

## 白鼠 관동맥 폐쇄와 재관류로 야기된 심근 경색의 크기에 미치는 엔도텔린 길항제들의 역할

서 봉 관 · 황 진 용

= Abstract =

### Effect of Endothelin Antagonists on Myocardial Infarct Size after Coronary Artery Occlusion and Reperfusion in Rat

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**Background :** Although experimental and clinical evidences suggest that endothelin-1(ET-1) may play a pathophysiological role in ischemic heart disease, it is still controversial whether ET-1 produced during myocardial ischemia and reperfusion affects the extent of necrotic myocardium. This study was performed to investigate the role of ET-1 and the effect of ET antagonists in infarct size determination.

**Methods :** Male Wistar rats(260 -400g) were anesthetized with pentobarbital(i.p. 50mg/kg) and ventilation was assisted via tracheostomy tube. The heart was exposed by midline incision and the left anterior descending coronary artery was ligated with 6-0 silk suture. The ligature was released after 1 hour and reperfusion was performed for 2 hours. In the first set of experiment, FR139317(ET-A antagonist) was given as bolus i.v.(3mg/kg) 10 minutes before reperfusion, followed by continuous infusion(total 24mg/kg) throughout reperfusion. In the other protocol, bosentan(ET-A/ET-B antagonist ; 10mg/kg) was given 10 minutes before coronary occlusion as i.v. bolus. At the end of reperfusion, the heart was excised and stained with Evans blue dye(1% w/v) and triphenyltetrazolium chloride(TTC ; 1%) to distinguish infarct region(not stained by TTC and Evans blue), ischemic but viable myocardium(stained brick-red by TTC but not stained by Evans blue) and nonischemic myocardium(dyed by Evans blue). These three regions of myocardium were separated and weighed for analysis. Infarct size(in percent) was expressed as the ratio of infarct region to ischemic myocardium(i.e. infarct region plus ischemic but viable myocardium).

**Results :** In the first protocol, infarct region was  $57.0 \pm 3.8\%$  of the ischemic myocardium in control(n = 9) and  $58.9 \pm 4.9\%$  in FR139317 group(n = 7) ; The difference was not significant statistically. Likewise, ET-A/ET-B antagonist bosentan given before coronary occlusion did not reduce infarct size significantly ; the ratio was  $74.2 \pm 3.2\%$  in control(n = 7) and  $69.5 \pm 2.0\%$  in bosentan group(n = 7).

**KEY WORDS :** Endothelin-1 · Myocardial infarction · Endothelin antagonist · FR139317 · Bosentan · Rat.

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## 연구대상 및 방법

### 1. 연구대상 및 실험방법

Pentobarbital(50mg/kg)  
(male Wistar rat ; 260 400g)  
stroke volume 10ml/kg, 60 (small animal ventilator, Harvard Apparatus Model 683)

(lead II) monitoring  
mon- appendage

6 - 0 2mm  
polyethylene(PE) . PE  
(鉗子)

1 2  
ET 2가 protocol

1) 재관류 직전에 시작하여 재관류 2시간 동안 계속 투여한 ET-A 길항제 FR139317의 효과

FR139317  
10 FR139317 3mg/kg(0.5ml/kg 20%  
DMSO ) bolus  
infusion pump 24mg/kg(3ml/kg  
DMSO ) 2  
(20% DMSO)

( )  
▽  
|-----|10분|-----|  
↑ 1시간 ↑ 2시간  
결찰 재관류

2) 관상동맥 결찰 직전에 투여한 ET-A/ET-B 길항제의 효과

ET  
ET - A/ET - B bosentan  
10mg/kg(1ml/kg )  
10 bolus 1  
2  
(1ml/kg)

▽  
|-----|10분|-----|  
↑ 1시간 ↑ 2 시간  
결찰 재관류

### 2. 심근경색의 크기 측정

가  
plastic cannula . Ca -  
nnula

6 - 0  
cannula Evans blue dye(1% w/v)  
( )

appendage  
2mm 6 7  
1% triphenyltetrazolium chloride(TTC)  
37 ℃ 10 incubation  
TTC

(viable myocardium)  
가 . TTC  
(viable myocardium)

( + )/( + + )  
( + )

( )

### 3. 사용약물

FR139317(Fujisawa, Tokyo, Japan) DMSO

Bosentan(Hoffmann - LaRoche, Basel, Switzerland)

blue dye(Sigma Chemical, USA)

1% w/v, triphenyltetrazolium chloride(Sigma Chemical, USA) phosphate buffered saline 1%

### 4. 통계처리

Student's unpaired t - test P  
0.05 가

## 결 과

5 8  
70%

15 20

Protocol 1. 재관류 직전에 시작, 재관류 2 시간 동안 계속 투여한 ET-A 길항제의 효과

/ (n=9)  
33.6 ± 4.6%, FR139317 (n=7) 31.8 ± 4.4%  
/ 57.0 ± 3.8%, FR139317  
58.9 ± 4.9%  
(Fig. 1).

Protocol 2. 관상동맥 결찰 직전에 투여한 ET-A/ET-B 길항제의 효과

/ (n=7)  
45.5 ± 2.3%, bosentan (n=7) 37.9 ± 3.4%  
/

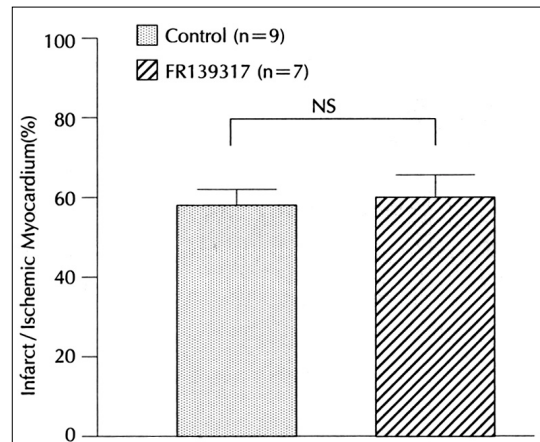


Fig. 1. Effect of FR139317, an ET-A receptor antagonist, on the myocardial infarct size after an episode of 1-hour coronary artery occlusion followed by 2-hour reperfusion. FR139317 was administered as i.v. bolus(3mg/kg) 10minutes before reperfusion, followed by continuous infusion(total 24mg/kg) throughout reperfusion. Data are expressed as mean ± SEM. NS ; not significant.

