Animal Disease Model and in Vivo Function Analysis (for Beginners)

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Animal Models of Heart failure

Species and technique	Selected references	Comments
Rat	1028[a]	
Coronary ligation	[44,49,59,60]	Clinical characteristics similar to human CHF; survival studies
Aortic banding	[62–64]	Studies of transition from hypertrophy to failure; survival studies
Salt-sensitive hypertension	[66,67]	Studies of transition from hypertrophy to failure
Spontaneous hypertension	[68–71]	Extracellular matrix changes; apoptosis; studies of transition from hypertrophy to failure
SH–HF/Mcc-facp	[72–76]	Altered NOS expression; altered calcium triggered calcium release
Aorto-caval fistula	[184,185]	Left ventricular hypertrophy; moderate LV dysfunction
Toxic cardiomyopathy	[186–189]	Decreased myocardial performance; myocyte loss with chronic ethanol application. Cardiomyopathy following catecholamine infusion or associated with *Diabetes mellitus*
Dog	148[a]	
Pacing tachycardia	[79–89,100–106]	Studies of remodeling and neurohumoral activation; studies on molecular mechanism of subcellular dysfunction; no hypertrophy
Coronary artery ligation	[111–115]	Studies on progression of heart failure; high mortality and arrhythmias
Direct-current shock	[115]	Studies of neurohumoral mechanisms
Volume overload -aorto-caval fistula -mitral regurgitation	[116–120]	Studies of neurohumoral mechanisms and therapeutic interventions
Vena caval constriction	[189]	Low cardiac output failure
Toxic cardiomyopathy	[190]	Left ventricular dysfunction
Genetic	[98]	Spontaneous cardiomyopathy in Doberman Pinscher dogs
Pig	43[a]	
Pacing tachycardia	[107–110]	Comparable with dog model for most aspects
Coronary artery ligation	[191]	Congestive heart failure; altered myocardial energetics

Rabbit	43[a]	
Volume and pressure overload	[122–126]	Myocardial alterations similar to failing human myocardium
Pacing tachycardia	[127–131,192]	Myocardial alteration similar to failing human myocardium
Toxic cardiomyopathy	[132]	Studies of functional consequences of altered ryanodine receptors
Guinea pig	31[a]	
Aortic banding	[134,135,193]	Myocardial function and alteration of calcium handling similar to human heart failure
Syrian hamster	10[a]	
Genetic	[136–147]	Hypertrophy and failure; alterations critically dependent on strain and age
Cat	11[a]	
Pulmonary artery constriction	[194,195]	Transition from compensated right ventricular hypertrophy to failure
Turkey	9[a]	
Toxic cardiomyopathy	[196]	Alteration of calcium handling and myocardial energetics
Bovine	25[a]	
Genetic	[197]	Similar to human heart failure regarding changes in β-adrenergic system
Sheep	17[a]	
Pacing tachycardia	[198,199]	Similar to dog and swine model of pacing tachycardia
Aortic constriction	[200]	Transition from compensated hypertrophy to left ventricular dysfunction

[a] Total number of references in this species (failure and animal species) 1993–1997.

Cardiovascular Research 39 (1998) 60–76

Animal Models of Atherosclerosis

Atherosclerosis

1. **Endothelial denudation model**
2. **Stent implantation model (mouse, rat, pig)**
3. **Transgenic mice**

Today

1. Myocardial infarction

(Rat and Mouse: Coronary artery ligation model)

2. Pressure over-load induced LV Hypertrophy

(Transverse aortic constriction model : TAC)

3. Restenosis

(Carotid artery balloon injury model)

Anesthesia (Rat and Mouse)

Barbiturate
Rompun (Xylazine HCL) : 23.3mg/mL
Ketamine : 50mg/1ml
Zoletil 50 (Tiletamine 125mg, Zolazepam 125mg/5ml)

표 19-1. 마우스 : 선별된 마취제와 진정제 용량[a]

Drug	Dose	Route	Duration
Tranquilizers			
Acepromazine	2–5 mg/kg	SC, IP	? min
Diazepam	5 mg/kg	IP	? min
Midazolam	5 mg/kg	IP	? min
Anesthestics			
Barbiturates			
Thiopental	25–50 mg/kg	IV	10 min
Methohexital	8–16 mg/kg	IV	2–5 min
Pentobarbital	30–50 mg/kg	IP	20–40 min
Dissociative agents and combinations			
Ketamine + acepromazine	100 mg/kg 5 mg/kg	IP	20–30 min
Ketamine + medetomidine	75 mg/kg 1 mg/kg	IP	20–30 min
Ketamine + xylazine	80–100 mg/kg 10 mg/kg	IP	20–30 min
Neuroleptanesthetics			
Fentanyl-fluanisone + (Hypnorm) midazolam	10.0 ml/kg[b]	IP	30–40 min
Inhalant agents			
CO_2/O_2	70–80%/20–30%	Inhalant	
Halothane	1–4%, to effect	Inhalant	
Isoflurane	1–4%, to effect	Inhalant	
Other agents			
Alphaxolone-alphadolone	10–15 mg/kg	IV	5–10 min
Propofol	12–26 mg/kg	IV	5–10 min
Tribromoethanol	240 mg/kg	IP	15–45 min
Analgesics			
Buprenorphine	0.05–0.1 mg/kg	SC	8 hr
Butorphanol	1–5 mg/kg	SC	4 hr
Morphine	2.5 mg/kg	SC	2–4 hr
Acetaminophen	200 mg/kg	PO	? hr
Miscellaneous			
Atropine	0.04 mg/kg	SC	? min

[a] Doses adapted with modifications from Wixson and Smiler (1997a) and Flecknell (1996a).
[b] See Flecknell (1996a) for mixing instructions.

표 19-2. 랫드:약물 용량 [a]

Drug	Dose	Route	Duration
Tranquilizers			
Acepromazine	2–5 mg/kg	SC, IP	? min
Diazepam	5–15 mg/kg	SC	? min
Midazolam	5 mg/kg	IP	? min
Anesthestics			
Barbiturates			
Thiopental	20–40 mg/kg	IV	5–10 min
Methohexital	10–15 mg/kg	IV	5–10 min
Pentobarbital	40–60 mg/kg	IP	20–60 min
EMTU (inactin)	80–100 mg/kg	IP	60–240 min
Dissociative agents and combinations			
Ketamine + acepromazine	75 mg/kg 2.5 mg/kg	IP	20–30 min
Ketamine + medetomidine	75 mg/kg 0.5 mg/kg	IP	20–30 min
Ketamine + xylazine	40–75 mg/kg 5–10 mg/kg	IP	20–40 min
Neuroleptanesthetics			
Fentanyl-fluanisone + (Hypnorm) midazolam	2.7 ml/kg[b]	IP	30–40 min
Inhalant agents			
CO_2/O_2	70–80%/20–30%	Inhalant	
Halothane	1–4%, to effect	Inhalant	
Isoflurane	1–4%, to effect	Inhalant	
Other agents			
α-Chloralose	55–65 mg/kg	IP	8–10 hr
Alphaxolone-alphadolone	10–15 mg/kg	IV	5–10 min
Chloral hydrate	300–450 mg/kg	IP	60–120 min
Propofol	7.5–10 mg/kg	IV	5–10 min
Urethane	1000–1500 mg/kg	IP	8–24 hr
Analgesics			
Buprenorphine	0.01–0.05 mg/kg	SC	8–12 hr
Butorphanol	2 mg/kg	SC	4 hr
Carprofen	5 mg/kg	SC	? hr
Ketoprofen	5 mg/kg	SC	? hr
Morphine	2.5 mg/kg	SC	2–4 hr
Miscellaneous			
Atropine	0.04 mg/kg	SC	? min
Atipamezole	0.1–1 mg/kg, varies with α_2 dose	IP, SC	

[a] Doses adapted with modifications from Wixson and Smiler (1997a) and Flecknell (1996a).
[b] See Flecknell (1996a) for mixing instructions.

Today

1. Myocardial infarction

(Rat and Mouse: Coronary artery constriction)

2. Pressure over-load induced LV Hypertrophy

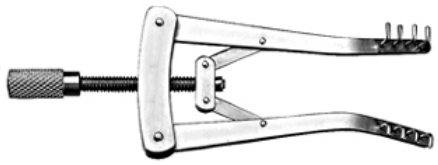
(Transverse aortic constriction model : TAC)

3. Restenosis

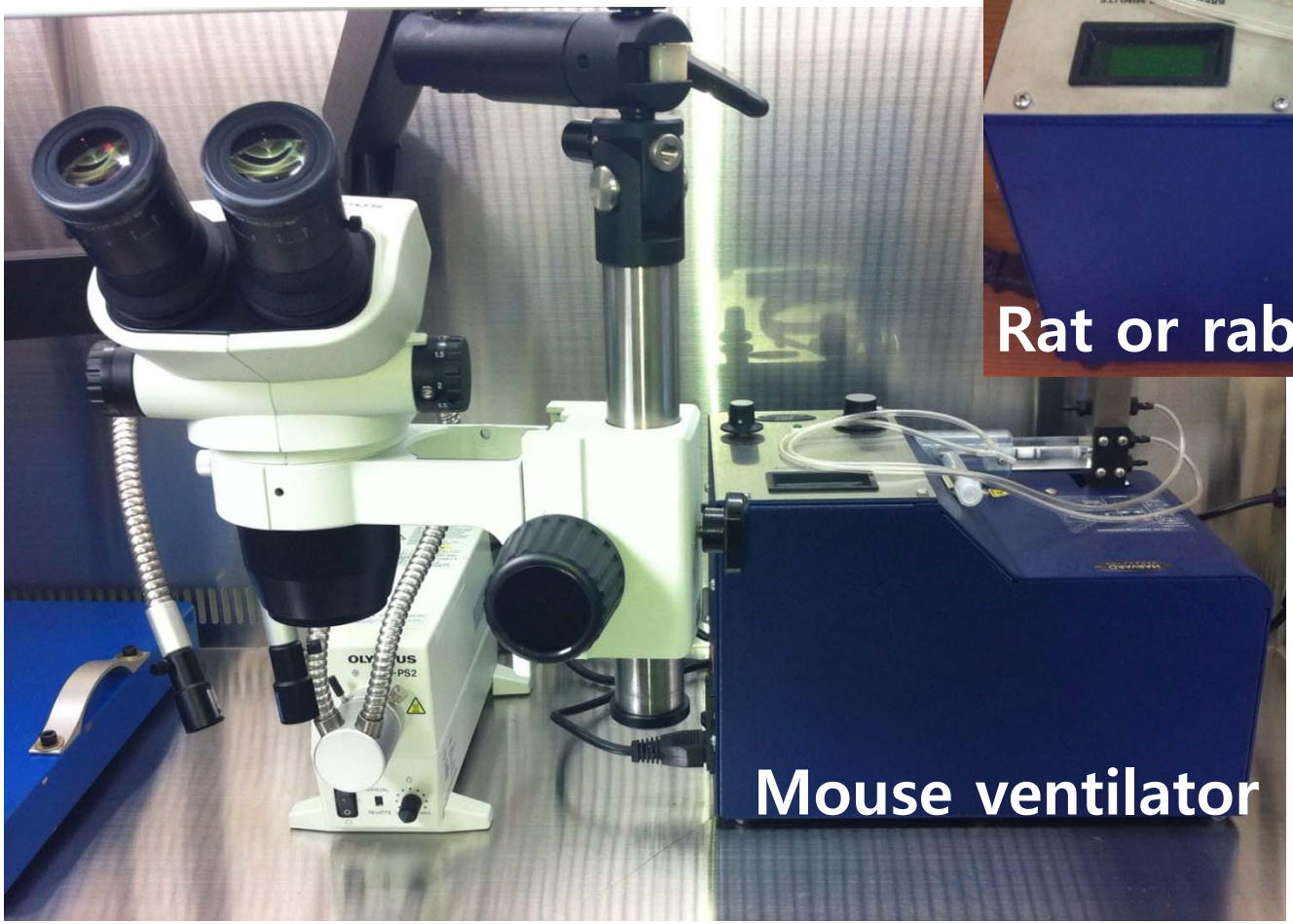
(Carotid artery balloon injury model)

Rat or Mouse Myocardial Infarction (MI)

1. Adson tissue forcep or forcep
2. Mosquito forcep
3. Metzenbaum scissors and straight scissor
4. Needle holder
5. Micro needle holder
6. Retractor
7. 5-0 silk (needle shape: taper)



Rat or Mouse Myocardial Infarction (MI)

