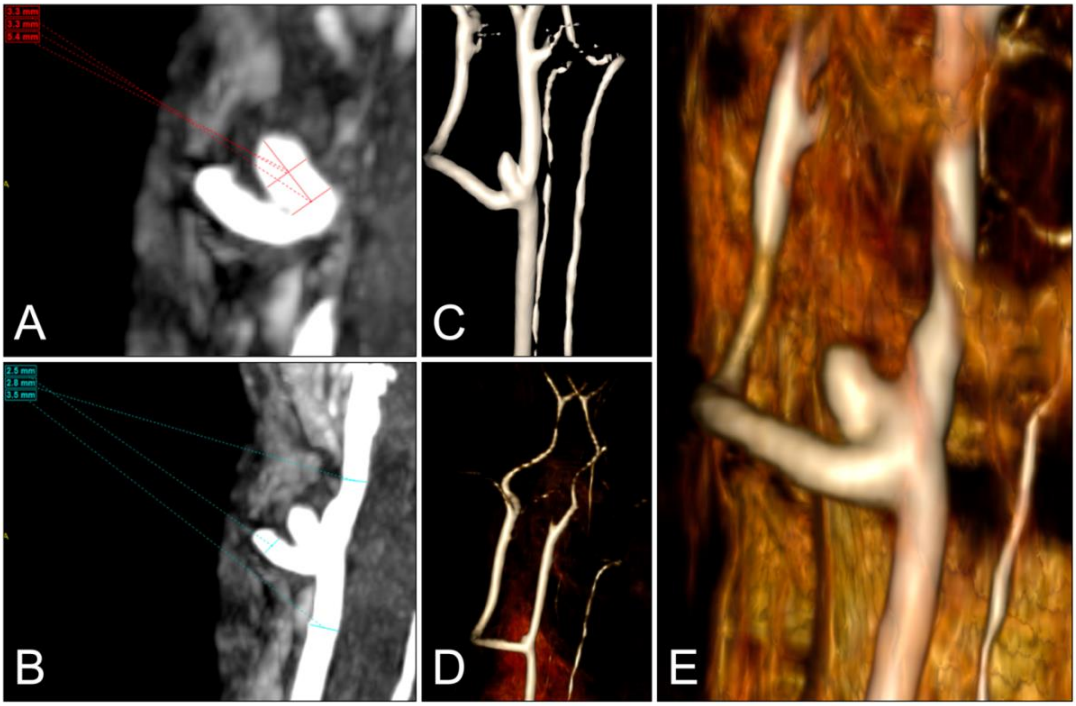
Supplementary Figure 1. Bar charts for surgical characteristics. Body weight in gram. Clamping, operation, and suture times in minutes. Total number of stitches includes stitches and restitches because of an initial insufficient anastomosis.



