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Comprehensive Preclinical Evaluation of the Valvuloplasty Embossing Balloon: Thrombo-Resistance and Safety in a Porcine Model

Minocha Dr. Pramod Kumar[1], Kothwala Dr. Deveshkumar[2], Shah Khusboo[3], Pandya Kamna[4], Kadam Aniket[5], Dave Arpit[6], Shah Hrishikesh[7]

Meril Medical Innovations Private Limited, Bilakhia House, Survey no.879, Muktanand Marg, Chala, Vapi, Dist-Valsad, Gujarat, 396191, India.

Corresponding Author: Shah Khusboo

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ABSTRACT

The study investigated the thrombo-resistance of the Valvuloplasty Embossing Balloon in a clinically relevant Porcine model. Male swine were chosen for preclinical testing, following standard fasting protocols to mitigate potential confounders. The balloon was deployed in the descending thoracic aorta, aided by a guide wire, to circumvent complications associated with valvular space inflation. Despite the inability to reach valvular space, the study aimed to evaluate the device's thrombogenicity. Angiographic imaging revealed the balloon's excellent performance, maintaining consistent retrograde contrast flow during inflation and deflation without thrombus formation. Post-procedural examination showed no signs of thrombus formation or coagulation on the balloon's surface or at the target sites. Additionally, necropsy findings confirmed the absence of thrombus formation or injury to the aorta's luminal interface, indicating the device's thrombo-resistance. Hematology and clinical chemistry parameters remained stable before and after the procedure, affirming the absence of systemic effects. The mean thrombus formation score of 0 indicated no significant thrombosis, highlighting the device's thrombo-resistant nature. Notably, no adverse events or safety concerns were reported. In conclusion, the Valvuloplasty Embossing Balloon demonstrated thrombo-resistance in the porcine model, supporting its safety and efficacy for clinical use in valvuloplasty procedures.

Keywords: Thrombo-resistance, Valvuloplasty Embossing Balloon, Porcine Model, thrombogenicity, angiographic imaging, necropsy findings, thrombus formation.

INTRODUCTION

The valvuloplasty embossing balloon is an advanced medical device integrating a balloon dilatation catheter with a nitinol embossing element (1). Designed for over-the-wire deployment, this system features a usable length of 115 cm to 145 cm and a balloon length of 25 mm to 55mm, with various balloon diameters available. It is compatible with a 0.035" guidewire, necessitating a minimum length of 245 cm to 275 cm. The purpose of this study is to evaluate the thrombo-resistance of the valvuloplasty embossing balloon using a clinically relevant porcine model (2).

Valvuloplasty is a minimally invasive procedure that increases blood flow and helps the heart work more efficiently, potentially postponing or avoiding valve replacement. Compared to open-heart surgery, it offers benefits such as faster recovery, less scarring, lower risk of

complications, and reduced pain. However, it is not a substitute for surgery when indicated by your healthcare provider (3). This single-arm study investigates both regional and downstream thrombo-resistance, alongside local safety and performance metrics. Key parameters evaluated include trackability, handling, visualization, haemostasis of the delivery system, qualitative and quantitative aspects of the deployment profile, and ease of withdrawal (4). Additionally, the study examines angiographic and necropsy data approximately 30 days post-valvuloplasty treatment to assess potential complications such as dissection, perforation, embossing effects, embolization, thrombus formation, aneurysm formation, filling defects, adverse cardiac events, and injury scores. Histopathological analysis of the target vessel is conducted to evaluate inflammatory changes and vascular injury (5).

Swine was chosen for this study due to the similarities between their circulatory system and that of adult humans (6). Key parameters such as size, cardiac output, heart rate, and blood pressure in swine are comparable to those in the human population targeted for this device (7). This physiological resemblance makes swine an ideal model for testing cardiovascular devices, ensuring the relevance and applicability of the study results to human clinical scenarios. (8)

Smaller species, which differ significantly in circulatory system parameters from adult humans, are less suitable for in vivo studies involving such devices (9) The use of swine in cardiovascular device testing is widely accepted, reflecting their physiological similarities to humans and thereby enhancing the pertinence of the study findings to the intended patient population.(9,10)

MATERIALS & METHOD

This study is designed to assess the acute thrombogenicity of the test items in a clinically relevant swine model as a standalone evaluation. Given that the observed score in the study was zero (0), there was no need for a side-by-side comparison with a control group (dual arm). The valvuloplasty embossing balloon is designed for the pre-dilatation of native stenotic cardiac valves before the implantation of trans-catheter heart valves. Its use extends to patients with aortic stenosis, including those with calcium deposits, bicuspid anatomy, pulmonary stenosis, and an extended indication for co-arctation of the aorta. The device is intended for limited contact duration, specifically less than 24 hours.

In terms of physical specifications, the balloon exhibits a thickness greater than 1.0 mm and is available in sizes of 18 x 40 mm, 20 x 40 mm, and 23 x 40 mm. The delivery device size is standardized at 12F to 16F in between. Overall, these characteristics define the intended use and physical attributes of the valvuloplasty embossing balloon.

Details of medication used in the study have been mentioned in table no.1.

Table 1: Details of Medication Used in the Study

Drug Name	Manufactured By	Batch / Lot No.	Expiry Date
Ketamine	NEON Laboratories Limited	1485002	Dec 2023
Xylazine	Indian Immulogicals Ltd	FHK 2004	Apr 2025
Isoflurane	Troikaa Pharmaceuticals Ltd	121256	Nov 2026
Thiopentone Sodium	Neon Lab	184526	Mar 2024
Heparin	Fusion Healthcare	L1182218B	Oct 2024
Tramadol	NEON	KP949131	Nov 2022
Atropine	Doctors Life Sciences (India) LTD	BY22001	Dec 2023

Device Design

The Valvuloplasty Embossing Balloon is designed as follows:

1. Components: It comprises a dilatation catheter equipped with a nitinol embossing element.

2. Design: The valvuloplasty balloon operates on an over-the-wire system, meaning it is inserted over a guidewire.
3. Specifications:

- Usable length: 115 cm to 145 cm
- Balloon length: 25 mm to 55 mm
- Balloon diameters: Available in various sizes to accommodate different procedural needs.
- Compatibility: Designed to be compatible with a 0.020" to 0.050" guidewire with a minimum length of 245 cm to 275 cm. The size matrix of a valvuloplasty balloon has been tabulated in table 2 for better understanding regarding the specifications.

This device is likely intended for use in valvuloplasty procedures, where it can be used to dilate and reshape heart valves. The inclusion of a nitinol embossing element suggests that it may provide enhanced control and precision during the procedure. The variety of balloon diameters allows for customization based on the specific anatomy and requirements of the patient. The complete assembly of a Valvuloplasty Embossing Balloon and the Delivery System has been depicted in the figure 1 with its technical specifications in table 3.

Table 2: Size Matrix

Valvuloplasty Balloon Diameter (mm)	Balloon Length (mm)
14	40
16	
18	
20	
23	
25	

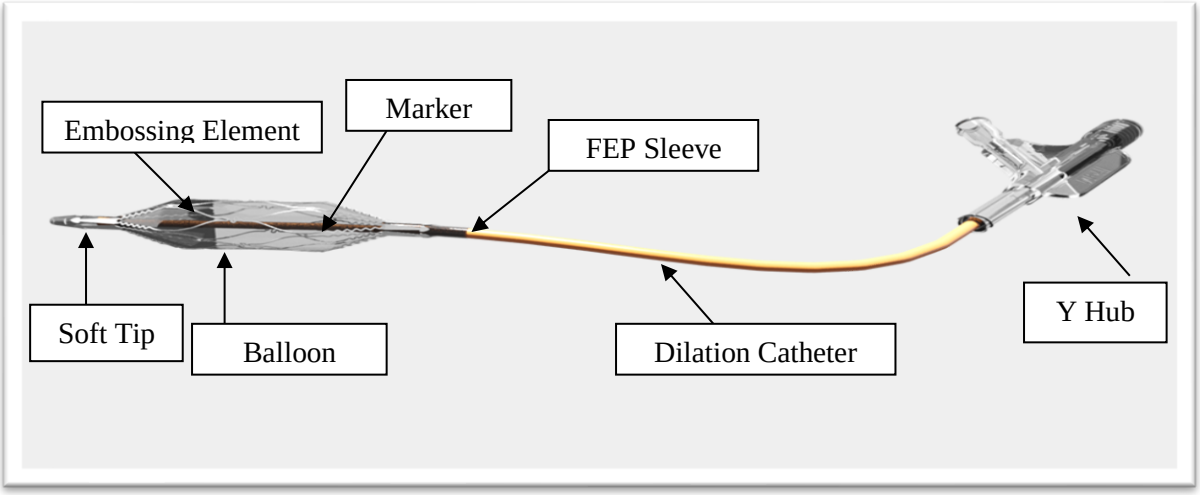


Figure 1: The Complete Assembly of a Valvuloplasty Embossing Balloon and the Delivery System