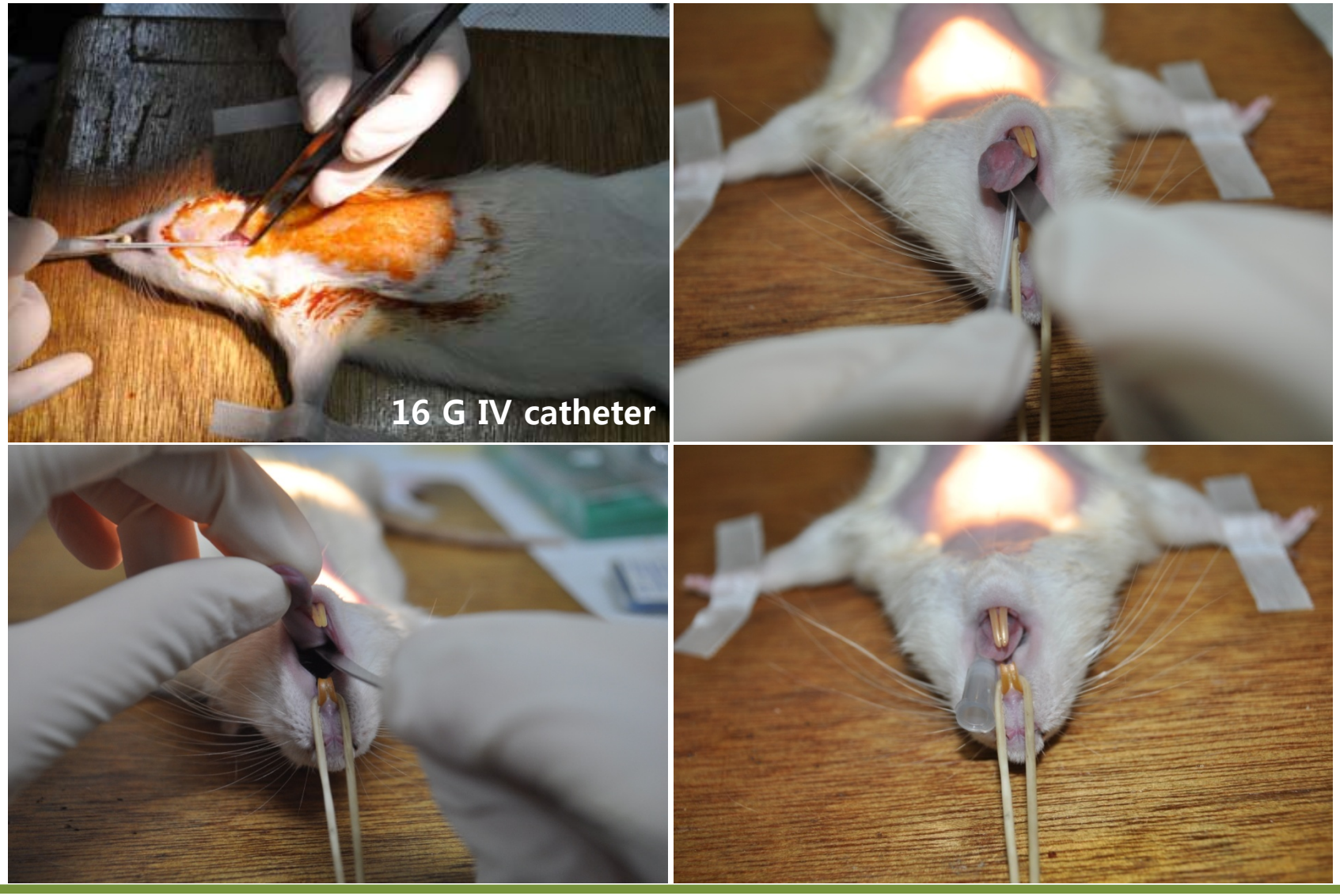


Microscopy
Light source

Rat or Mouse Myocardial Infarction (MI)

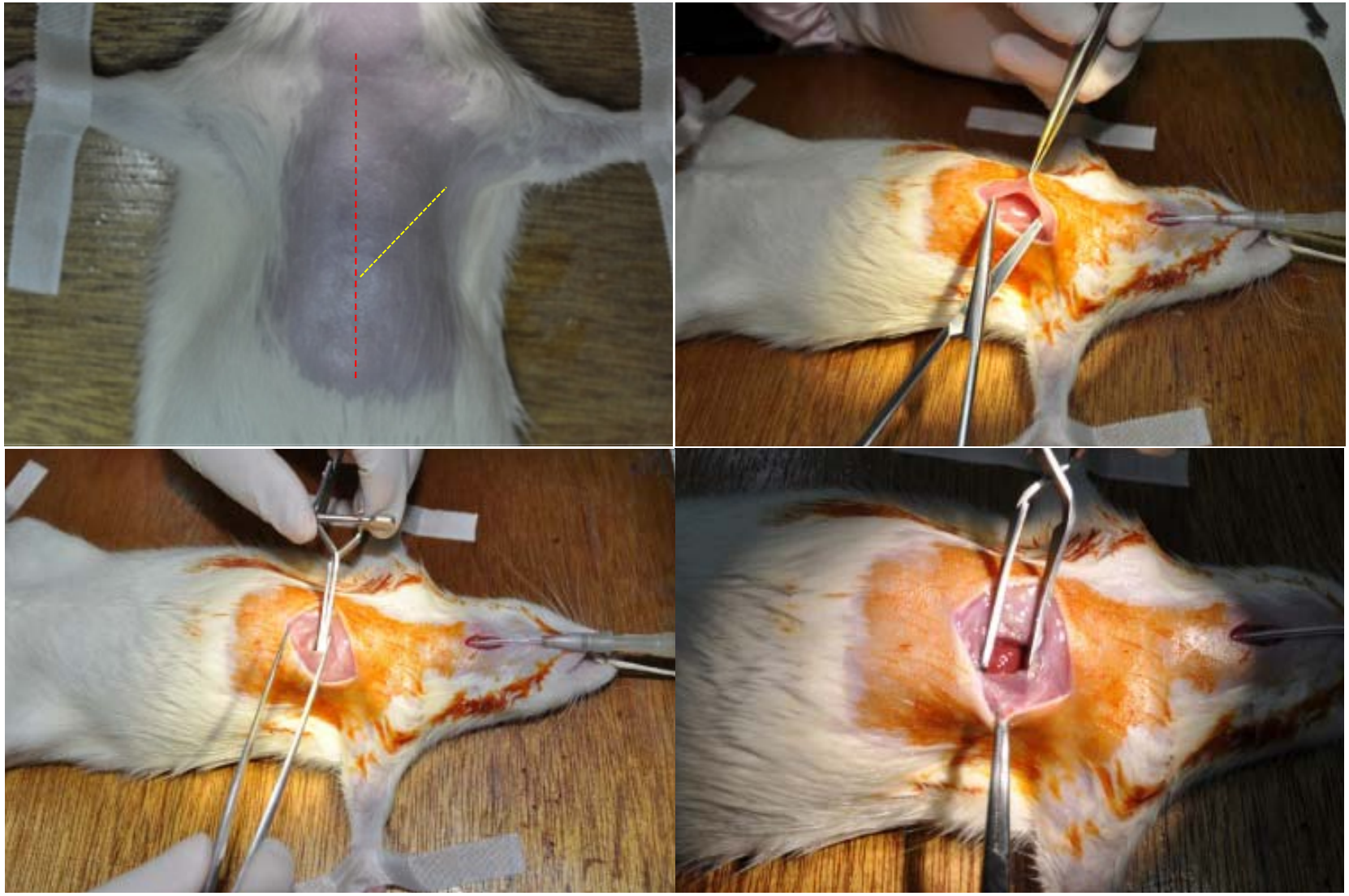
Rat or Mouse Myocardial Infarction (MI)

-기관 절개술 (Tracheostomy), 기관 삽입 (Endotracheal Inturbation)



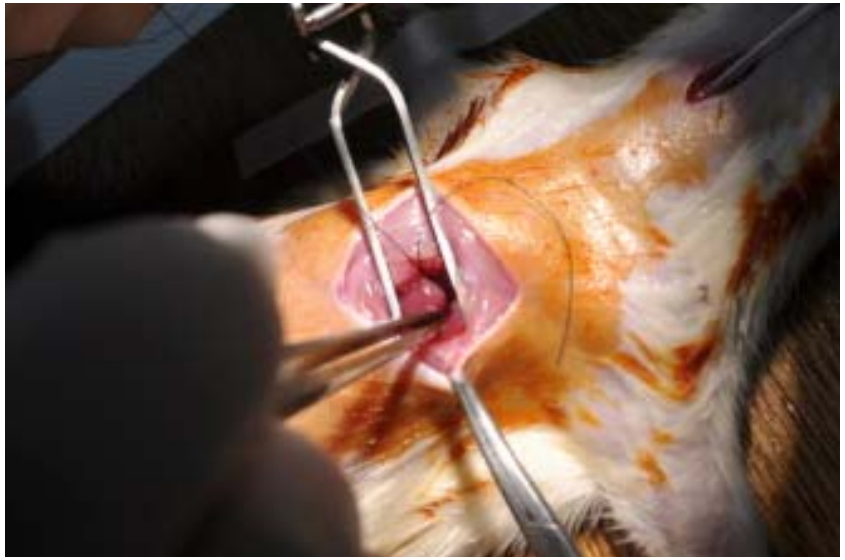
Rat or Mouse Myocardial Infarction (MI)

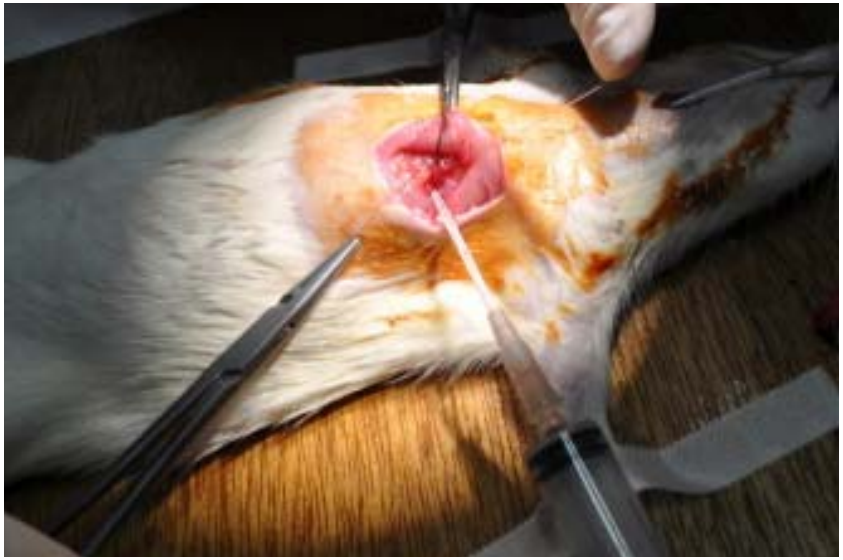
-Thoracotomy

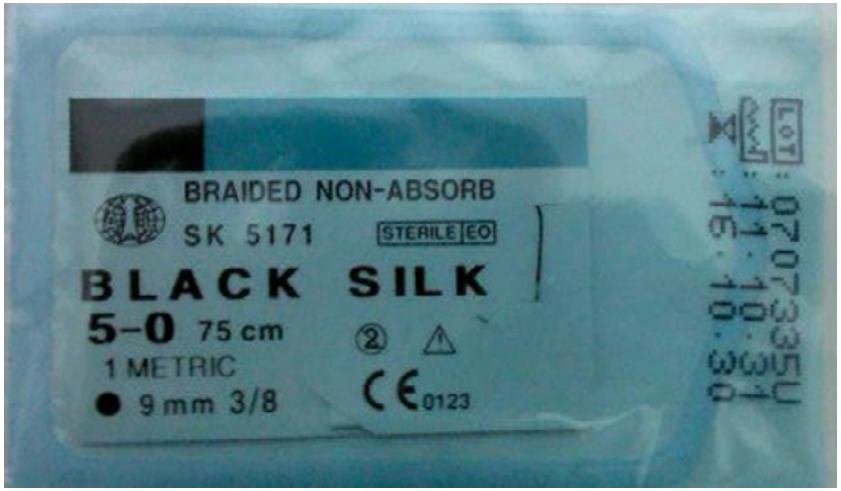


Rat or Mouse Myocardial Infarction (MI)

-Thoracotomy

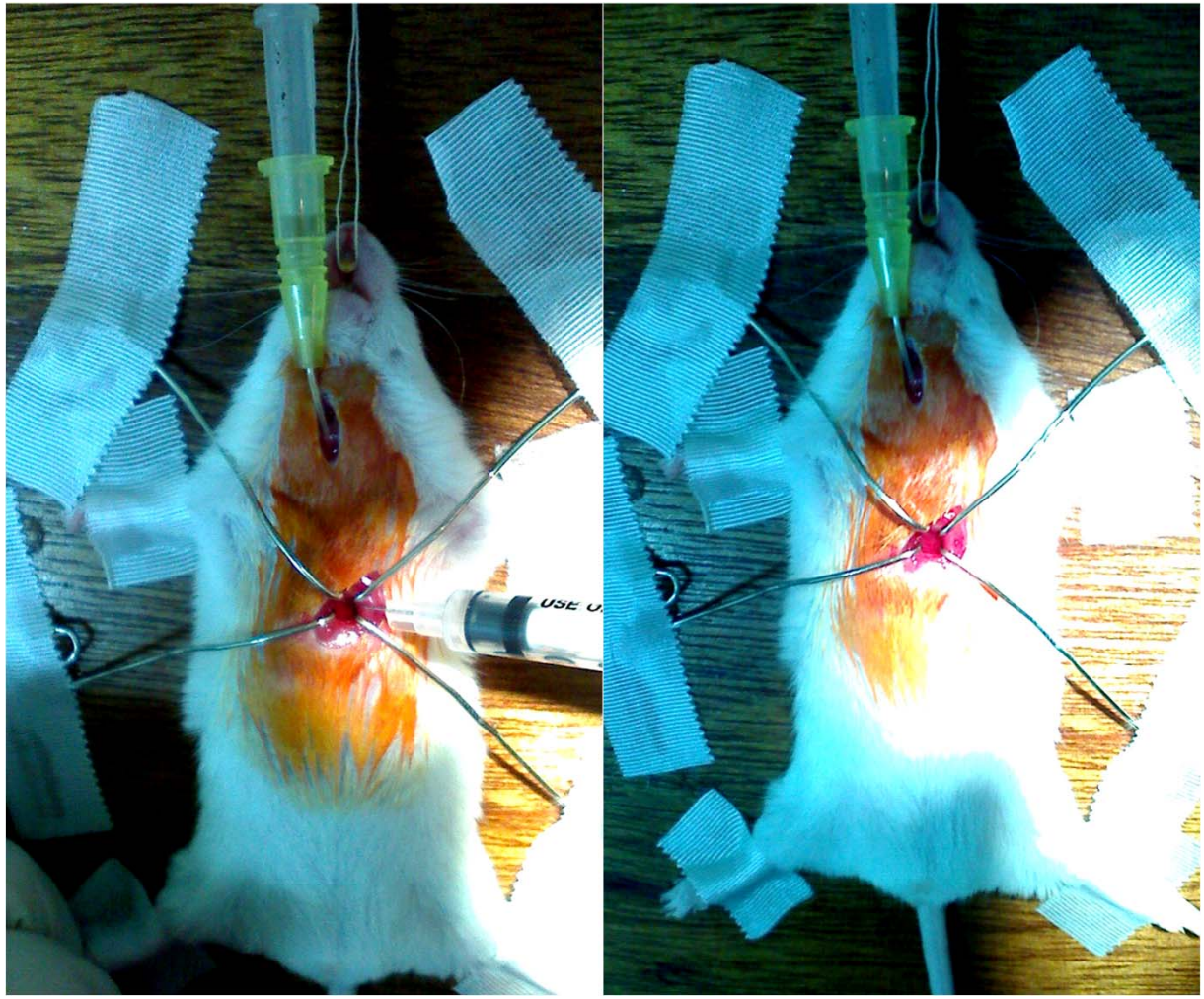






Mouse Myocardial Infarction (MI)