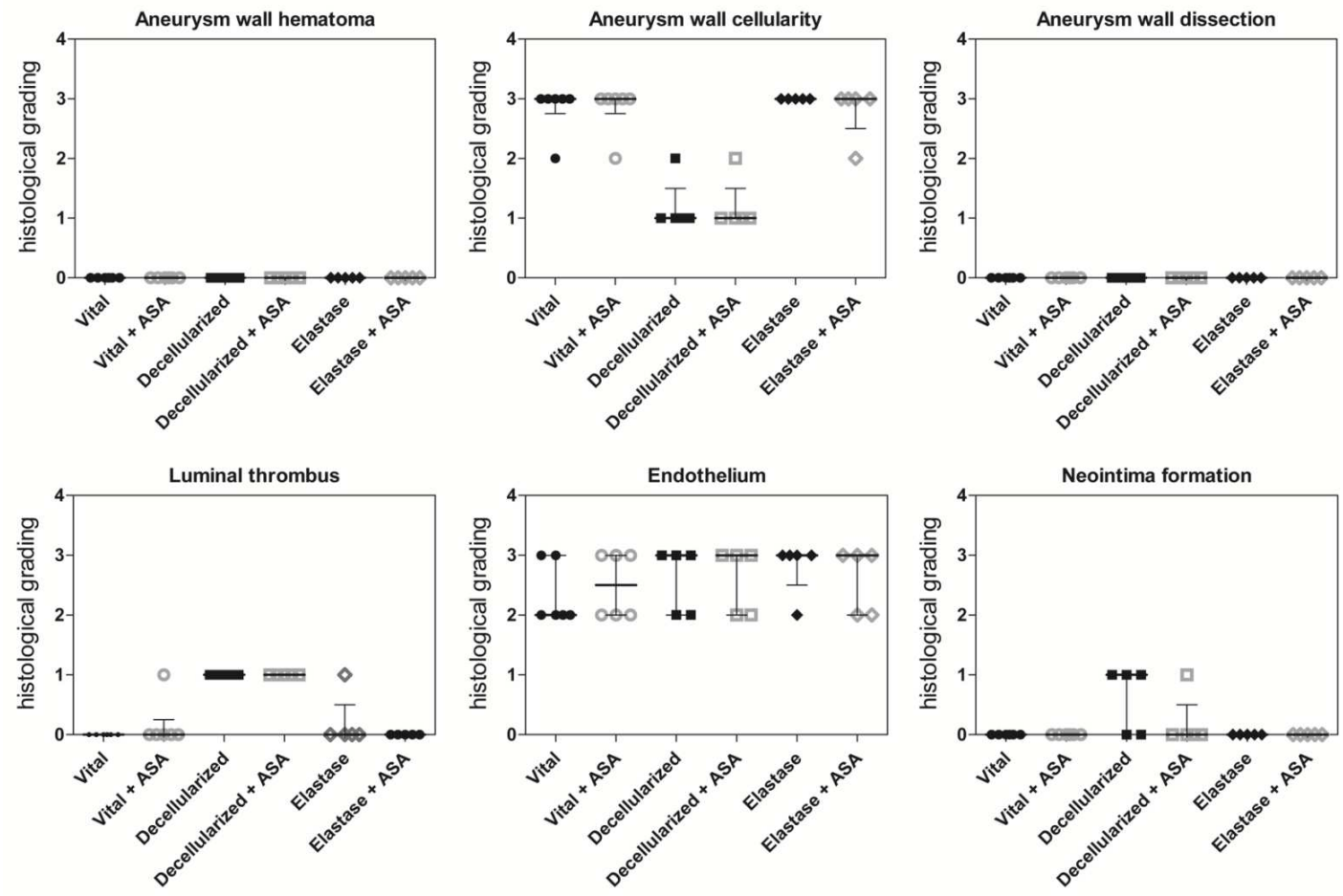
Supplementary Figure 2. Photographs illustrating the macroscopic findings. Intraoperative situs upon aneurysm creation of vital, (A) decellularized (B), and elastase-degraded (C) vessel pouches. Intraoperative macroscopic findings after opening the arterial clamp show aneurysm perfusion in vital (A.1), decellularized (B.1), and elastase-degraded aneurysms (C.1). Intraoperative flow assessment after administration of fluorescein in vital (A.2), decellularized (B.2), and elastase-degraded (C.1) aneurysms. Legend: § right CCA, # left CCA, * aneurysm pouch. Note: (1) dotted and non-dotted arrows in (C) show the aggressive co-effect of elastase preincubated pouches on the angioarchitecture pronounced proximally to the anastomosis. (2) kinking of the elastase-degraded pouch in (C.1) compared to decellularized (B.1) and vital pouches (A.1).