Table 3: Technical Specifications of the Product

Embossing Element Material	:	Nitinol
Embossing element design	:	Anchor head and zig-zag strut at proximal and distal end with serpentine link at intermediate region
Delivery system		
Delivery system	:	Over the wire (OTW) 0.020" to 0.050"
Balloon catheter	:	Semi-compliant
Balloon material	:	Nylon
Balloon diameter (mm)	:	14.0, 16.0, 18.0, 20.0, 23.0, 25.0
Usable balloon length (mm)	:	25 mm to 55 mm

Soft tip	:	Pebax
Nominal pressure	:	3 ATM
Usable catheter length	:	130 cms
Sheath compatibility	:	12F To 16F
Guidewire compatibility	:	0.035" (0.889mm)

Experimental Procedure

Animals were maintained on anticoagulant therapy, receiving aspirin at a dosage of 300 mg per animal and clopidogrel at a dosage of 75 mg per animal for duration of 3 days preceding the procedural day, which preceded the implantation of the test item. This treatment regimen was sustained from day 0 (the day of implantation) until the conclusion of the study, with a subsequent adjustment to a lower dosage of aspirin 150mg.

Diet and Water

The feed used in the study, which is available for purchase and tailored to the specific species, was determined to be adequate by the study veterinarian, including any necessary supplemental hay. The hay and other non-formulated dietary components were found to be acceptable for consumption. The water source was borewell water that had been purified using a reverse osmosis (RO) water plant located on the premises, and it was provided to the animals at all times.

Animal Preparation

The animals underwent a fasting period and were deprived of water for at least 12 hours prior to the procedure. They were then weighed (weight tabulated in table no.4), anesthetized, instrumented, and monitored. Atropine was administered as preanesthetic at a dose rate of 0.05mg/kg (IM), followed by ketamine at 15 mg/kg (IM), xylazine at 2.5 mg/kg (IM), and propofol at 0.5mg/kg (IV bolus if necessary), with subsequent inhalation anesthesia at a concentration of 1-3% administered through a facemask. Clipping of hair from the chest and thigh areas facilitated the aortic root approach and application of ECG leads, respectively.

Table 4: Body Weight of Animal

Animal number	Sex	Treatment (Day 0)	Follow up day (Day 30)
P1	Male	84.2 kg	85.8 kg
P2	Male	92.4 kg	93.8 kg

Animal Trial Procedure

The right and left groin areas were prepared by shaving. A percutaneous approach was employed using the Seldinger method, where a vessel was punctured with a 16G to 20G needle to introduce an 8 to 12 French sheath into the femoral artery. Activated clotting time (ACT) measurements were taken before and after heparinization to maintain ACT values between 250 to 550 seconds. An initial heparin bolus dose of 100 IU/kg was administered intra-arterially.

A 0.020-inch to 0.050-inch diameter, 245 cm to 275 cm length guidewire was utilized to guide the Python sheath, through which the test item was advanced into the descending aorta. Radiographic cine images of the balloon were captured to confirm its placement. The inflation device was used to inflate and deflate the balloon, which was maintained inflated for 30 seconds before removal for thrombogenicity assessment.

Following the simulated procedure, the mammoth catheter was withdrawn, and thrombus evaluation was conducted, with photographic documentation for thrombogenicity assessment. The same procedure was then repeated in the second animal (P2).

After the procedures, the animals were allowed to recover and returned to their respective cages for observation. P2 was euthanized immediately after the test item withdrawal on day 0, while P1 was

euthanized on day 30. Post-euthanasia, a comprehensive necropsy was performed, involving a detailed examination of the heart, external body surfaces, orifices, thoracic and abdominal cavities, and their contents.

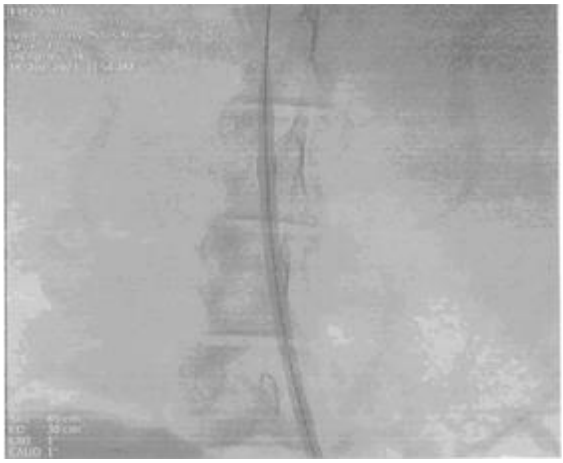
Radiographic Thrombogenicity Assessment

Radiographic confirmation and assessment of thrombogenicity were conducted for each device in both experimental animals. Radiographic images were captured, Specified in radiographic images figure 2(P1) and 3(P2).

- Trackability Assessing the handling, visualization, and hemostasis of the test item delivery system.
- Qualitative Evaluation of test item deployment profile evaluating the inflation, deflation, and slippage characteristics of the test item deployment.
- Quantitative evaluation of test item deployment profile providing quantitative analysis of the test item deployment.
- Ease of withdrawal

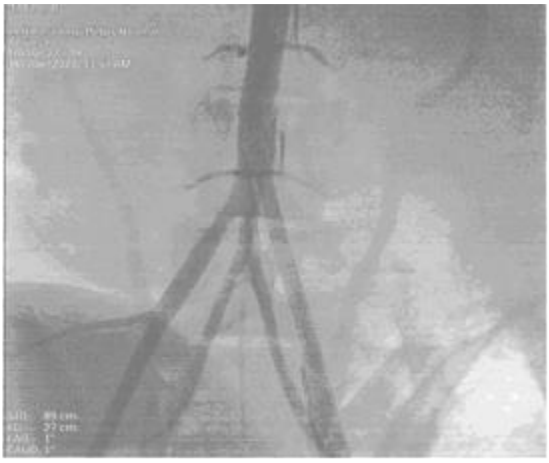
Radiographic Images

Animal P1



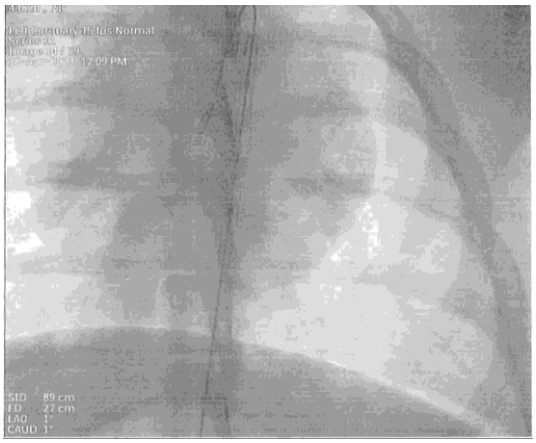
Animal P1 Long Sheath was Inserted in Abdominal Aorta

Animal P1 Ascending Aorta Measurement and Baseline Angion of Descending and Arch of Aorta

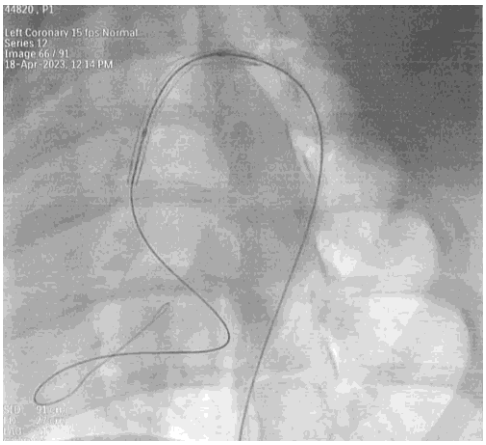


Animal P1, Long Sheath Angiogram

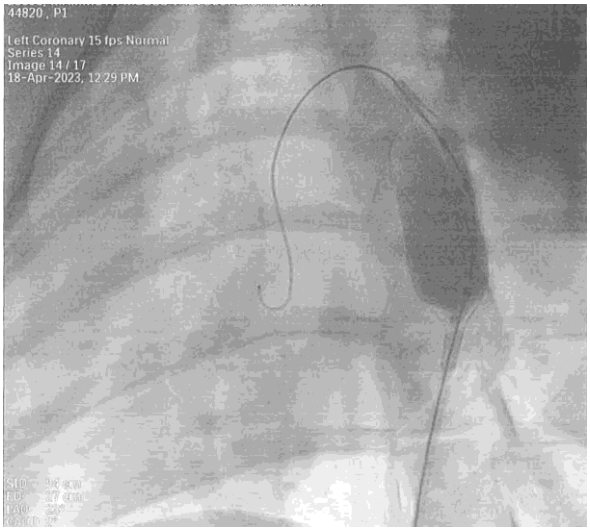
Animal P1, Valvuloplasty Embossing Balloon was Passing on the Guide Wire 0.035x260cm Illio Femoral Artery, Abdominal Aorta, and Thorax