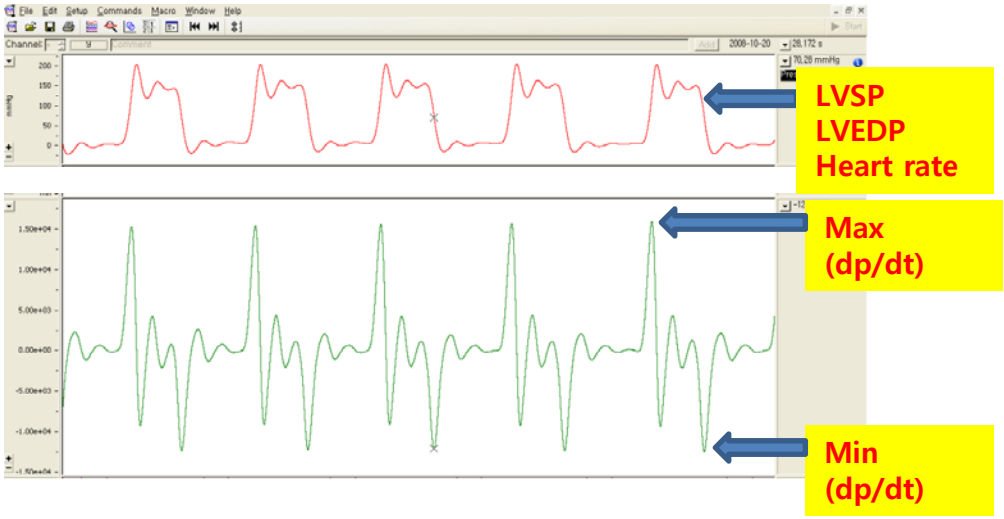
-Thoracotomy



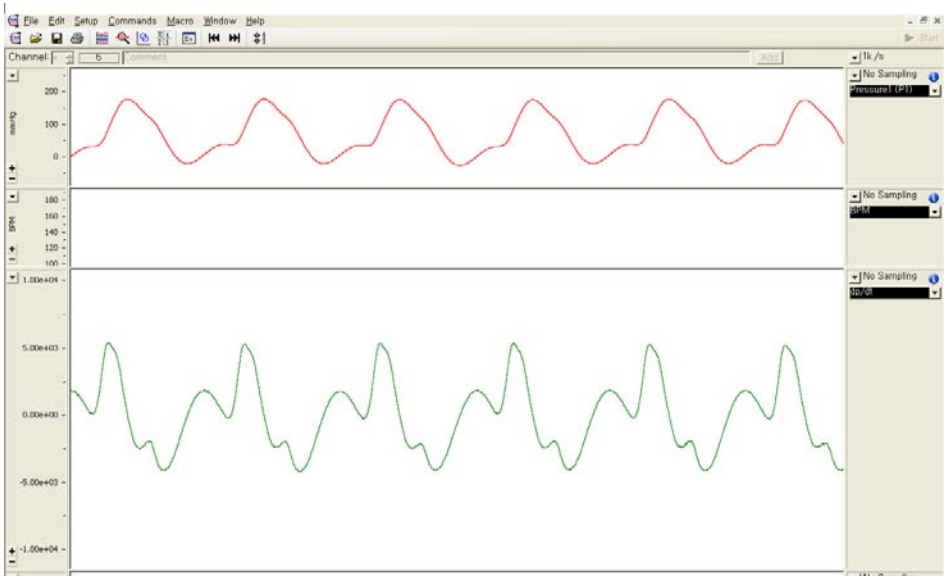
20-24G IV catheter
7-0, 8-0 Silk
Micro needle holder

Function analysis : Echo and Millar system

Hemodynamic factor



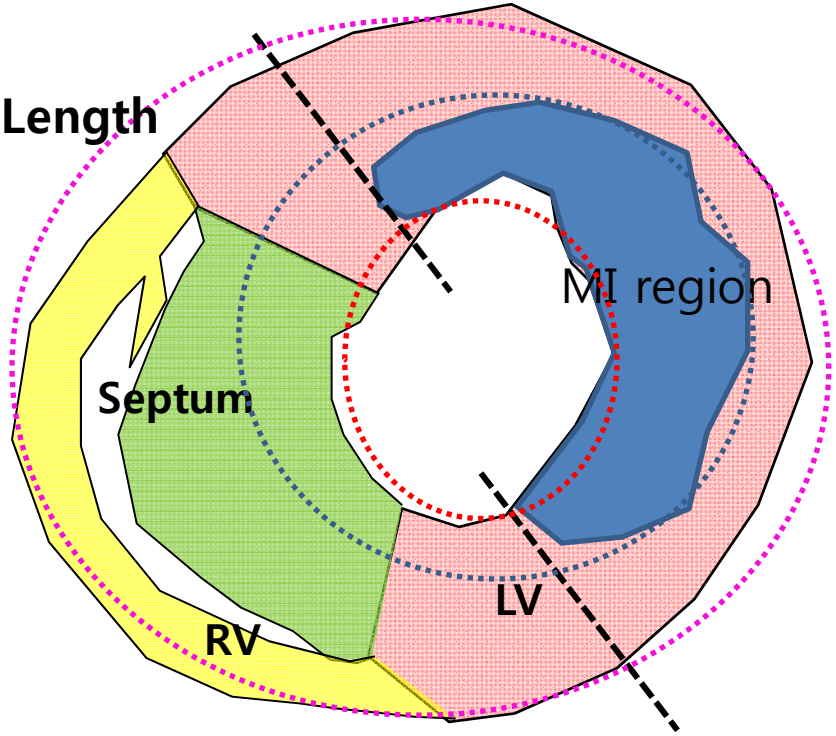
Echocardiography

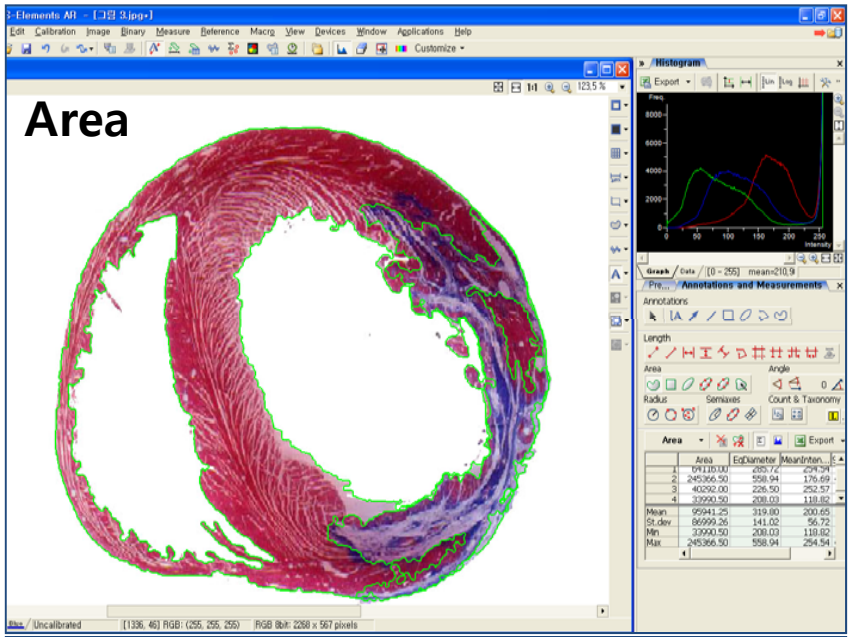


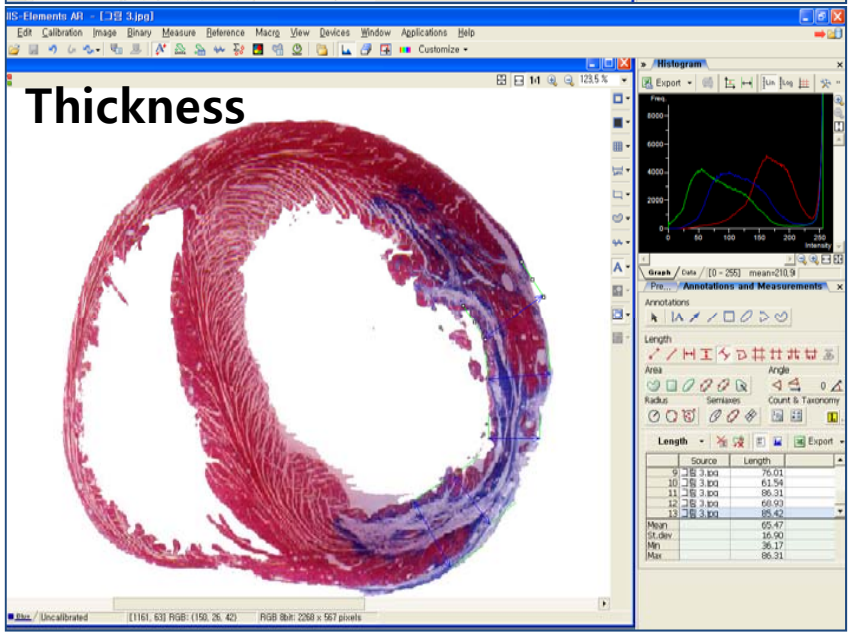
Histomorphometric analysis

1. MI region evaluation

- Area and Area ratio
- Thickness
- Length and Length ratio







Today

1. Myocardial infarction

(Rat and Mouse: Coronary artery ligation model)

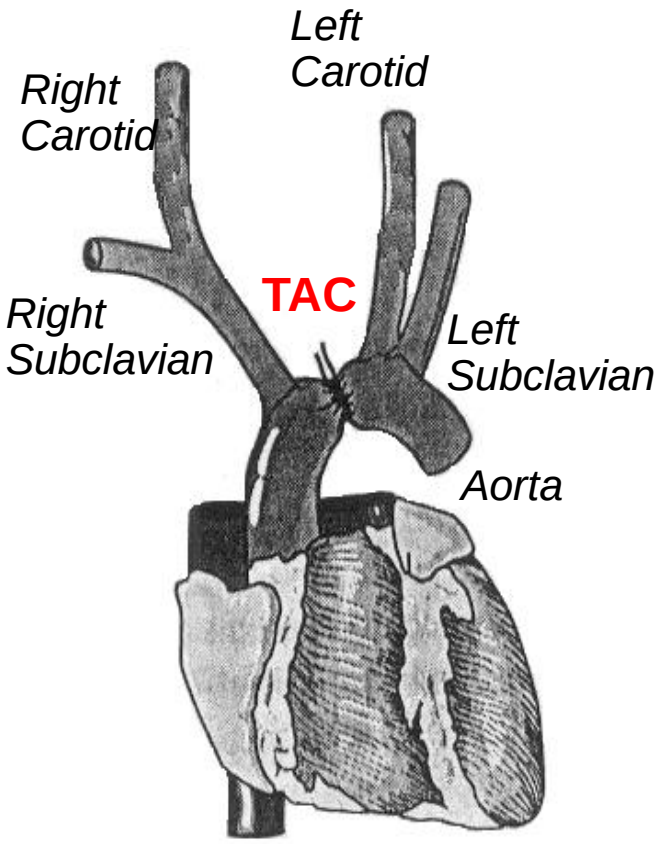
2. Pressure over-load induced LV Hypertrophy

(Transverse aortic constriction model : TAC)

3. Restenosis

(Carotid artery balloon injury model)

Transverse Aortic Constriction (TAC)



Surgical Metods

- Anesthesia : ketamine/xylazine
- Artificial ventilation
- TAC with 7-0 nylon tying over 26G bunt needle or untying
- Pneumothorax reduction