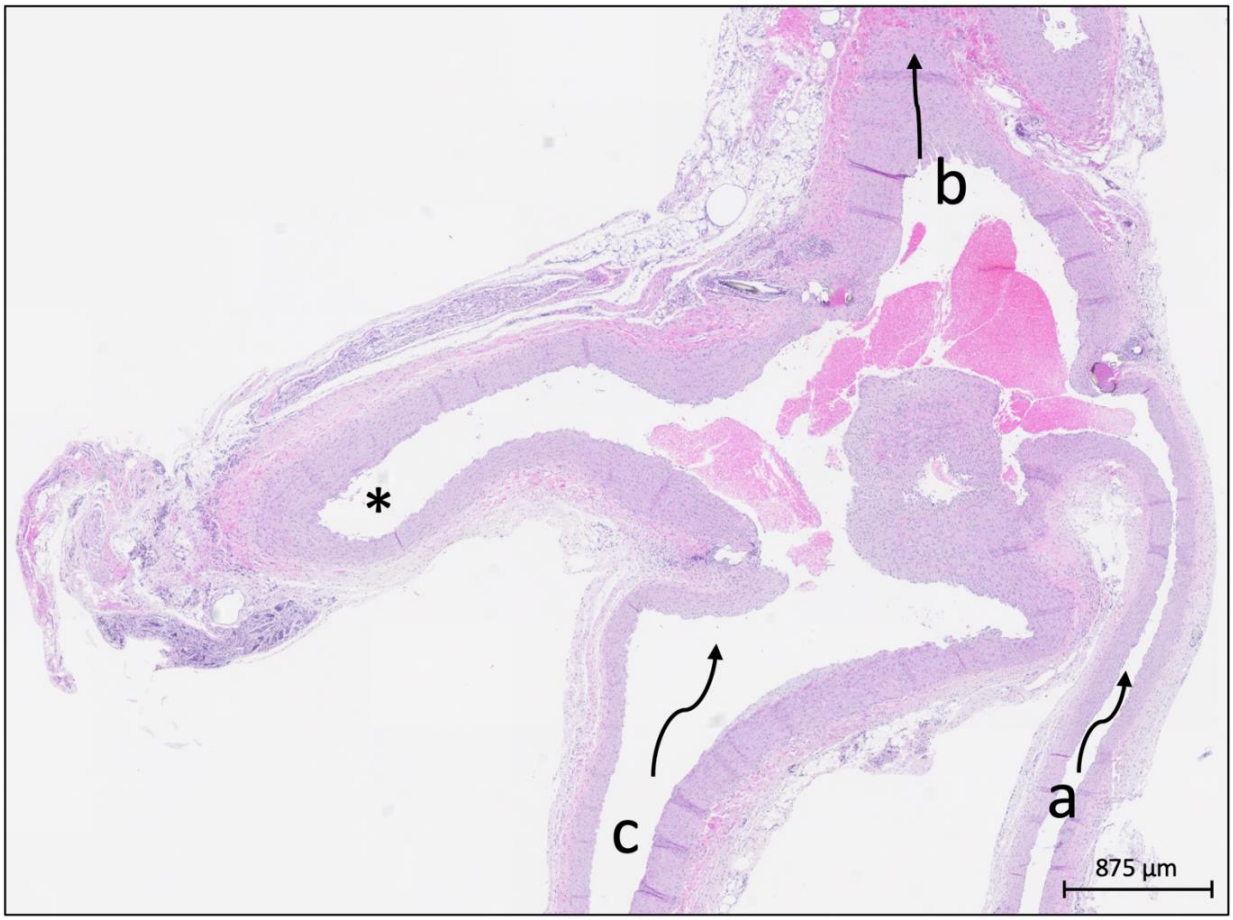
Supplementary Figure 3. Illustrative panel of MR Angiography including CE-3D-MRA-morphometric measurements. Example of maximum intensity projections (8.5) of a vital arterial pouch aneurysm at 28-day follow-up. (A) Measurement of aneurysm height, width and depth. (B) Diameter of the corresponding parent arteries in the sagittal view. (C-E) show 3D-MRA reconstructions of vital, decellularized and elastase-degraded arterial pouches, respectively.



Supplementary Figure 4. Detailed histological grading. Comparison of various parameters in the vital, decellularized, and elastase-degraded groups without and with ASA treatment (0 = none, 1 = mild, 2 = moderate, 3 = severe).