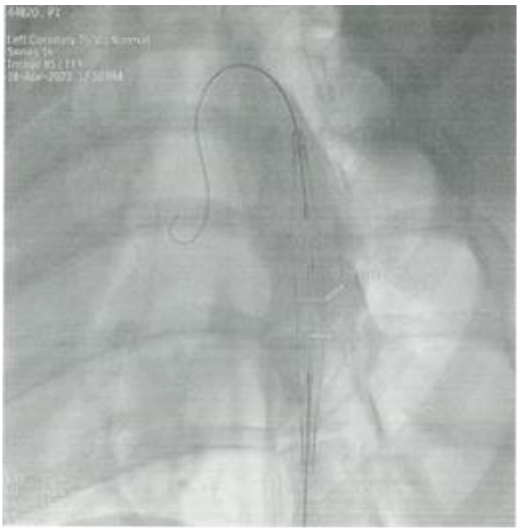
Animal P1, Valvuloplasty Embossing Balloon was Passing on The Guide Wire 0.035x260cm at the Descending Aorta.



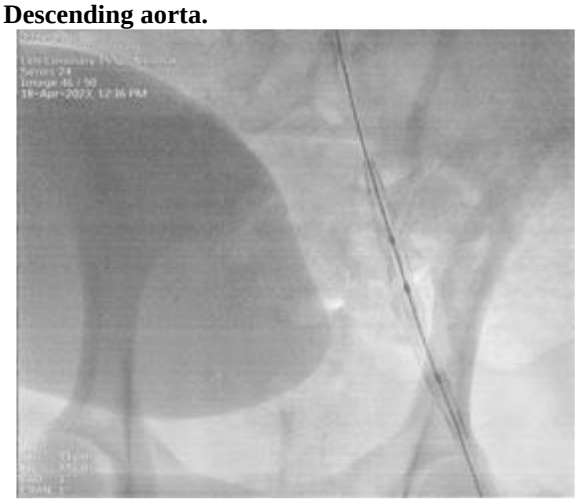
Animal P1, Valvuloplasty Embossing Balloon was Crossing on the Guide Wire 0.035x260cm at the Arch of Aorta.



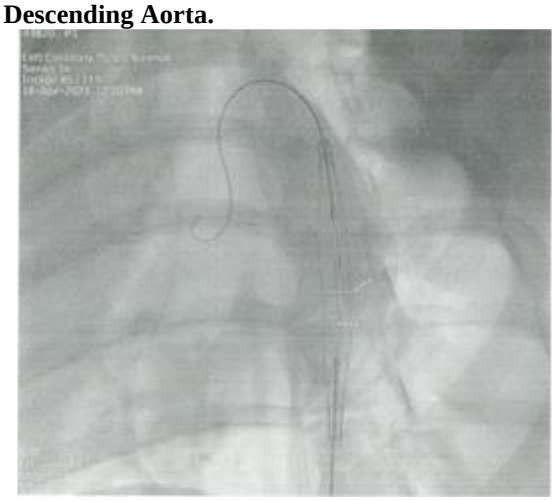
Animal P1, Valvuloplasty Embossing Balloon was Dilation with Inflation device Support in Descending aorta.



Animal P1, Valvuloplasty Embossing Balloon was Deflation with Inflation Device Support in Descending Aorta.



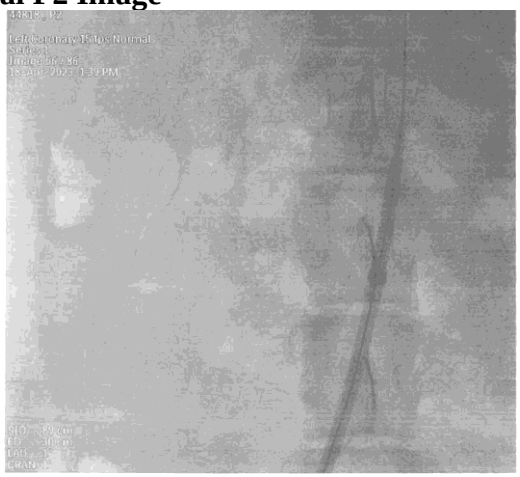
Animal P1 Valvuloplasty Embossing Balloon was Withdrawing into Thorax Aorta and Abdominal Aorta



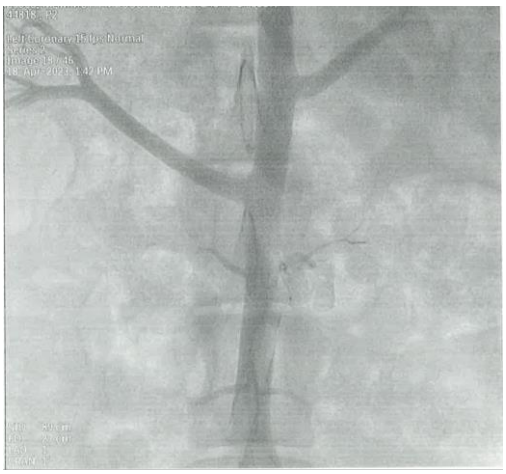
Animal PI, Valvuloplasty Embossing Deflated Balloon Length and Width Measurement.

Figure 2: Radiographic Images Animal P1

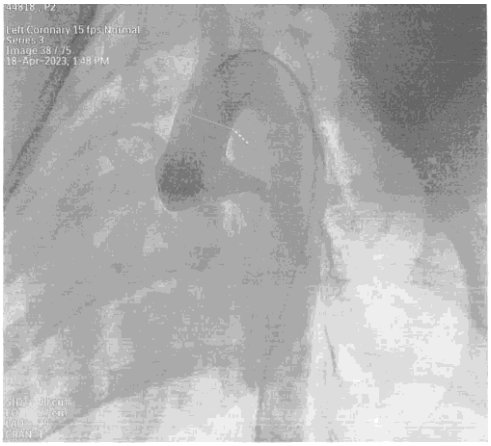
Animal P2 Image



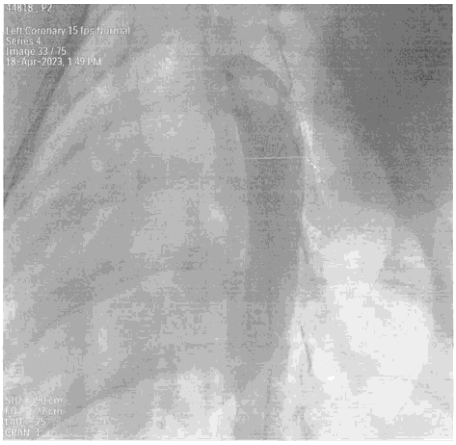
Animal P2, Long Sheath was Inserted in Abdominal Aorta.



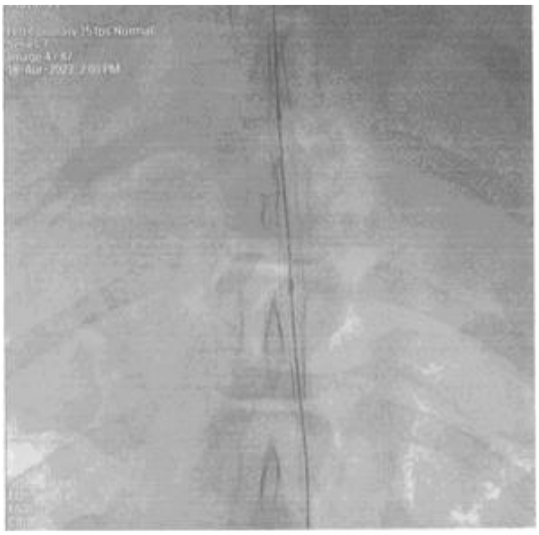
Animal P2, Long Sheath Angiogram.