Changes by TAC

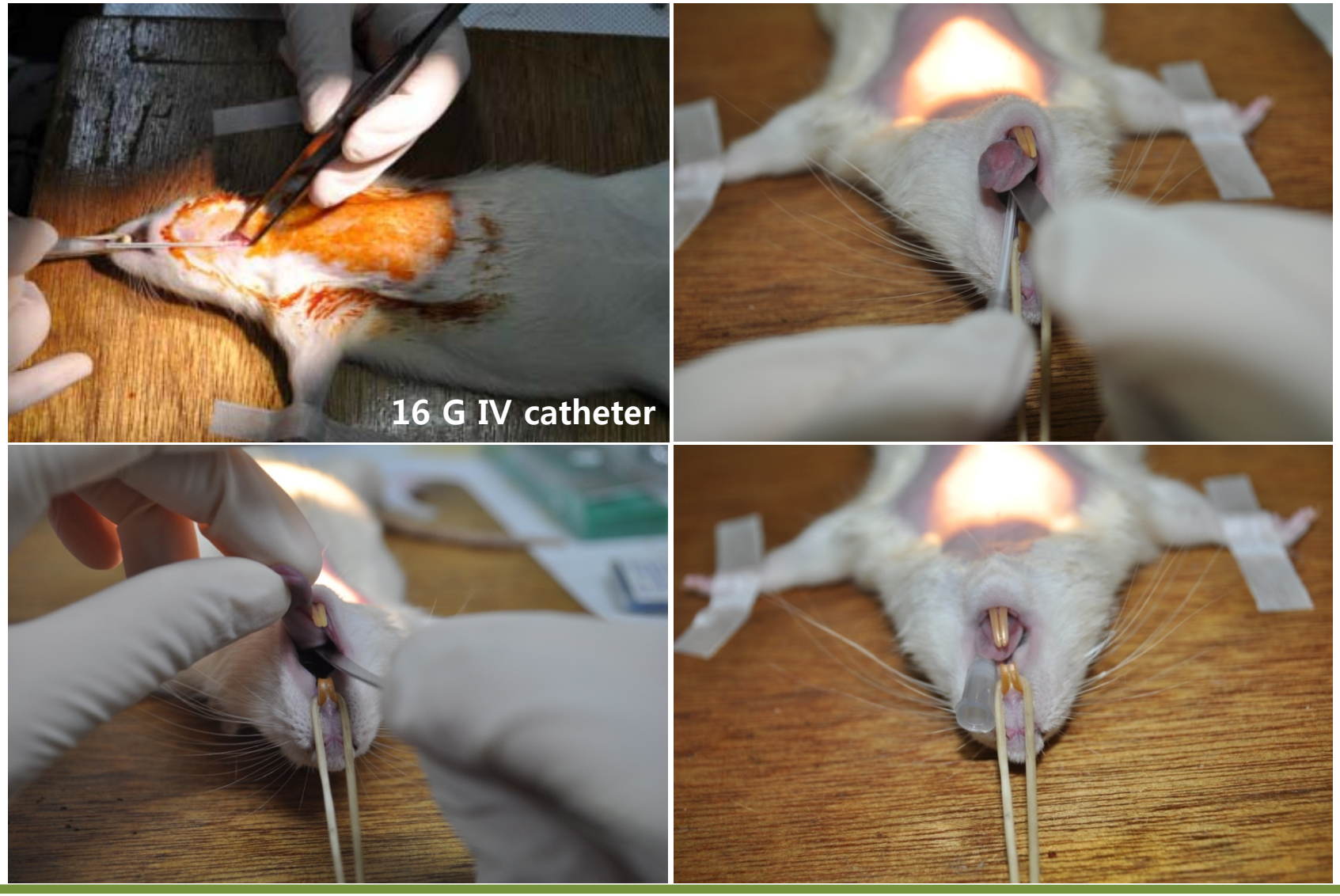
- Pressure gradient : 30-40mmHg
- Increase in HW (35%)
- Increase in ANF mRNA
- Increase in β-MHC
- Decrease in SERCA

Transverse Aortic Constriction (TAC)

1. Adson tissue forceps
2. Mosquito forceps
3. Scissor
4. Retractor
5. Needle holders
6. 4-0 or 5-0 silk)
7. Micro forceps
8. Modified needle

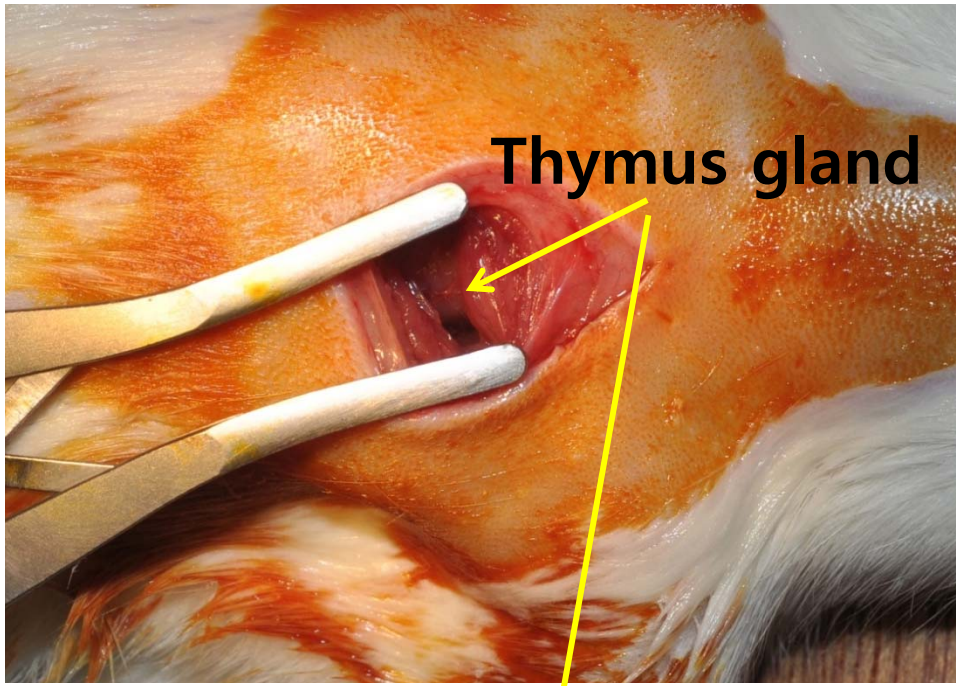
Rat or Mouse Myocardial Infarction (MI)

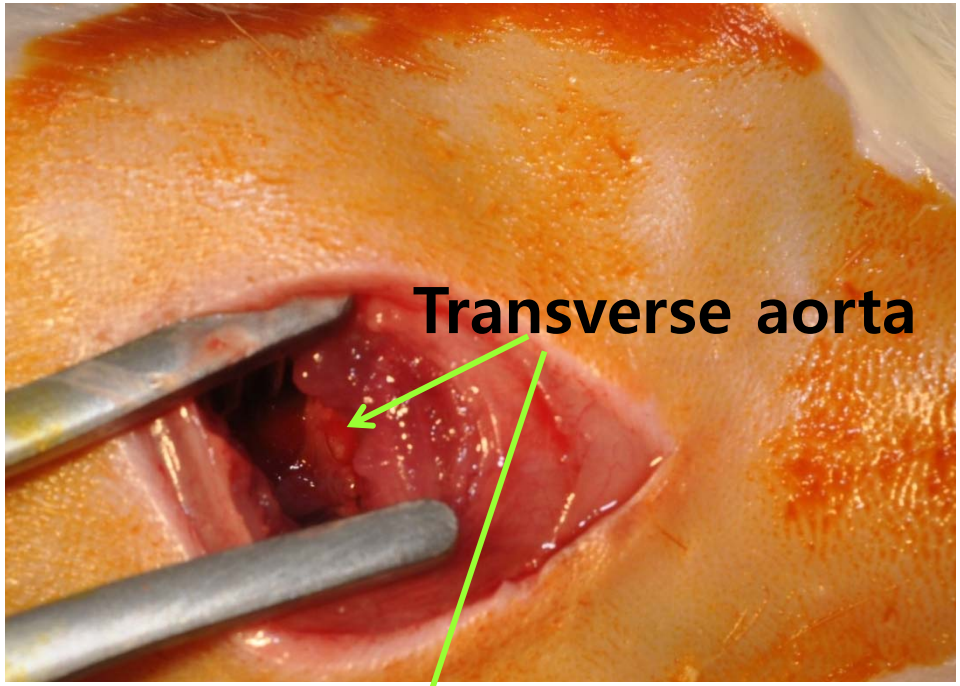
-기관 절개술 (Tracheostomy), 기관 삽입 (Endotracheal Inturbation)

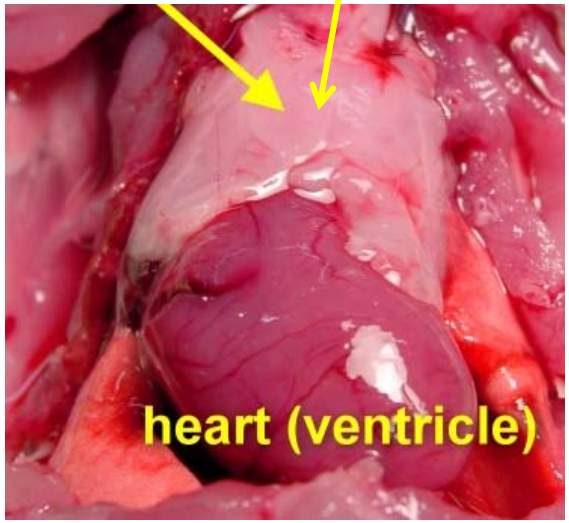


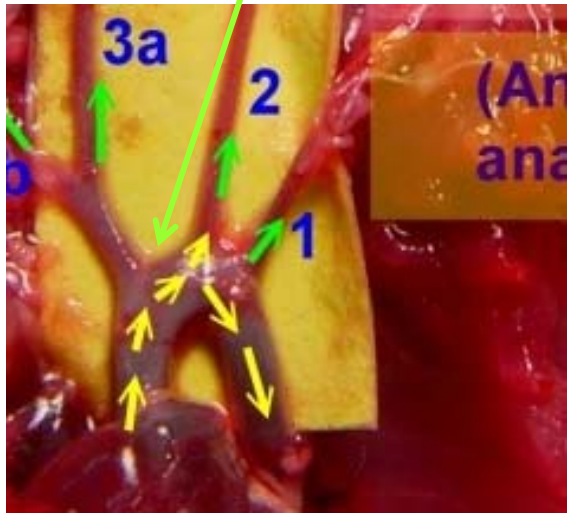
Pressure-Overload Induced LV Hypertrophy by Transverse Aortic Constriction (TAC)

Transverse Aortic Constriction (TAC)