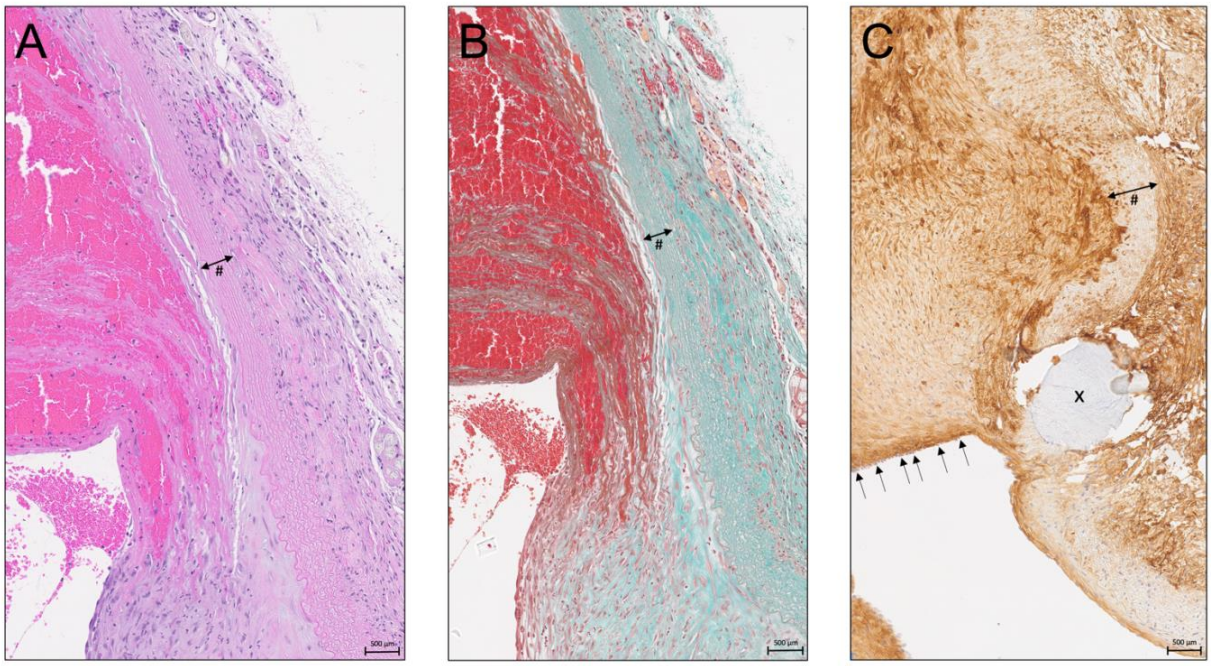


Supplementary Figure 5. Microphotograph of a cross-section shows a vital aneurysm (*) and its parent artery complex without ASA treatment. Proximal left (a), distal left (b), and right CCA (c) are marked.



Supplementary Figure 6. Representative microphotographs depicting thrombus formation.

Note the apical thrombus formation with consecutive neointima formation and endothelial lining from the lumen of a decellularized aneurysm without ASA treatment. Staining with HE (A), MASA (B) and F8 (C) are shown. Cell-depleted muscular media (#). (C) endothelial cells demonstrating neointima formation (black arrows). Crosscut (x) of the underlying 9-0 non-absorbable suture material.