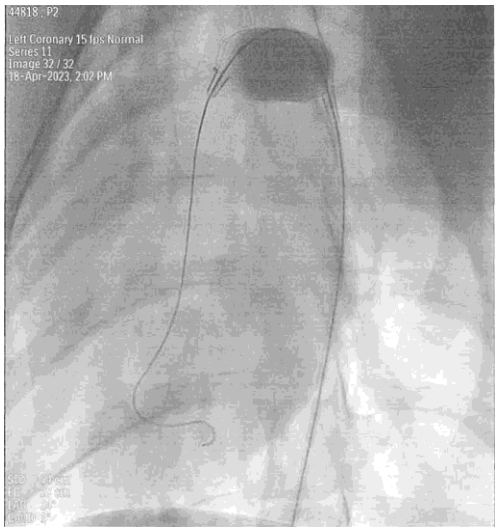
Animal P2, Ascending Aorta Measurement and Baseline Angio of Ascending and Arch of Aorta.



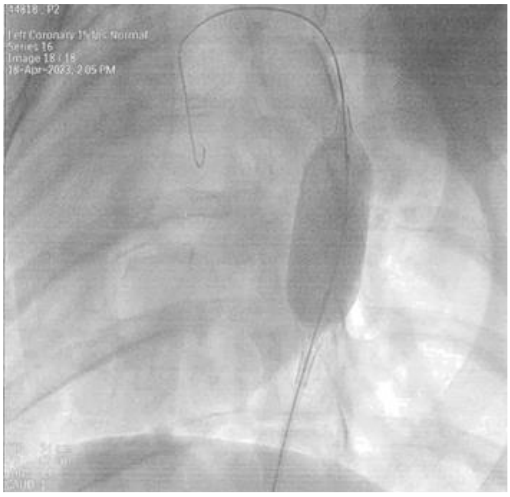
Animal P2, Descending Aorta Measurement and Baseline Angio of Descending Aorta



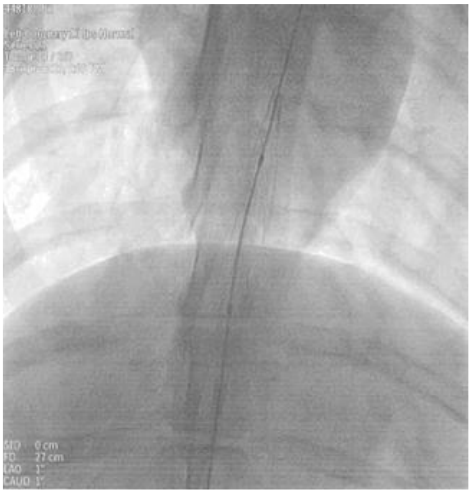
Animal P2, Valvuloplasty Embossing Balloon was Crossing on the Guide Wire 0.035x260cm at the Iliofemoral Artery, Abdominal Aorta and Thorax



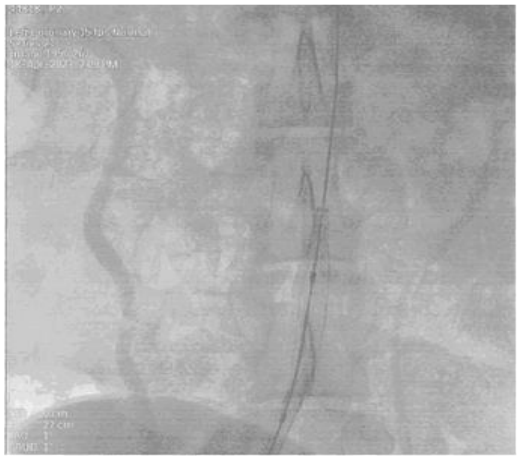
Animal P2, Valvuloplasty Embossing Balloon was Dilation with Inflation Device Support in Arch of Aorta.



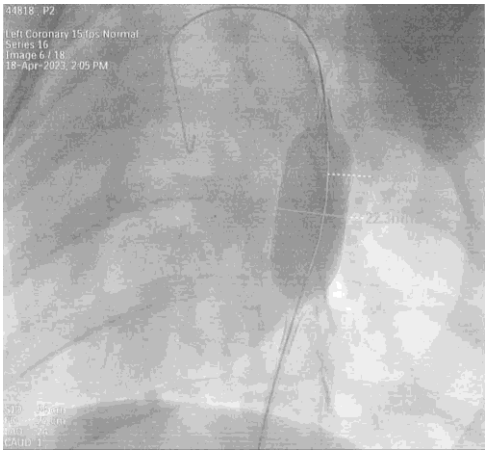
Animal P2, Valvuloplasty Embossing Balloon was Dilation with Inflation Device Support in Descending Aorta



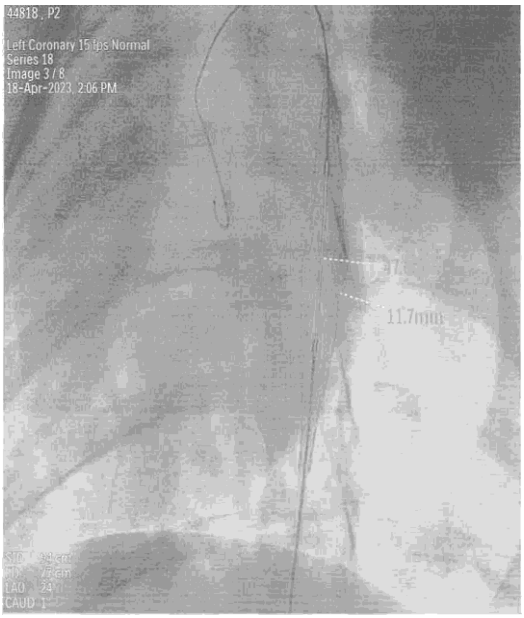
Animal P2, Valvuloplasty Embossing Balloon was Withdrawing into Descending Aorta



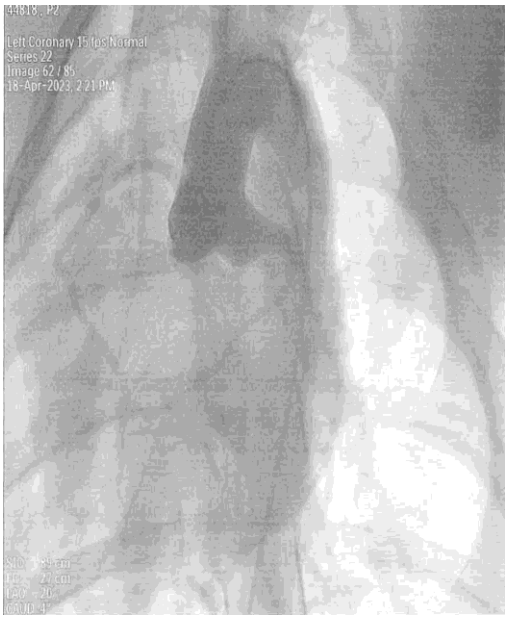
Animal P2, Valvuloplasty Embossing Balloon was Withdrawing into Illio Femoral Artery, Abdominal Aorta and Thorax Aorta.



Animal P2, Valvuloplasty Embossing Inflated Balloon Length and Width Measurement.



Animal P2, Valvuloplasty Embossing Deflated Balloon Length and Width Measurement.



Animal P2, After Withdrawal the Valvuloplasty Embossing Balloon Aortic Root Check Angio was Done (No Dissection, Injury, and Thrombus Were Seen).

Figure 3: Radiographic Images Animal P2

Table 5: Mortality Table

Animal Number		**44820 & 44818**	**P1 & P2**
Sex		**Male**	**Male**
Day of Observations	**Mortality**	**Incidences**	**Incidences**
Acclimatization Phase (Day 1-3)	Mortality	0/2	-
Treatment / Experiment Phase (Day 30)	Mortality	-	0/2

Table 6: Clinical Signs

Days of Observation					
Animal Id	**Acclimatization**			**Animal Number**	**Treatment**
	1	**2**	**3**		
44820	1	1	1	P1	1
44818	1	1	1	P2	1

Clinical Pathology

At pre-balloon application and on the terminal day, standard hematology and clinical chemistry parameters were analyzed. Adequate blood volumes were collected from each animal at both pre- and post-surgical procedure time points. To ensure accurate results, appropriate anticoagulants were used to mix the blood for haematology (K2 EDTA), clinical chemistry (lithium heparin), and electrolyte (sodium citrate 3.2% w/v) parameters. Clinical Signs was mentioned in table no.6.