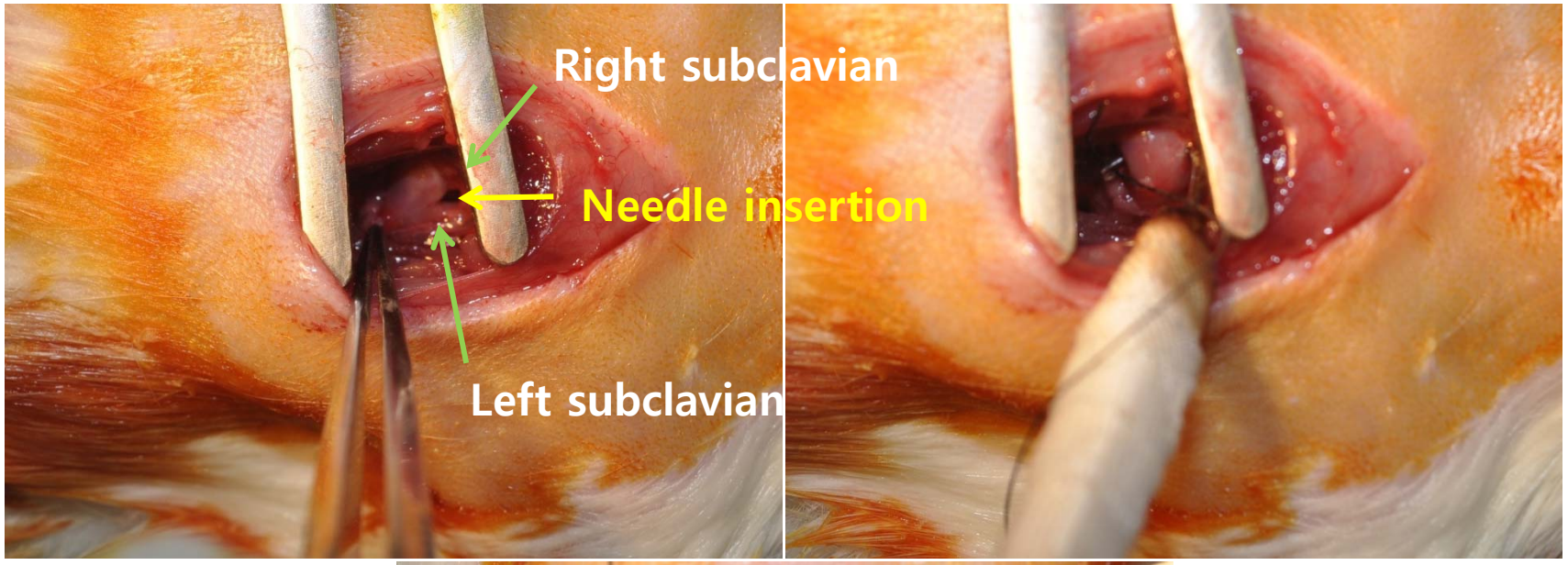


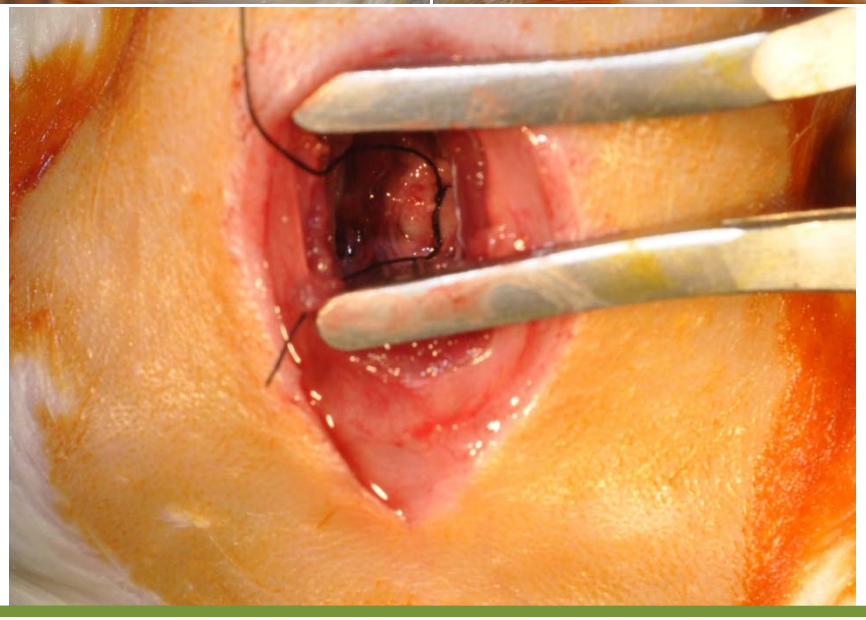


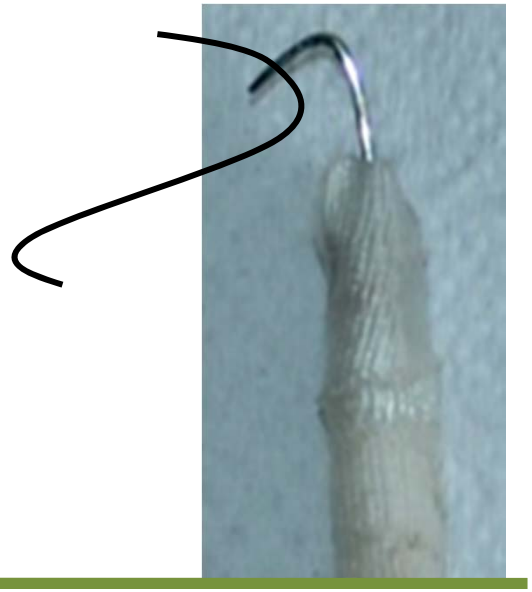




Transverse Aortic Constriction (TAC)







Histomorphometric analysis

Postmortem pathologic measurements of MAFbx KO

	WT		MAFbx KO	
	Sham	**Banding**	**Sham**	**Banding**
n	7	7	6	9
BW (g)	27.3±1.0	26.6±1.0	26.0±1.7	25.2±1.2
TL(mm)	17.7±0.3	17.8±0.3	17.2±0.1	17.2±0.2
LV (mg)	87.1±1.6	151.3±9.0[B]	86.3±4.6	112.2±5.7[A,D]
RV (mg)	17.4±0.6	23.4±2.1[A]	19.0±2.1	18.0±1.0
LVW/BW	3.22±0.12	5.68±0.20[B]	3.36±0.20	4.49±0.22[B,D]
Lung W/BW	5.08±0.10	10.10±1.77[B]	4.93±0.21	6.00±0.73[C]
Liver W/BW	47.2±2.2	44.5±1.5	44.9±3.8	40.9±2.0
LVW/TL	4.92±0.13	8.35±0.40[B]	4.76±0.23	6.52±0.29[B,D]
Lung/TL	7.79±0.19	12.56±1.34[B]	7.39±0.53	8.55±0.74[D]
Liver/TL	72.4±3.5	65.6±3.4	66.9±10.9	60.3±4.6

Data are mean ± SEM, [A]$P<0.05$, [B]$P<0.01$ compared with same genotype sham mice.
[C]$P<0.05$, [D]$P<0.01$ compared with WT 2 weeks after banding.

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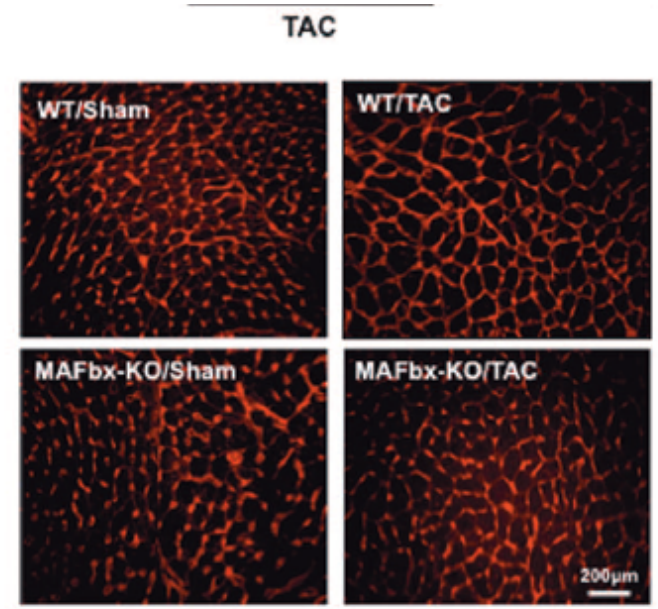
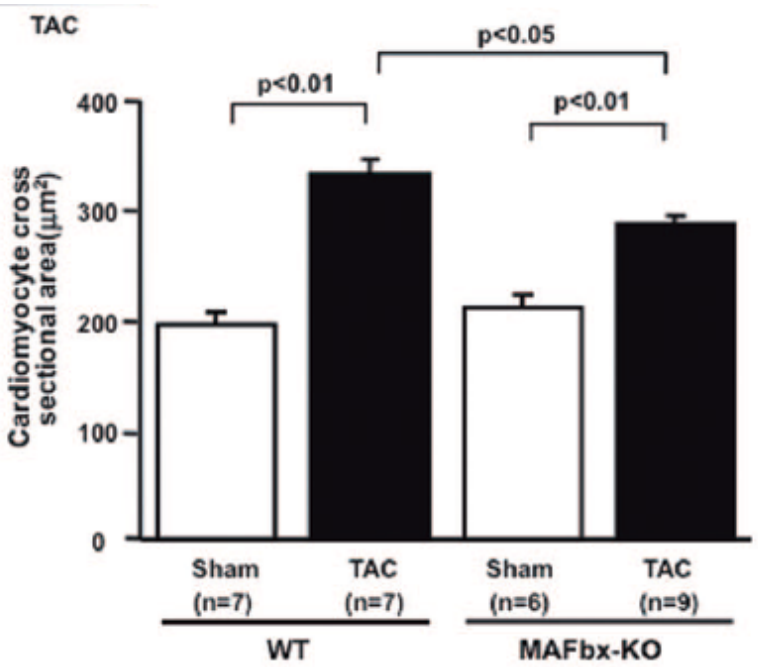
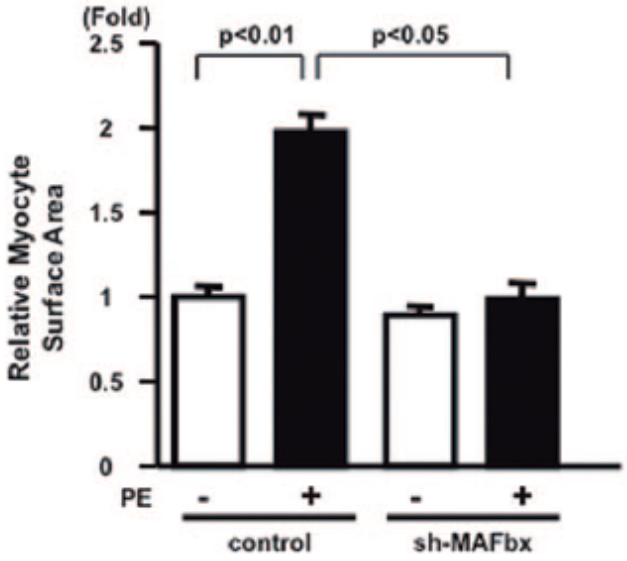
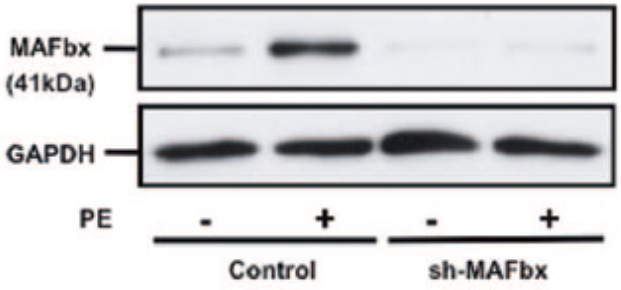
Histomorphometric analysis









Wheat germ agglutinin Ab

Soichiro Usui et al . 2011 Circ Res 2011;109:161-171

Today

1. Myocardial infarction

(Rat and Mouse: Coronary artery ligation model)

2. Pressure over-load induced LV Hypertrophy

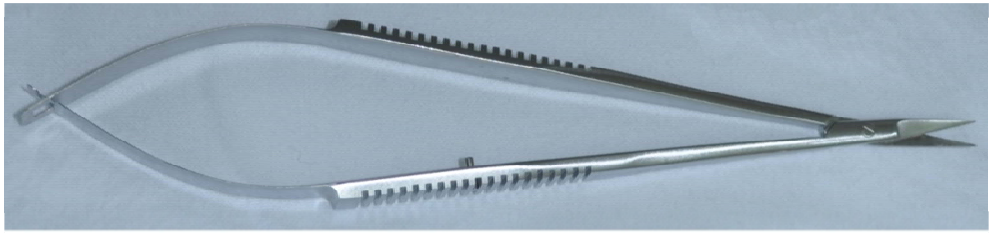
(Transverse aortic constriction model : TAC)

3. Restenosis

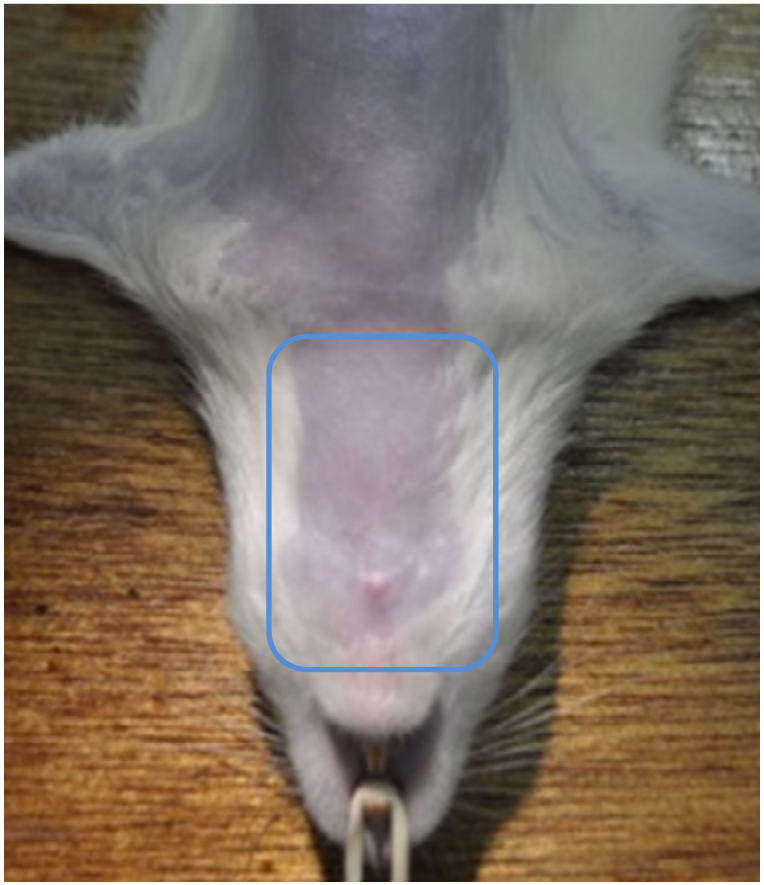
(Carotid artery balloon injury model)