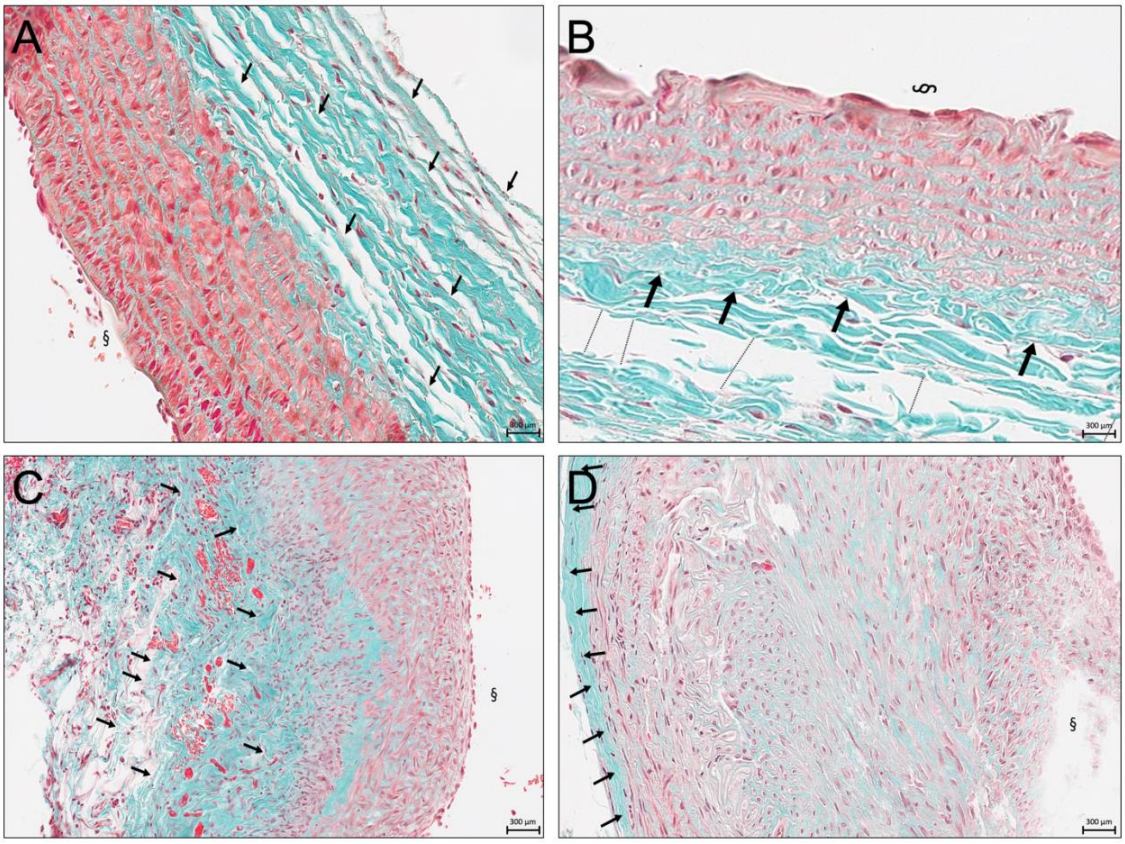
Supplementary Figure 7. Representative microphotographs depicting periadventitial fibrosis. Different magnifications of MASA stained histological samples of vital and elastase-degraded aneurysms treated without and with ASA. Collagen fibers (green) and blood (red). Scale bar: 300 μm.

(A) Adventitia in a vital aneurysm without ASA treatment. Note the intense periadventitial fibrosis (black arrows).

(B) Masson-Goldner trichrome staining of the adventitia in vital aneurysm with ASA pretreatment (§ shows the luminal side, black arrows the periadventitial fibrosis). Note the rarefication of collagen fibers (dotted line) compared to (A). Scale bar: 300 μm.

(C) Masson-Goldner trichrome staining of the adventitia in an elastase treated aneurysm without ASA (§ shows the luminal side of the aneurysm). Note the intensive fibrosis (black arrows), likewise the diffuse infiltration of neutrophils in between (black arrowheads). Scale bar: 300 μm.

(D) Masson-Goldner trichrome staining of the adventitia in an elastase treated aneurysm with ASA. (D) shows a 40-fold magnification of the outer periadventitia. Note the diminished periadventitial fibrosis (black arrows) compared to (C). Furthermore, no neutrophils can be detected. Scale bar: 300 μm.