Table 7: The Following Haematological Parameters were Analyzed

• Haemoglobin concentration	• White blood cell (WBC) count
• Red blood cell (RBC) count	• Platelet count
• Haematocrit	• WBC differential Reticulocytes
• Reticulocytes	

Table 8: Individual Data of Hematology Parameters (Baseline)

Animal No.	P1/Base	P2/Base	Mean	SD	Lower Limit	Upper Limit
WBC (10^9/L)	13.44	16.08	14.76	1.87	9.37	19.89
RBC (10^{12}/L)	4.79	5.07	4.93	0.20	4.65	7.07
HGB (g/L)	115	97	106.00	12.73	84.21	123.56
HCT (L/L)	0.338	0.296	0.32	0.03	0.27	0.39
PLT (10^9/L)	277	252	264.50	17.68	317.21	559.21
NEUT (%)	59.4	63.5	61.45	2.90	20.55	68.25
LYM (%)	36.5	33.5	35.00	2.12	22.73	60.87
MONO (%)	2.0	2.3	2.15	0.21	1.51	14.82
EOS (%)	0.6	0.1	0.35	0.35	0.18	0.62
LUC (%)	1.4	0.4	0.90	0.71	0.49	4.16
BASO (%)	0.2	0.1	0.15	0.07	0.18	0.62
Retic (%)	1 .44	0.37	0.91	0.76	0.5	1.5
PT (Seconds)	16.9	17.8	17.35	0.64	12.3	15.1
APTT (Seconds)	33.8	23.7	28.75	7.14	21	35

Abbreviation: WBC- White blood cells, RBC- Red blood cells, HGB- Haemoglobin, HCT- Haematocrit, PLT- Platelet, Neut- Neutrophil, LYM- Lymphocyte, MONO- Monocyte, BASO- Basophil, EOS-Eosinophil and Retic- Reticulocytes

Table 9: Individual Data of Haematology Parameters (Terminal)

Animal No.	**P1/Terminal (0 Day)**	**P2/Terminal (30 Day)**
WBC (10^9/L)	26.17	11.84
RBC (10^{12}/L)	6.53	7.94
HGB (g/L)	474	188
HCT (L/L)	0.460	0.578
PLT (10^9/L)	177	178
NEUT (%)	34.3	37.2
LYM (%)	60.5	53.9
MONO (%)	2.3	4.1
EOS (%)	1.1	2.9
LUC (%)	1.8	1.3
BASO (%)	0.0	0.6
Retic (%)	0.31	2.36
PT (Seconds)	17.2	20.4
APTT (Seconds)	27.1	25.8

Abbreviation: WBC- White blood cells, RBC- Red blood cells, HGB- Haemoglobin, HCT-Haematocrit, PLT- Platelet, Neut- Neutrophil, LYM-Lymphocyte, MONO-Monocyte, BASO-Basophil, EOS- Eosinophil and Retic- Reticulocytes

Table 10: Following Clinical Chemistry Parameters were Analyzed

• Albumin	• Gamma-glutamyl transferase (GGT)
• Alkaline phosphate (ALP)	• Glucose
• Alanine aminotransferase (ALT)	• Inorganic phosphorus
• Aspartate aminotransferase (AST)	• Potassium
• Calcium	• Sodium
• Chloride	• Total bilirubin
• Cholesterol	• Total protein
• Creatinine	• Triglycerides

Table 11: Individual and Summary Data of Clinical Chemistry (Baseline)

Animal No.	P1/Baseline	P2/Base1ine	Mean	SD	Lower Limit	Upper Limit
ALB (g/L)	44.8	21.8	33.30	16.26	35	50
Alp (U/L)	44	95	69.50	36.06	16.90	120.20
ALT (U/L)	22	12	17.00	7.07	7	56
AST (U/L)	29	18	23.50	7.78	27.20	89.90
Ca (mmol/L)	2.82	2.08	2.45	0.52	2.12	2.62
T. Choi (mmol/L)	1.41	1.17	1.29	0.17	0.5	1.61
Creat (pmol[L)	152	81	116.50	50.20	75.10	228.70
GGT (U/L)	53	34	43.50	13.44	20.80	157.80
Glu (mmol/L)	5.63	5.21	5.42	0.30	5.1	12.55
P1 (mmol/L)	1.83	1.92	1.88	0.06	1.5	3.2
T. Bil (pmol/L)	3.33	1.07	2.20	1.60	1.1	13.9
T. Pro (g/L)	83.9	57.8	70.85	18.46	51.20	90.63
Trig (mmol/L)	0.14	0.08	0.11	0.04	0	1.7
BUN (mm01/L)	2.54	2.03	2.29	0.36	1.61	5.57
Sodium (mmol/L)	138.3	142.3	140.30	2.83	136	145
Potassium(mmol/L)	3.95	3.63	3.79	0.23	3.6	5.2
Chloride (mmol/L)	96.0	99.7	97.85	2.62	96	106

Abbreviations: ALB- Albumin, ALP- Alkaline phosphates, ALT- Alanine amino transferase, AST- Aspartate amino transferase, Ca- Calcium, T.Chol- Total cholesterol, Creat- Creatinine, GGT- Gama glutamyl transferase, GLU- Glucose, P1- Inorganic phosphorus, T.BiI- Total bilirubin, T. Pro- Total protein, Trig- triglycerides, BUN- Blood urea nitrogen, Na- Sodium, K- potassium and Cl- Chloride.
Note: Convert BUN to urea in mmol/L by using following formula:
BUN [mmol/L] x 1 = Urea [mmol/L]

Table 12: Individual and Summary Data of Clinical Chemistry (Terminal)

Animal No.	P1/Termina1 (30 Day)	P2/Terminal (0 Day)
ALB (g/L)	21.4	48.0
Alp (U/L)	240	50
ALT (U/L)	23	30
AST (U/L)	21	26
Ca (mmol/L)	2.63	3.06
T. Choi (mm01/L)	2.12	1.77
Creat (pmol/L)	66	148
GGT (U/L)	52	63
Glu (mmol/L)	2.64	5.86
P1 (mmol/L)	1.68	1.95
T. Bil (pmoVL)	2.39	2.45
T. Pro (g/L)	53.7	103.4
Trig (mmol/L)	0.54	0.24
BUN (mmoVL)	3.96	4.36
Sodium (mmol/L)	141.9	148.4
Potassium(mmol/L)	3.16	5.02
Chloride (mmol/L)	106.7	116.6

Abbreviations: ALB- Albumin, ALP- Alkaline phosphates, ALT- Alanine amino transferase, AST- Aspartate amino transferase, Ca- Calcium, T. Chol- Total cholesterol, Creat- Creatinine, GGT- Gama glutamyl transferase, GLU- Glucose, P1- Inorganic phosphorus, T.Bil- Total bilirubin, T. Pro- Total protein, Trig- triglycerides, BUN- Blood urea nitrogen, Na- Sodium, K- potassium and CI- Chloride.
Note: Convert BUN to Urea in mmol/L by using following formula:
BUN [mmol/L] x 1 = Urea [mmol/L].

Pathology

Euthanasia

The animals were euthanized using an intravenous injection of pentobarbital sodium or thiopental sodium at a dose of 100 mg/Kg. Specifically, P2 was euthanized immediately after the test item was removed on day 0, while P1 was euthanized on day 30. If necessary, additional doses were administered to ensure euthanasia. The death of the animals was confirmed through the observation or auscultation of the heart and lungs, a systolic ECG, and zero oxygen saturation.

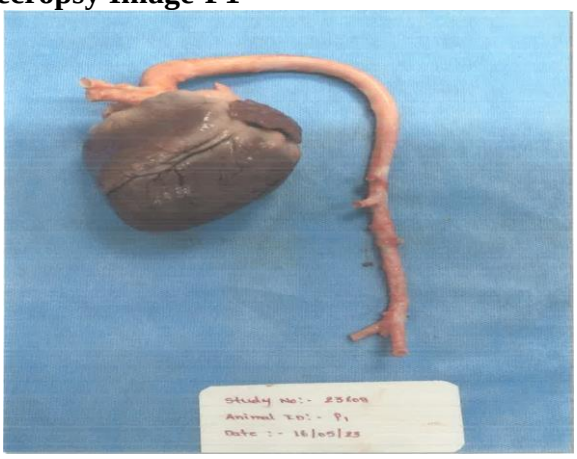
Necropsy

The data is about a study evaluating the thrombo-resistance of the Valvuloplasty Embossing Balloon in a porcine model. The balloon was deployed in the descending thoracic aorta with a guide wire and the dwelling time was 30 seconds, which is a clinically relevant time. The study found that the test item showed prominent visualization under angiographic imaging with good retrograde contrast flow throughout the balloon inflation and deflation time in the target site with no evidence of thrombus formation. The test item and the target sites showed no sign of thrombus or coagulation due to the surface material of the test item throughout the balloon inflation and deflation time in the target sites and after explanting the test item from target sites. The post procedural necropsy of the animals showed no thrombus formation or injury to the luminal interface in the aorta. Thrombus formation or thromboembolism was not observed in any examined tissue. The mean thrombus formation score was 0 (No significant thrombosis) for the test item, (Thrombus formation score has been specified in the table no. 13). The test item was thromboresistant in both the regional and downstream areas. No adverse events or safety concerns were found in this study. The study concluded that the Valvuloplasty Embossing Balloon does not have thrombogenicity activity in the test animals and can perform activities related to valvuloplasty with no device related coagulopathic local lesions.