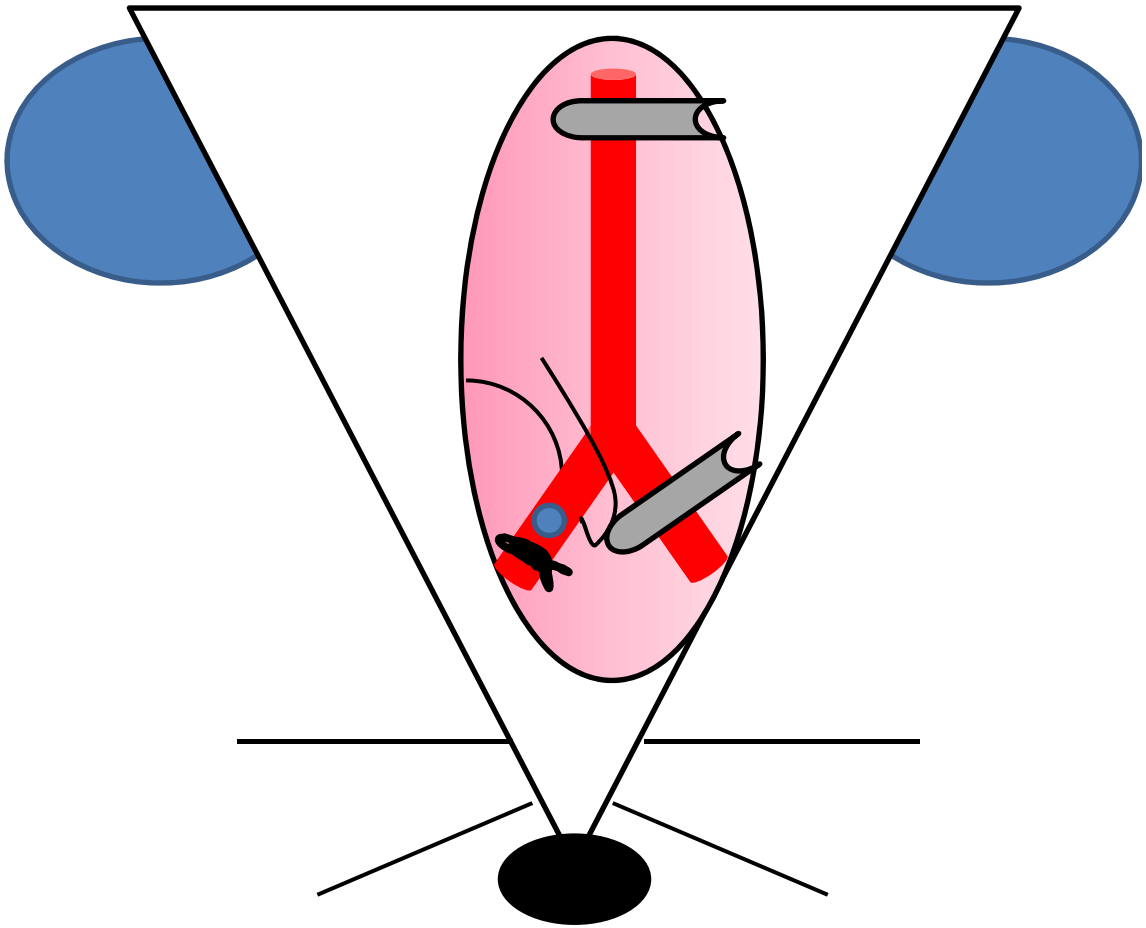
Common carotid endothelial cell denudation model (Carotid balloon injury model)

1. Adson tissue forcep, micro needle holder
2. Mosquito forcep
3. Metzenbaum scissors, Scissor)
4. Hemoclips (S&T, Microvascular clamp, 2-single clamps, 0.4-1.0 mm)
5. Needle holders
6. 5-0 silk
7. Hamilton Syringe
8. Forgaty balloon catheter



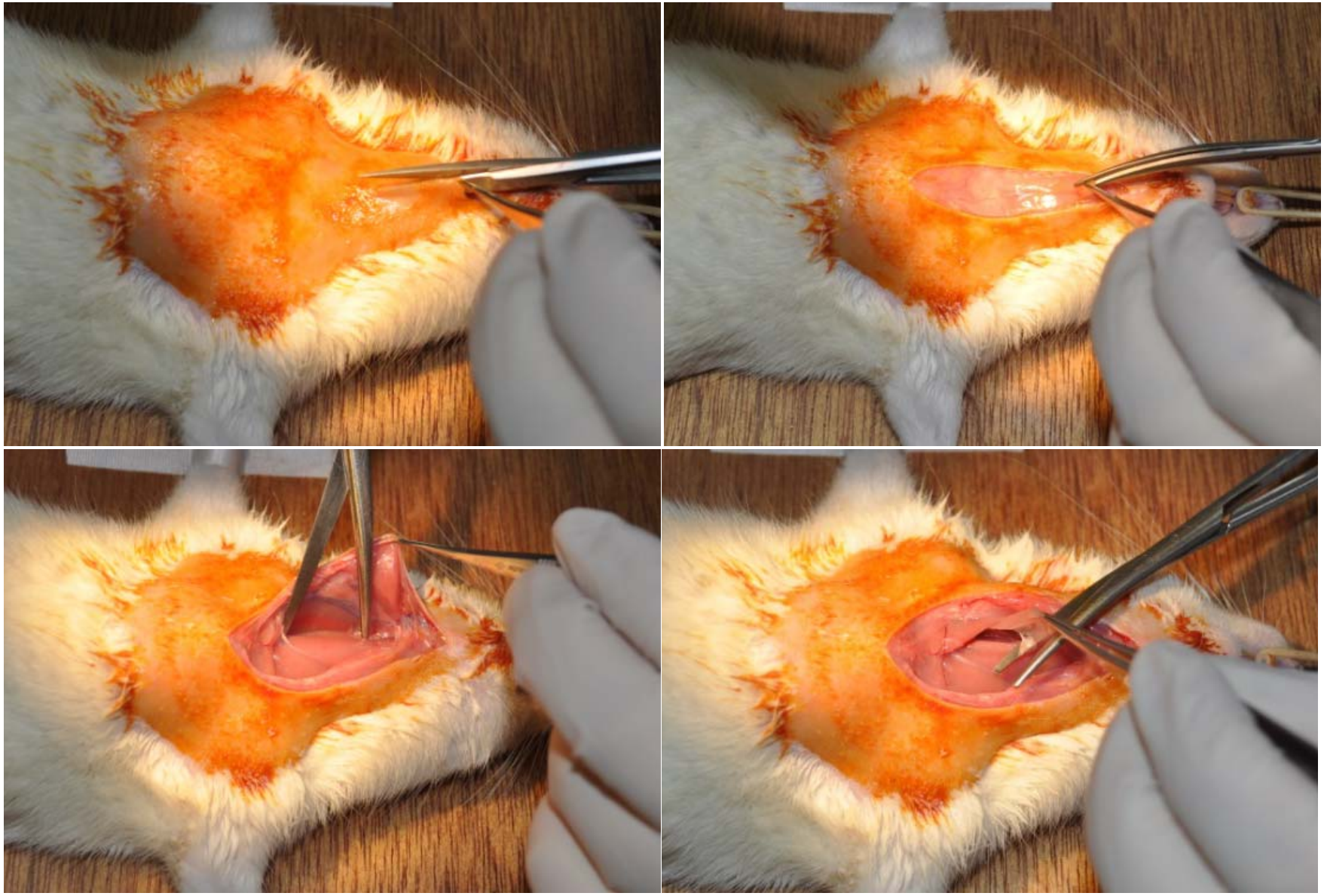
Carotid artery balloon injury model





1. Right common carotid artery exposure

Carotid artery balloon injury model



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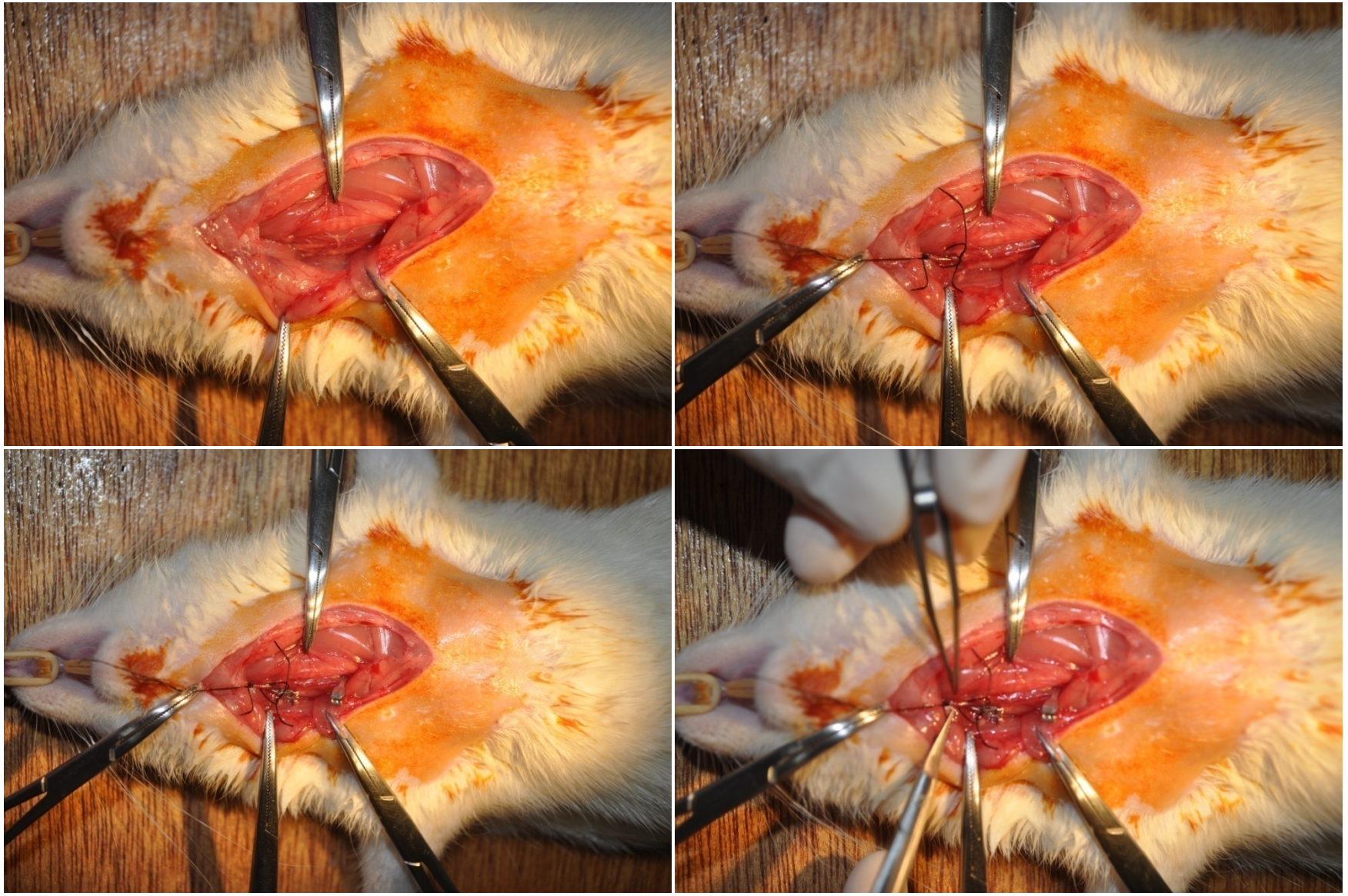

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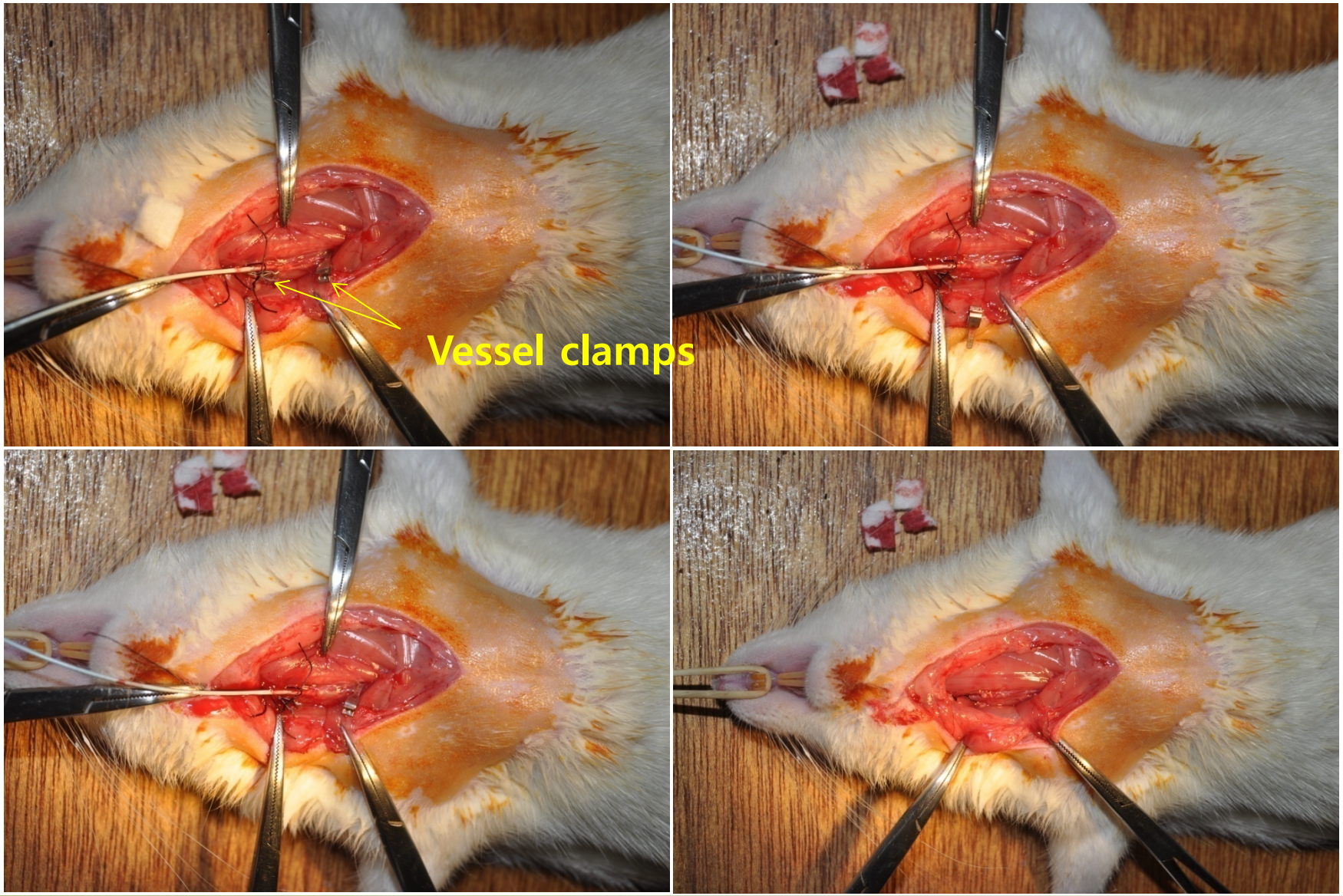
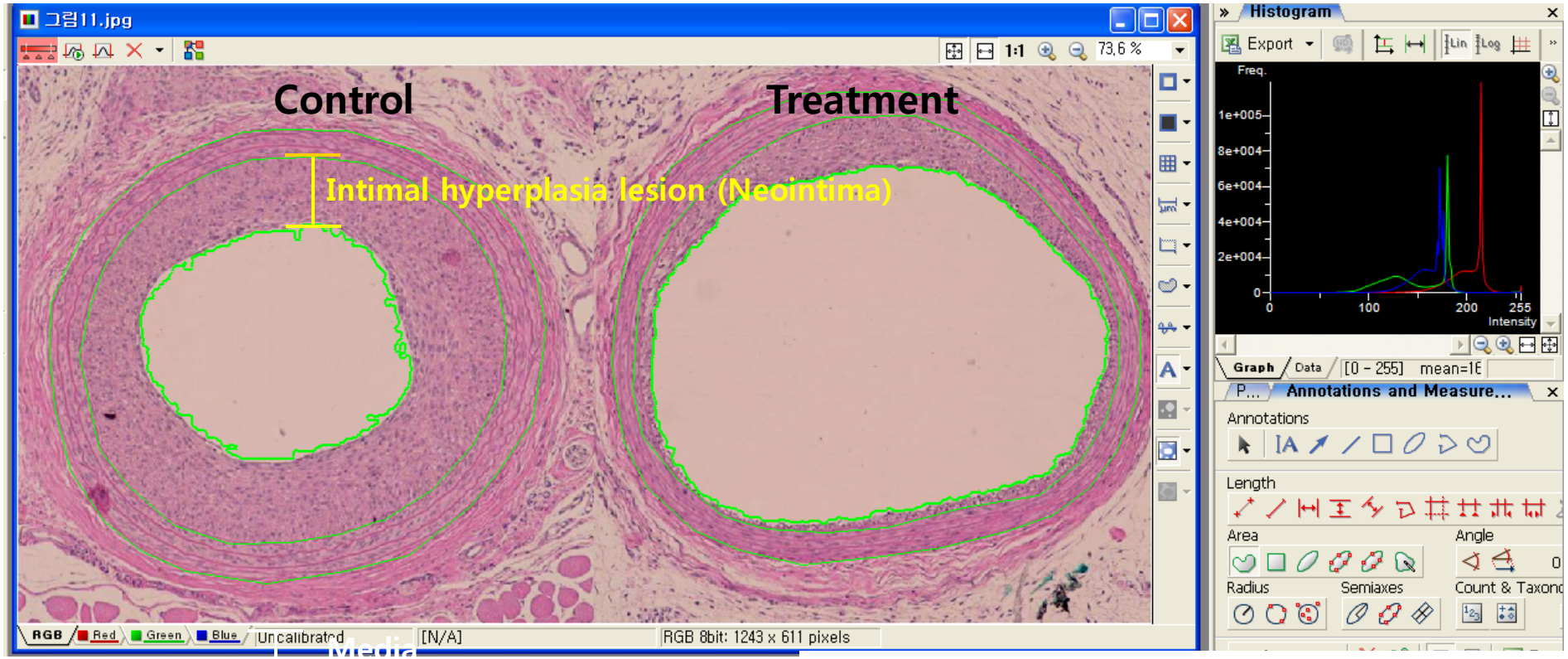