A
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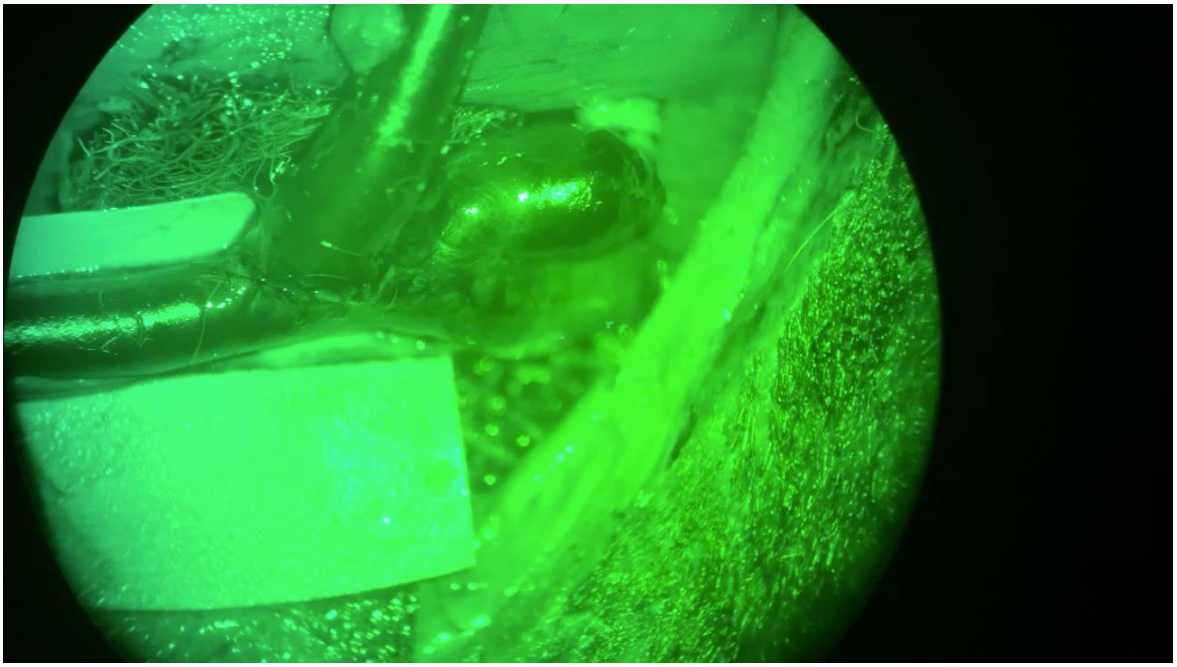
Supplementary Videos

Supplementary Video 1. Fluorescence angiography. Video shows an elastase- degraded arterial pouch with consecutive fluorescein application to confirm intraoperative patency.



***Supplementary Table 1*.** Surgery times averaged 164.0 ± 25.0 min (range 122-187) with 51.2 ± 8.6 min (range 42-66) needed for aneurysm creation in the vital group w/o ASA and 170.0 ± 25.0 min (range 148-203) with 48.8 ± 6.4 min (range 42-60) for the w ASA group. Duration of the surgical procedure in the decellularized group w/o ASA was 203.3 ± 43.5 min (range 159-285), for aneurysm creation 72.7 ± 25.0 min (range 50-120); for the w ASA group 148.5 min ± 14.3 min (range 135-172) and 46.3 min ± 9.1 min (range 30-57) were needed. For the elastase w/o ASA group, values were 200.8 min ± 31.9 min (range 158-250 minutes) for surgery in whole, for aneurysm creation 57.5 min ± 13.3 min (range 43-76); the w ASA group values were 186.2 min ± 28.7 min (range 158-240), for aneurysm creation 55.3 min ± 24.7 min (range 26-100).

(A). Surgical characteristics and morphometric measurements of vital aneurysms without medical treatment**.** Values are expressed as means ± SD. * $p_{Vol(BL-FU)} < 0.05$.

n	***Weight (g)***	***Ischemia (min)***	***Suture time (min)***	***Stitches (#)***	***Re-stitches (#)***	***Surgery (min)***	***Volume baseline (mm^3)***	***MRT (days)***	***Volume follow-up (mm^3)***	***CCA left proximal (mm)***	***CCA left distal (mm)***	***CCA right (mm)***
1	4100.00	98.00	54.00	24	0	187.00	1.96	24	5.00	2.50	2.80	1.80
2	4200.00	121.00	53.00	24	1	183.00	2.35	23	7.73	3.30	2.90	2.50
3	3800.00	102.00	66.00	26	0	163.00	4.71	37	28.03	3.40	3.00	2.60
4	3600.00	67.00	42.00	22	0	122.00	6.28	37	47.37	2.80	2.80	3.20
5	3700.00	88.00	45.00	24	3	180.00	10.99	22	15.82	3.20	3.00	2.80
6	3700.00	89.00	47.00	21	0	149.00	12.56	31	15.11	2.30	2.20	1.90
Mean ± SD	**3850.00 ± 242.90**	**94.17 ± 17.88**	**51.17 ± 8.61**	**23.50 ± 1.76**	**0.67 ± 1.21**	**164.00 ± 25.04**	**6.48 ± 4.43**	**29.00 ± 6.96**	**19.85 ± 15.69***	**2.92 ± 0.45**	**2.78 ± 0.30**	**2.47 ± 0.54**