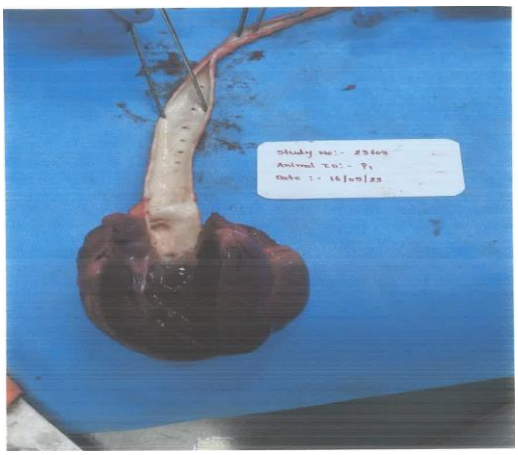
Table 13: Thrombus Formation Score

Thrombus formation score description	Score
No significant thrombosis (a very small clot is acceptable at insertion)	0
Minimal thrombosis, one location	1
Minimal thrombosis, multiple location	2
Significant thrombosis, >1/4 to 61/2 the surface of the implant, vessel patent	3
Significant thrombosis, >1/2 the surface of the implant, vessel patent	4
Vessel completely occluded	5

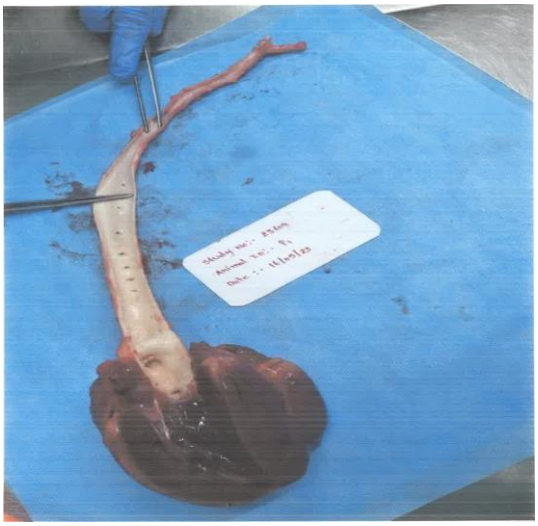
Necropsy Image-P1



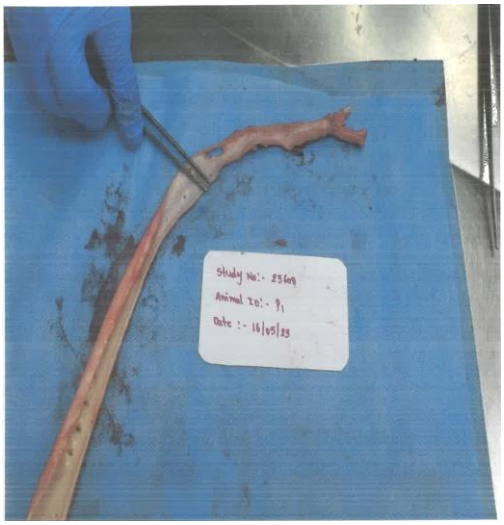
Heart with Whole Aorta Collection



Heart with LV, Ascending and Arch of Aorta Internal Wall of the Artery (No Injury, Dissection and Thrombus were Seen)



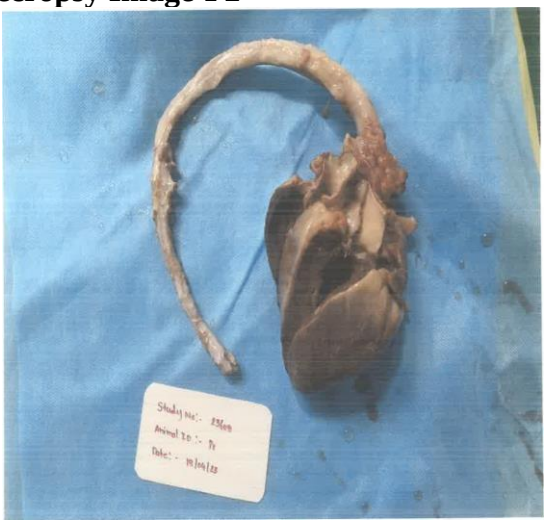
Heart with LV, Ascending Aorta, Descending Aorta and Arch of Aorta Internal Wall of the Artery (No Injury, Dissection and Thrombus were Seen)



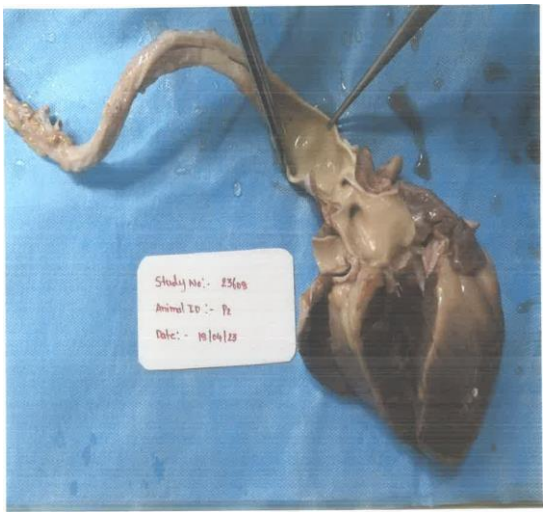
Descending Aorta, Abdominal Aorta and Ilio Femoral Artery Internal Wall of the Artery (No Injury, Dissection and Thrombus were Seen).

Figure 4: Necropsy Image-P1

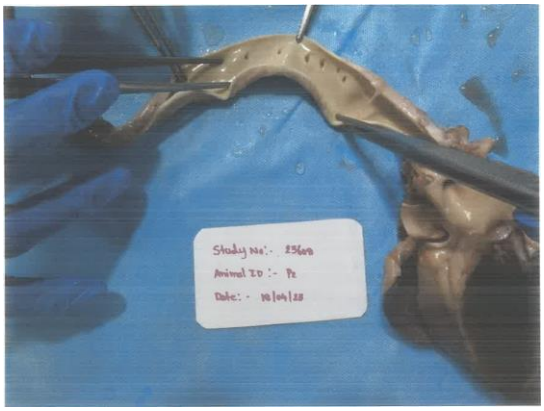
Necropsy Image-P2



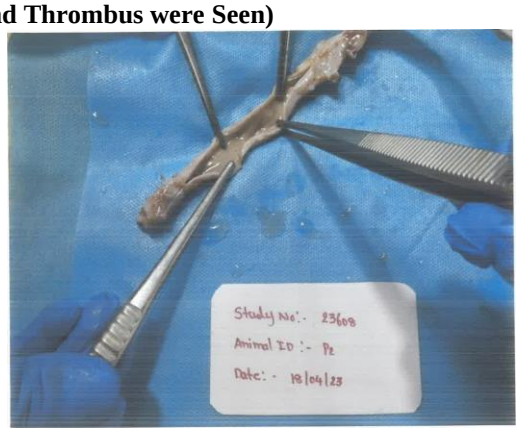
Heart with Whole Aorta Collection



Heart rate with LV, Ascending and Arch of Aorta Internal Wall of the Artery (No Injury, Dissection and Thrombus were Seen)



Descending Aorta and Thorax Aorta Internal Wall of the Artery (No Injury, Dissection and Thrombus were Seen)



Abdominal Aorta, and Ilio Femoral Artery Internal Wall of the Artery (No Injury, Dissection and Thrombus were Seen)

Figure 5: Necropsy Image-P2

Histopathology

The histopathology of the target vessel was performed for inflammatory changes and vascular injury at the terminal time point, which was day 30 on the descending aorta, including 2-3cm above and below the inflated region together with the inflated region. The histological analysis was conducted to evaluate the presence of inflammatory cells, vascular injury, smooth muscle cell loss, fibrin deposition, and endothelial loss in any part of the descending aorta (proximal, mid, and distal) in P1 and P2 animals.

Histological Analysis

The histological analysis of the descending aorta was conducted to evaluate the presence of inflammatory cells, vascular injury, smooth muscle cell loss, fibrin deposition, and endothelial loss in any part of the descending aorta (proximal, mid, and distal) in P1 and P2 animals. The search results provide insights into the histopathology of aortic aneurysms, which are characterized by a degenerative process involving the extracellular matrix and loss of smooth muscle cells in the medial layer of the aortic wall. Inflammatory cell infiltration and (eP1) genetic changes can modulate vascular smooth muscle cell (vSMC) functions, leading to disturbances in processes such as changes in TGF-β signaling and regulatory RNA expression.