Histomorphometric analysis

-2 weeks later



Neointimal area (mm^2), Stenosis ratio (%),
Lumen area (mm^2),
Thickness, Neointima area/Media area ratio

TABLE 2. Angiographic and Histomorphometric Indices of Oral Paclitaxel Groups

	Paclitaxel Dose, mg/kg			
	0 (n=13)	5 (n=9)	7.5 (n=10)	10 (n=8)
MLD, AU	4.67±1.45	6.28±2.09*	6.97±1.79*	7.97±1.57*
Neointimal area, mm^2	0.13±0.05	0.05±0.05*	0.04±0.03*	0.05±0.03*
Neointima/media area ratio	1.20±0.38	0.46±0.45*	0.39±0.29*	0.40±0.28*
% Area stenosis, %	33.9±8.6	15.2±15.1*	12.1±7.7*	13.5±9.7*
Media area, mm^2	0.11±0.01	0.11±0.02	0.11±0.02	0.11±0.02

Results are reported as mean±SD
*P<0.05 vs control

Kim DW et al. Circulation. 2004;19:242-7

Histomorphometric analysis

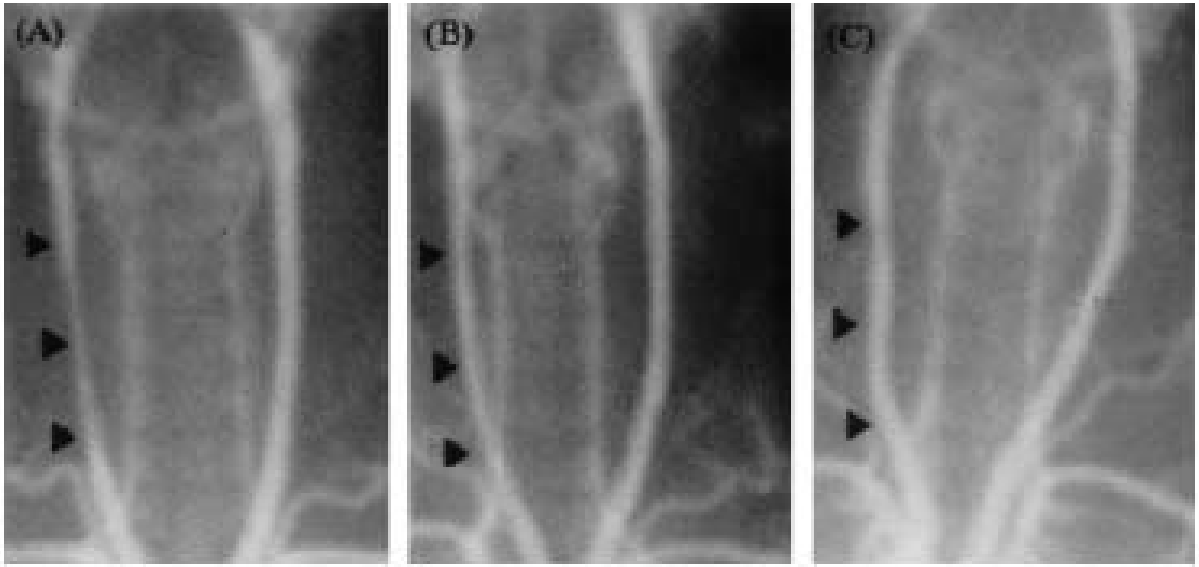
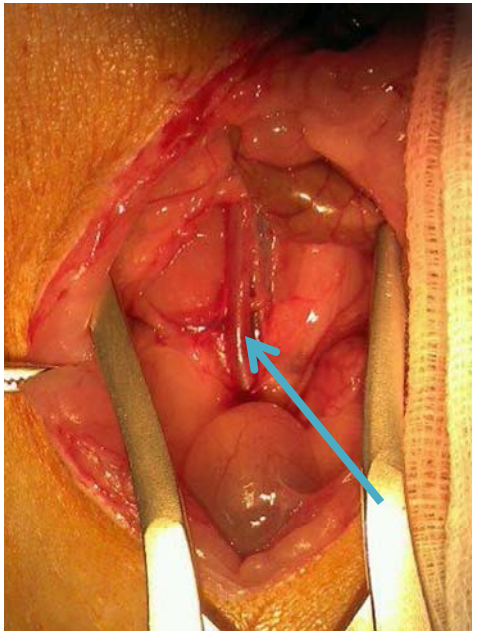


Figure 2. Representative carotid angiographic finding performed 11 days after balloon injury. Diffuse narrowing of right carotid artery was observed in control group. Oral paclitaxel–treated groups showed significant reduction in luminal narrowing of carotid artery compared with control group. A, Control (vehicle alone); B, low-dose paclitaxel (5 mg/kg); and C, high-dose paclitaxel (10 mg/kg).



Abdominal aorta
16G~18 G IV catheter or 7F angio catheter

Kim DW et al. Circulation. 2004;19:242-7

Today

1. Myocardial infarction

(Rat and Mouse: Coronary artery ligation model)

2. Pressure over-load induced LV Hypertrophy

(Transverse aortic constriction model : TAC)

3. Carotid artery balloon injury model

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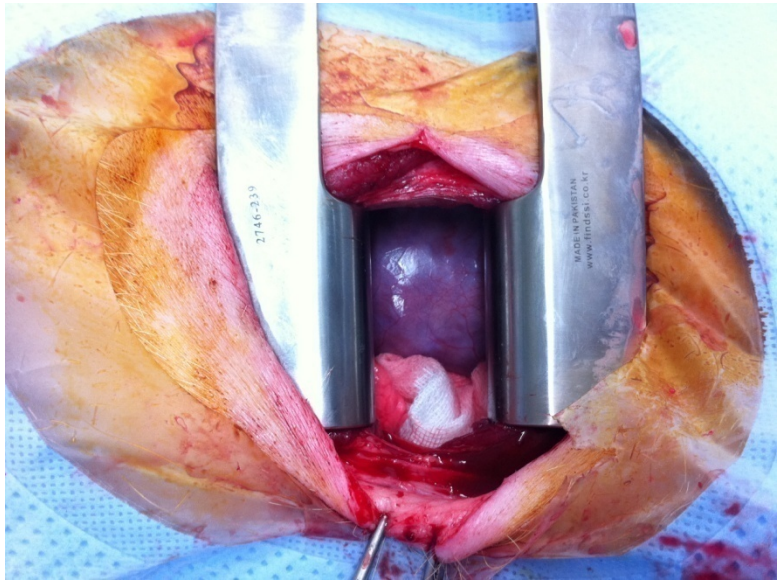
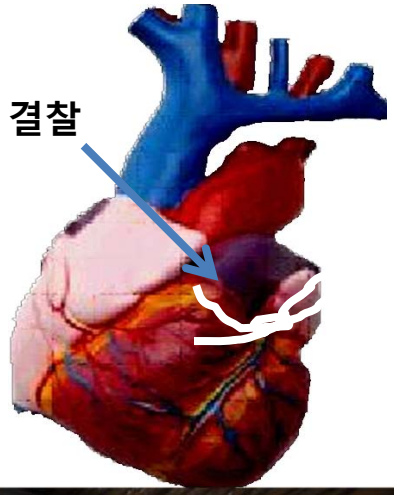
심장질환 치료기술 개발 특성화 센터 (Heart Research Center)

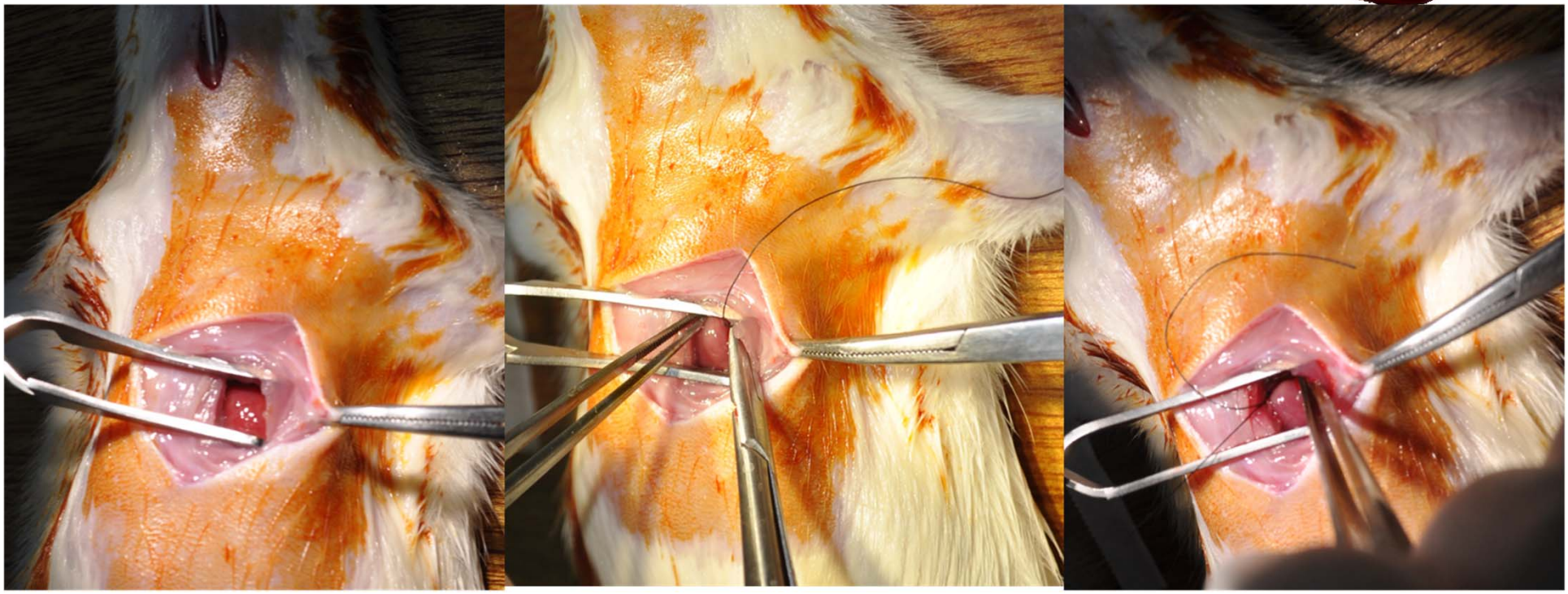
1. Pig coronary artery balloon occlusion model (MI)
2. Pig coronary artery chronic total occlusion model (CTO)
3. Pig coronary artery delivery system
4. Pig thoracotomy model
5. Rat aorta and Rabbit iliac artery stent implantation model
6. Pig Coronary artery stent implantation model
7. Mouse hind limb ischemia model