Abbreviations: CCA (common carotid artery); CE-3D-MRA (contrast enhanced 3D magnetic resonance angiography); g (gram); min (minute/s); mm^3 (cubic millimeter)

(B). Surgical characteristics and morphometric measurement of vital aneurysms with ASA treatment**.** Values are expressed as means ± SD.

n	***Weight (g)***	***Ischemia (min)***	***Suture time (min)***	***Stitches (#)***	***Re-stitches (#)***	***Surgery (min)***	***Volume baseline (mm^3)***	***MRT (days)***	***Volume follow-up (mm^3)***	***CCA left proximal (mm)***	***CCA left distal (mm)***	***CCA right (mm)***
1	3900.00	89.00	44.00	25	0	148.00	12.56	26	14.13	2.70	2.40	2.40
2	3855.00	98.00	51.00	21	0	198.00	9.42	23	10.72	2.50	2.80	2.30
3	3700.00	87.00	47.00	27	0	148.00	11.77	23	17.00	3.30	2.70	2.60
4	3850.00	121.00	60.00	22	0	203.00	7.06	24	23.17	2.60	2.60	2.70
5	3850.00	88.00	49.00	22	1	170.00	8.24	26	14.36	2.90	2.80	2.30
6	3980.00	85.00	42.00	25	0	153.00	10.99	26	4.04	2.50	2.40	2.20
Mean ± SD	**3855.00 ± 91.35**	**94.67 ± 13.66**	**48.83 ± 6.37**	**23.67 ± 2.34**	**0.17 ± 0.41**	**170.00 ± 25.02**	**10.01 ± 2.13**	**24.67 ± 1.51**	**13.91 ± 6.38**	**2.75 ± 0.31**	**2.62 ± 0.18**	**2.42 ± 0.19**

(C). Surgical characteristics and morphometric measurement of decellularized aneurysms without medical treatment**.** Values are expressed as means ± SD.

n	***Weight (g)***	***Ischemia (min)***	***Suture time (min)***	***Stitches (#)***	***Re-stitches (#)***	***Surgery (min)***	***Volume baseline (mm^3)***	***MRT (days)***	***Volume follow-up (mm^3)***	***CCA left proximal (mm)***	***CCA left distal (mm)***	***CCA right (mm)***
1	3400.00	159.00	120.00	22	0	285.00	8.83	83	14.44	3.10	2.50	2.30
2	3900.00	177.00	76.00	19	0	208.00	4.71	33	4.00	2.70	2.40	2.30
3	4200.00	121.00	68.00	22	0	198.00	20.60	29	14.34	2.70	2.70	2.70
4	3300.00	107.00	50.00	23	3	192.00	6.28	40	19.62	3.10	3.30	2.80
5	3400.00	109.00	55.00	29	0	159.00	11.77	40	19.62	3.40	2.90	2.70
6	3700.00	104.00	67.00	20	0	178.00	7.06	41	14.56	-	-	-
Mean ± SD	**3650.00 ± 350.71**	**129.50 ± 30.91**	**72.67 ± 25.03**	**22.50 ± 3.51**	**0.50 ± 1.23**	**203.33 ± 43.49**	**9.88 ± 5.79**	**44.33 ± 19.53**	**14.43 ± 9.02**	**3.00 ± 0.30**	**2.76 ± 0.36**	**2.56 ± 0.24**

(D). Surgical characteristics and morphometric measurements of decellularized aneurysms with ASA treatment. Values are expressed as means ± SD.