Additionally, vSMCs are paramount for providing structural and functional integrity of the aortic wall and ECM synthesis, and their plasticity allows them to adapt to environmental stimuli and mechanical stress

METHODOLOGY

On the scheduled sacrifice dates, P2 (on Day 0) and P1 (on Day 30) were euthanized using an overdose of Thiopentone sodium followed by exsanguinations. A pathologist examined all animals for external and internal gross pathological changes. The descending aorta was harvested from both animals and preserved in 10% neutral buffered formalin. Sections of the descending aorta (proximal, mid, and distal parts) were processed to achieve a section thickness of 3-5 microns. These tissue sections were stained with hematoxylin and eosin (H&E) and subsequently examined under a light microscope by the study pathologist to evaluate histopathological lesions.

RESULTS

Gross Pathology

External Findings:

No lesions of pathological significance were observed during the external examination of male animals (P1 and P2).

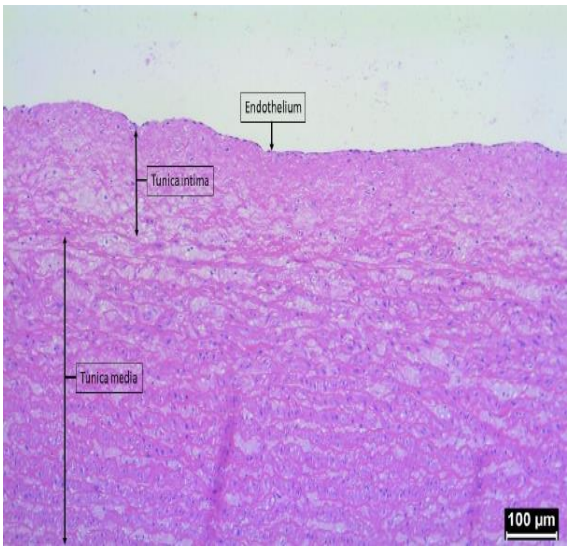
Internal Findings:

No pathological abnormalities were detected during the internal examination of male animals (P1 and P2).

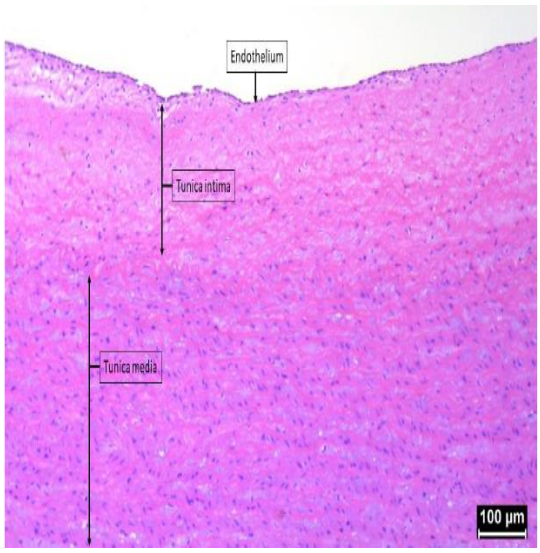
Animal no. P1- The microscopic examination of the descending aorta in animal no. P1 revealed no inflammatory cells, vascular injury, smooth muscle cell loss, fibrin deposition, or endothelial loss in any part of the descending aorta. The total histopathology score was 0, indicating a normal histopathological appearance.

Animal no. P2 - Microscopically, a few inflammatory cells were observed in the mid part of the descending aorta. This occurrence may be attributed to the embossing balloons exerting direct pressure on the endothelium, resulting in minimal inflammation. Such findings are commonly associated with this type of procedure and are not indicative of issues related to the test item. No vascular injury, smooth muscle cell loss, fibrin deposition, or endothelial loss was detected in any part of the descending aorta. The total histopathology score was 0.3. Safe limit was acceptable limit should be mentioned, (The safer histopathology score component ranges from 0 to 5).

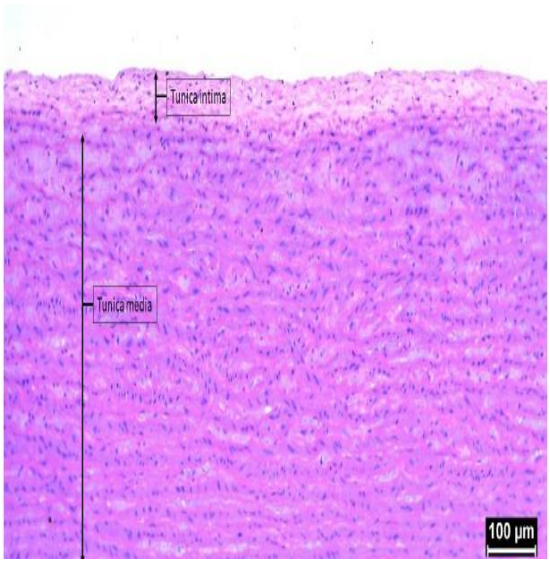
Hematoxylin and Eosin (H&E) Image of animal P1&P2:



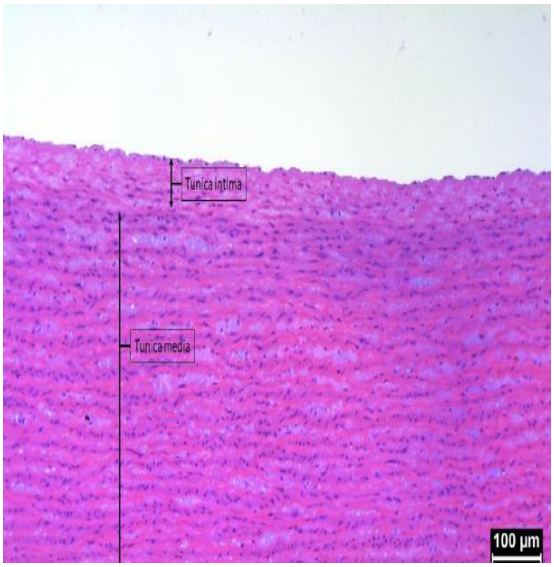
Animal No. P1 Aortal Proximal H&E 10X



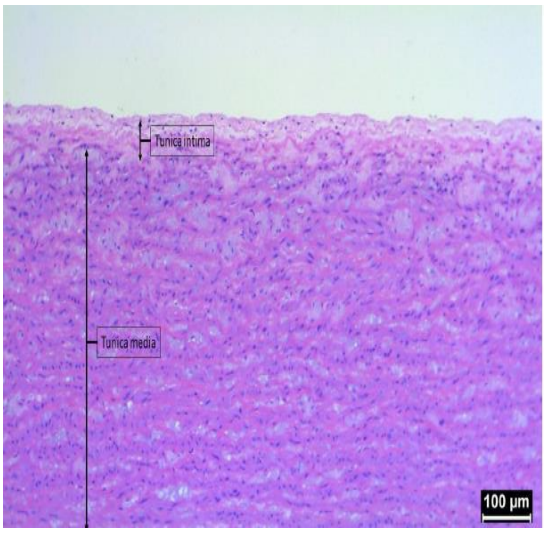
Animal No. P1 Aortal Mid H&E 10X



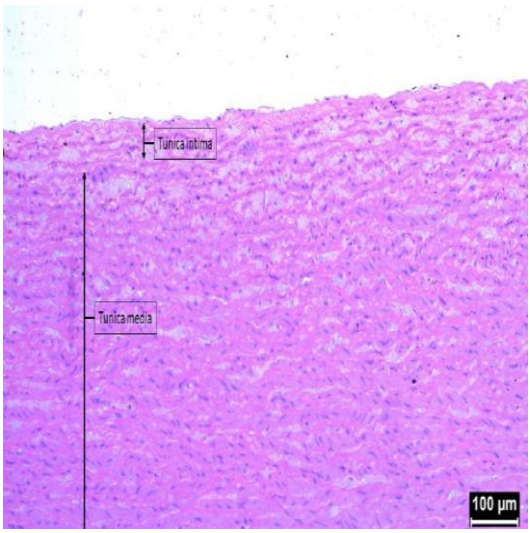
Animal No.P1 Aortal distal H&E 10X



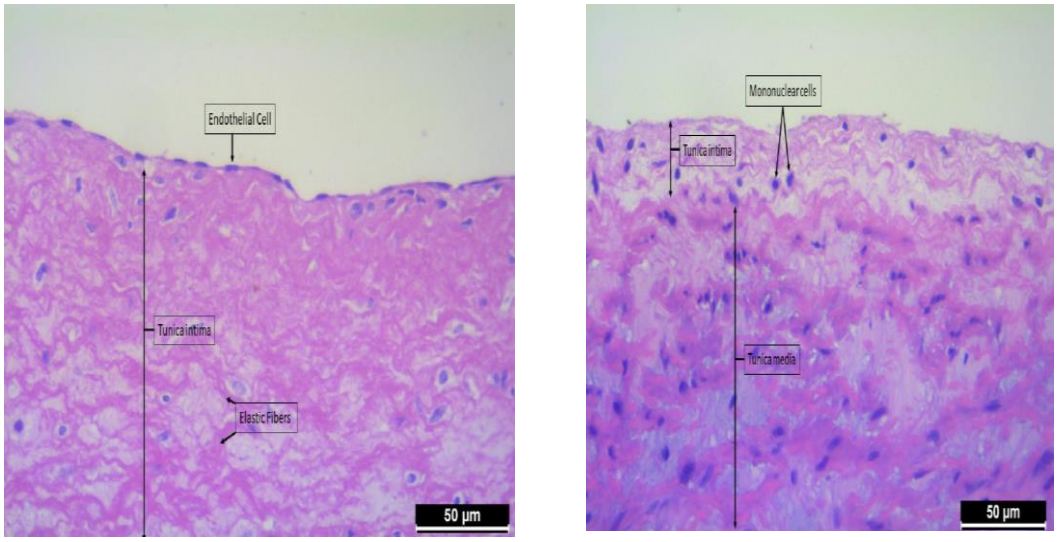
Animal No.P2 Aortal Proximal H&E 10X



Animal No.P2Aortal Mid H&E 10X



Animal No.P2Aortal Distal H&E 10X



Animal No.P1 Aortal Mid H&E 40X **Animal No.P2 Aortal Mid H&E 40X**

Figure 6: Hematoxylin and Eosin (H&E) Image of animal P1&P2:

The study found that the Valvuloplasty Embossing Balloon did not have thrombogenicity activity in the test animals. The balloon was tested for inflation and deflation in the target region of descending aorta with the support of a stiff guidewire and was found to perform activities related to valvuloplasty with no device-related coagulopathic local lesions. The balloon showed prominent visualization under angiographic imaging with good retrograde contrast flow throughout the balloon inflation and deflation time in the target site with no evidence of thrombus formation. The test item and the target sites showed no sign of thrombus or coagulation due to the surface material of the test item throughout the balloon inflation and deflation time in the target sites and after explanting the test item from the target sites. The post-procedural necropsy of the animals showed no thrombus formation or injury to the luminal interface in the aorta. Thrombus formation or thromboembolism was not observed in any examined tissue. There was no difference in the hematology, clinical chemistry parameters in the blood samples taken before and after the procedure. The mean thrombus formation score was 0 (No significant thrombosis) for the test item. The test item was thromboresistant in both the regional and downstream areas. No adverse events or safety concerns were found in this study.

DISCUSSION

The study aimed to evaluate the thrombo-resistance of the Valvuloplasty Embossing Balloon in a clinically relevant porcine model. The selection of male swine for this study was in line with accepted standards for preclinical testing. By fasting the animals overnight without access to water, potential confounding factors related to food intake were minimized.

The deployment of the balloon in the descending thoracic aorta, supported by a stiff guide wire, was a strategic choice to avoid complications such as cardiac arrest and difficulty in balloon deflation in the valvular space. DesP1te attempts to reach the valvular space, the absence of calcified or hard lesions at the valve and high ventricular pressure necessitated targeting the descending aorta adjacent to the valve. This decision did not impact the study outcome, as the primary objective was to assess the device's thrombogenicity.

During the study, the Valvuloplasty Embossing Balloon demonstrated excellent performance under angiographic imaging, with consistent retrograde contrast flow throughout inflation and deflation. Importantly, no evidence of thrombus formation or coagulation was observed on the balloon's surface or at the target sites, both during and after the procedure. The absence of thrombus formation or

thromboembolism in the examined tissues further supported the device's thrombo-resistance.

Hematology and clinical chemistry parameters remained consistent before and after the procedure, indicating the absence of systemic effects related to the device. The mean thrombus formation score of 0 indicated no significant thrombosis, affirming the thromboresistant nature of the test item in both regional and downstream areas. Notably, no adverse events or safety concerns were identified during the study, underscoring the device's safety profile.

CONCLUSION

The Valvuloplasty Embossing Balloon, tested for thrombo-resistance in a porcine model the study adhered to ISO 10993-4:2017 and standard operating procedures. Two male swine were used, and the balloon was deployed in the descending thoracic aorta using a guide wire. The balloon showed excellent trackability and rapid inflation and deflation within 30 seconds using the Introducer Sheath. It achieved good retrograde contrast flow without thrombus formation or coagulation at the target site. Post-procedure necropsy revealed no thrombus or injury in the aorta, and blood analyses showed no significant changes. The Valvuloplasty Embossing Balloon exhibited thromboresistance in both regional and downstream areas, with no observed adverse events, confirming its safety and efficacy for clinical use. The results of the study indicate that the balloon does not have thrombogenicity activity in test animals. The balloon was tested for inflation and deflation in the target region of the descending aorta with the support of a guidewire and performed activities related to valvuloplasty without device-related coagulopathic local lesions.

Declaration by Authors

Ethical Approval: Approved

Acknowledgement: None

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Conflict of Interest: The authors declare no conflict of interest.

REFERENCES

1. Bao, B., Saito, N., Watanabe, S., Tokushige, A., Yamamoto, E., Kawase, Y., … Inoue, K. (2013). A novel device for antegrade percutaneous balloon aortic valvuloplasty: Feasibility of the looped inoue balloon technique in swine model. Catheterization and Cardiovascular Interventions, n/a–n/a. doi:10.1002/ccd.24908
2. Seconds Count. Valvuloplasty for Heart Valve Problems (http://www.secondscount.org/sc-news-detail-2/valvuloplasty-heart-valve-problems-2#.YVIlOLhKhPZ). Accessed 6/9/2022.
3. Zain Z, Zadinello M, Menahem S, Brizard C. Neonatal isolated critical aortic valve stenosis: balloon valvuloplasty or surgical valvotomy. Heart Lung Circ. 2006 Feb;15(1):18-23. doi: 10.1016/j.hlc.2005.02.003. Epub 2005 Jul 25. PMID: 16473786.
4. Kuhn MA, Latson LA, Cheatham JP, Fletcher SE, Foreman C. Management of pediatric patients with isolated valvar aortic stenosis by balloon aortic valvuloplasty. Cathet Cardiovasc Diagn. 1996 Sep;39(1):55-61. doi:10.1002/(SICI)1097-0304(199609)39:1<55:AID-CCD12>3.0.CO;2-N. PMID: 8874948.
5. Merck Manual (Consumer Version). Biology of the Heart (https://www.merckmanuals.com/home/heart-and-blood-vessel-disorders/biology-of-the-heart-and-blood-vessels/biology-of-the-heart). Accessed 10/6/2021.
6. Galon, M. Z., Takimura, C. K., Figueira Chaves, M. J., de Campos, J. C., Eduardo Krieger, J., Gutierrez, P. S., … Neto, P. A. L. (2013). Porcine Model for the Evaluation and Development of Catheter-Based Coronary Devices: An Essential Preclinical Tool. Revista Brasileira de Cardiologia Invasiva (English Edition), 21(4), 378–383. doi:10.1016/s2214-1235(15)30162-9
7. Demkow M, Ruzyllo W, Ksiezycka E, Szaroszyk W, Lubiszewska B, Przyluski J, Rozanski J, Wilczynski J, Hoffman P, Rydlewska-Sadowska W. Long-term follow-up results of balloon valvuloplasty for congenital aortic stenosis: predictors of late

outcome. J Invasive Cardiol. 1999 Apr;11(4):220-6. PMID: 10745516.

8. Galbas MC, Straky HC, Meissner F, Reuter J, Schimmel M, Grundmann S, Czerny M, Bothe W. Cardiac dimensions and hemodynamics in healthy juvenile Landrace swine. Cardiovasc Ultrasound. 2024 Jan 16;22(1):3. doi: 10.1186/s12947-023-00321-9. PMID: 38229189; PMCID: PMC10790455.
9. Kim, J., Koo, BK. & Knoblich, J.A. Human organoids: model systems for human biology and medicine. *Nat Rev Mol Cell Biol* 21, 571–584 (2020). https://doi.org/10.1038/s41580-020-0259-3
10. Biological Evaluation of Medical Devices - Part 2: Animal welfare requirements. ISO 10993-2:2006(E).

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