Rat or Mouse Myocardial Infarction (MI)

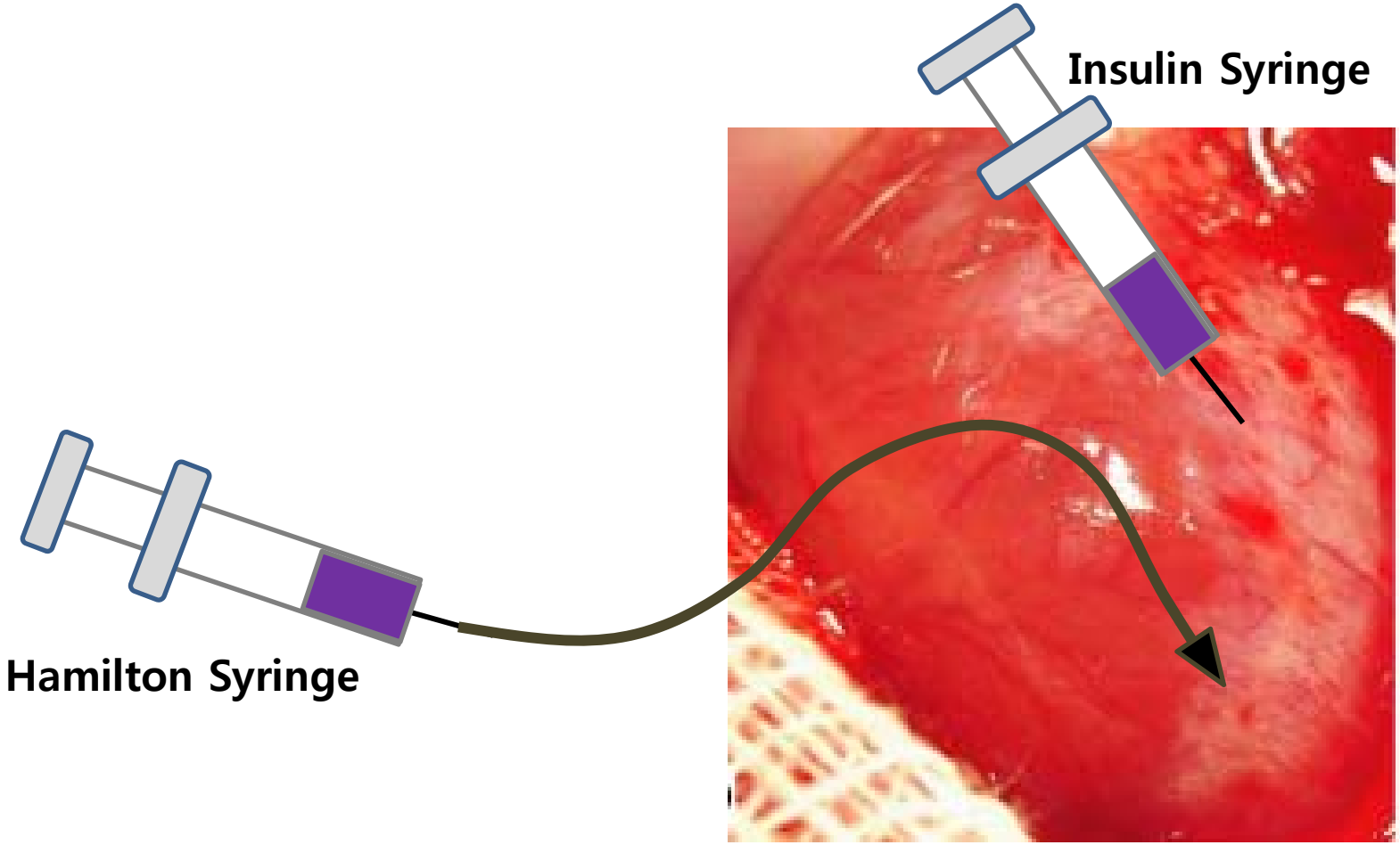


Rat or mouse
Body weight : rat 260±20g (~7 weeks)
: mouse 30±5 g (~12 weeks)



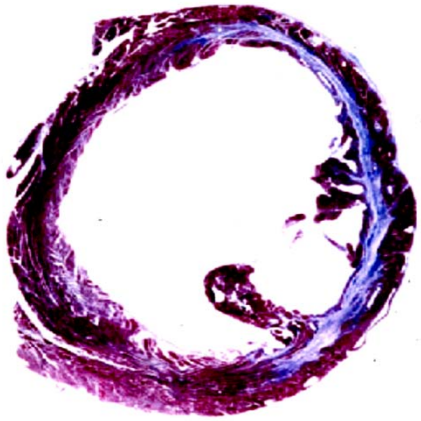
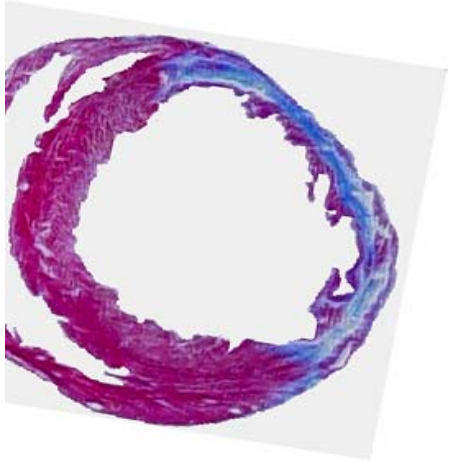
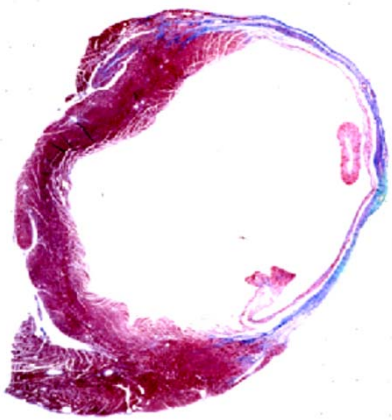
Rat or Mouse Myocardial Infarction (MI)

-Re-opening surgery : cell or plasmid delivery and polymer implantation



Histomorphometric analysis

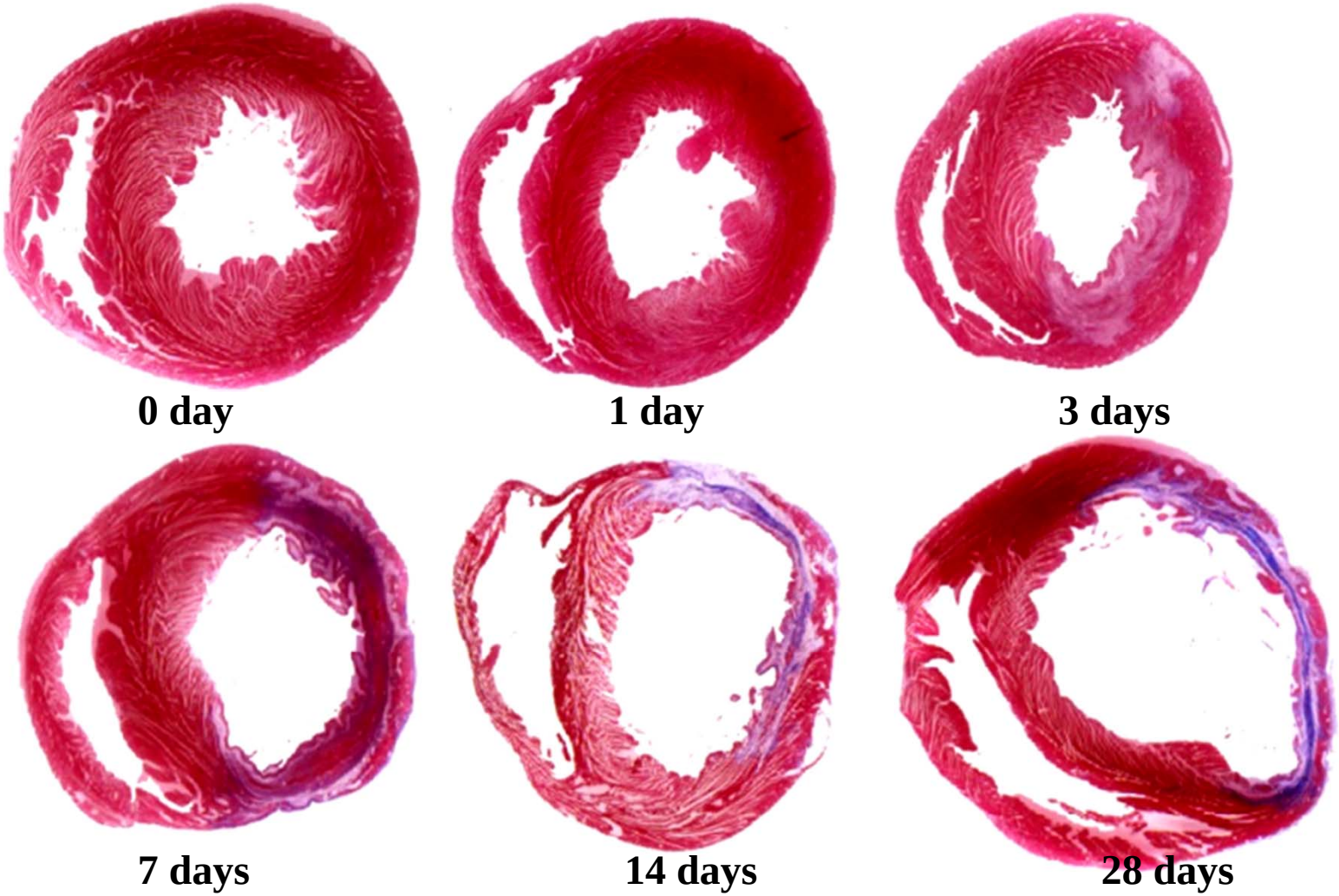
- **Left coronary artery ligation in Sprague-Dawley rats**

ER group (Subendocardial MI)	LR group (Nontransmural MI)	NR group (Transmural MI)
Early reperfusion with myocardial salvage	Late reperfusion without myocardial salvage	Non-reperfusion
45-minute LCA ligation followed by reperfusion	5-hour LCA ligation followed by reperfusion	Permanent LCA ligation
		

From Tae jin Youn (SNU) and Myeong-chan Cho (NIH)

Histomorphometric analysis

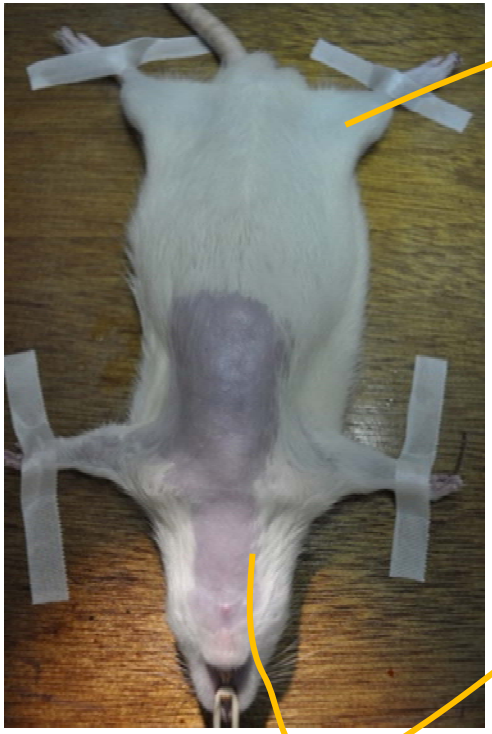
- 4 hours LCA ligation followed by reperfusion (Masson and Trichrome stain or Sirius red stain)

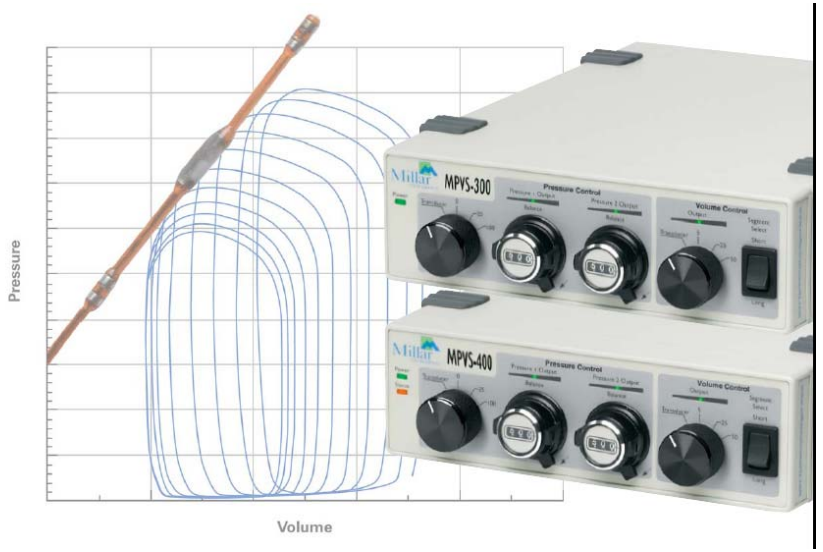


Kwon JS et al. J Mol Cell Cardiol 2011 50:814-

Blood pressure analysis

Femoral artery (Normal blood pressure)





Right common carotid artery (High blood pressure)