n	***Weight (g)***	***Ischemia (min)***	***Suture time (min)***	***Stitches (#)***	***Re-stitches (#)***	***Surgery (min)***	***Volume baseline (mm^3)***	***MRT (days)***	***Volume follow-up (mm^3)***	***CCA left proximal (mm)***	***CCA left distal (mm)***	***CCA right (mm)***
1	3760.00	100.00	57.00	24	0	160.00	12.56	26	18.84	2.50	1.90	2.10
2	3800.00	77.00	49.00	24	0	143.00	15.70	25	11.77	2.60	2.60	2.80
3	3760.00	87.00	50.00	25	0	140.00	15.70	24	17.66	2.80	2.50	2.00
4	3550.00	89.00	48.00	20	2	172.00	15.70	26	10.99	3.00	2.80	2.60
5	3700.00	88.00	44.00	23	0	141.00	9.42	24	14.13	2.80	2.90	2.60
6	3800.00	66.00	30.00	23	0	135.00	12.56	31	34.34	3.20	3.30	2.50
Mean ± SD	**3728.33 ± 94.75**	**84.50 ± 11.64**	**46.33 ± 9.05**	**23.17 ± 1.72**	**0,33 ± 0,82**	**148.50 ± 14.32**	**13.61 ± 2.56**	**26.00 ± 2.61**	**17.95 ± 8.61**	**2.82 ± 0.26**	**2.67 ± 0.47**	**2.43 ± 0.31**

(E). Surgical characteristics and morphometric measurements of elastase aneurysms without medical treatment**.** Values are expressed as means ± SD. * $p_{Vol(BL-FU)} < 0.05$.

n	***Weight (g)***	***Ischemia (min)***	***Suture time (min)***	***Stitches (#)***	***Re-stitches (#)***	***Surgery (min)***	***Volume baseline (mm^3)***	***MRT (days)***	***Volume follow-up (mm^3)***	***CCA left proximal (mm)***	***CCA left distal (mm)***	***CCA right (mm)***
1	3400.00	114.00	76.00	26	0	158.00	9.42	29	12.26	2.90	2.60	2.70
2	3400.00	135.00	43.00	27	0	180.00	10.99	32	46.16	3.50	2.80	2.50
3	3900.00	197.00	70.00	27	0	250.00	6.59	31	10.10	3.50	3.20	3.30
4	4200.00	140.00	45.00	28	0	208.00	9.42	30	24.00	3.00	2.60	2.10
5	3660.00	109.00	53.00	18	0	192.00	8.24	25	8.00	2.80	2.80	2.80
6	3200.00	115.00	58.00	24	1	217.00	3.53	46	25.16	2.70	2.80	3.10
Mean ± SD	**3626.67 ± 371.30**	**135.00 ± 32.82**	**57.50 ± 13.31**	**25.00 ± 3.69**	**0.17 ± 0.41**	**200.83 ± 31.87**	**8.03 ± 2.64**	**32.17 ± 7.20**	**20.95 ± 14.31***	**3.07 ± 0.35**	**2.80 ± 0.22**	**2.75 ± 0.43**

(F). Surgical characteristics and morphometric measurements of elastase aneurysms with ASA treatment**.** Values are expressed as means ± SD. * $p_{Vol(BL-FU)} < 0.05$.

n	***Weight (g)***	***Ischemia (min)***	***Suture time (min)***	***Stitches (#)***	***Re-stitches (#)***	***Surgery (min)***	***Volume baseline (mm^3)***	***MRT (days)***	***Volume follow-up (mm^3)***	***CCA left proximal (mm)***	***CCA left distal (mm)***	***CCA right (mm)***
1	3700.00	158.00	100.00	24	0	240.00	18.84	27	26.70	3.30	3.20	2.00
2	4200.00	81.00	26.00	22	2	168.00	7.06	23	32.36	3.00	2.70	2.50
3	3600.00	99.00	45.00	20	0	182.00	7.06	33	13.84	2.90	2.80	2.40
4	3750.00	114.00	58.00	23	1	179.00	9.42	45	11.65	3.30	3.40	3.00
5	3300.00	109.00	57.00	21	0	190.00	8.24	44	10.17	2.10	2.00	2.00
6	3700.00	80.00	46.00	21	0	158.00	8.38	40	21.98	-	-	-
Mean ± SD	**3625.00 ± 357.42**	**106.83 ± 28.72**	**55.33 ± 24.74**	**21.83 ± 1.47**	**0.50 ± 0.84**	**186.17 ± 28.65**	**9.91 ± 4.48**	**35.33 ± 9.14**	**19.45 ± 8.99***	**2.92 ± 0.49**	**2.82 ± 0.54**	**2.32 ± 0.50**