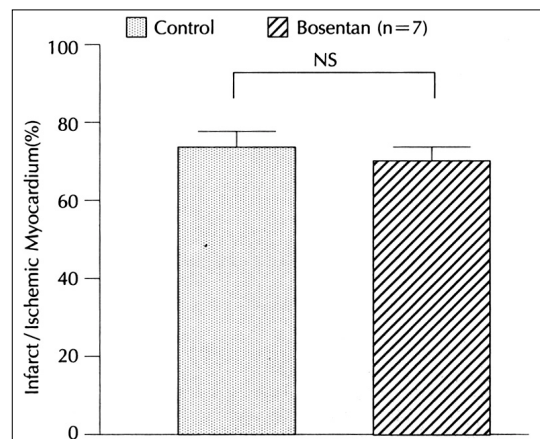


Fig. 2. Effect of bosentan, an ET-A/ET-B receptor antagonist, on the myocardial infarct size after an episode of 1-hour coronary artery occlusion followed by 2-hour reperfusion. Bosentan(10mg/kg) was administered as single i.v. bolus 10min before coronary artery ligation. Data are expressed as mean ± SEM. NS ; not significant.

74.2 ± 3.2%, bosentan  
69.5 ± 2.0%  
가 (Fig. 2).

## 고 안

ET - 3 ET - 1  
 가 ET  
 가 가 Arai<sup>17)</sup>  
 ET - 1 가 ET - 3  
 ET 가, Sakurai<sup>18)</sup> 3  
 가 ET 가  
 - 1(endothelin ; ET) 21 amino acid peptide Yanagisawa  
 1988 ET - A, ET - B  
<sup>26)</sup> ET - A  
<sup>1)</sup> ET - 1 ET peptide  
 가 ET - B  
<sup>1)</sup> ET peptide  
 ET - 1 nitric oxide prostacyclin  
 angiotensin II  
 (tonus)가 가 ( .  
 , , )  
 ET - 1 (saphenous  
<sup>24)</sup> , 3가 isoform(ET - 1, ET - 2, vein)  
<sup>25)</sup> , ET ET ET - A 가 ET - B  
 ET - 3) (ET receptor)  
 peptide (subtype)<sup>26)</sup> , A ET - B 가 ET -  
 ET - 1 big - ET - 1 1 ET - 1 가  
 ET - 1 (endothelin converting enzyme)가  
 cloning<sup>27,28)</sup> 가 가 3), 4),  
<sup>29)</sup> ET  
 ET  
 (ET receptor antagonist) ET - 1  
 ET - 1 가  
 ET bosentan<sup>23)</sup>  
 SB209670<sup>30)</sup> (spontaneous hypertensive rat)  
 ET - 1  
 가 ET - 1  
 ET - 1 가 bosentan  
 ET  
 angiotensin 가  
 ET - 1  
 3) 가 3가 isoform(ET - 1, 2,  
 genomic library  
<sup>25)</sup> ET - 1  
 3가 ET isoform

. , 가 가 - 가 가  
 (ischemia - reperfusion)  
 ET - 1 가 ET ET 가  
 31) 32) 가  
 ET - 1 ET phosphoramidon<sup>33)</sup>, ET -  
 receptor 1 monoclonal antibody  
 binding assay / 125I - ET - 1 11)  
 binding 가 10), , ET  
 / 가 ET - 1 - Grover ET - A  
 8,9) BQ - 123가 - 12)  
 ET - 1 ET - 1 , Krause BQ - 123가  
 가 / 13)  
 ET - A FR139317  
 가 ET 가 14,15)  
 FR139317 가 ET  
 bosentan , bosentan  
 가 (i.v. bolus) TAK - 044 ET - A/ET - B  
 2 ET - A 16,34)  
 FR139317 ET - A FR  
 가 139317  
 ET bosentan  
 Richard 16) ET 가 35)  
 bosentan bosentan 가  
 가 ET bosentan 가 가  
 가 3mg/kg  
 ET 가 16,23) bos -  
 entan bosentan (protocol 2)  
 FR139317 / 가 FR139317  
 (protocol 1) 가 가  
 , bosentan 가 가  
 가 3 (Dr. M. Clozel 私信)  
 bosentan 가 가  
 , protocol 1 /  
 33.6% protocol 2

45.5% 가 . , ET FR139317(ET - A )  
가 protocol 2 protocol 1 10 3mg/kg 2  
가 24mg/kg . bosentan  
가 (ET - A/ET - B ) 10mg/kg 10  
protocol 1 FR139317 .  
DMSO protocol 1 .  
bosentan  
가 . , FR139317 가 ( 57.0 ± 3.8%, n = 9 ; FR139317 58.9 ± 4.9%, n = 7). bosentan ( 74.2 ± 3.2%, n = 7 ; bosentan 69.5 ± 2.0%, n = 7).  
결 과 :  
결 론 :  
ET - A  
ET - A/ET - B 가 ET - A FR139317  
ET - 1 ET - A/ET - B bosentan  
가 가  
ET  
감사의 글 1996  
요 약  
FR139317 Fujisawa  
bosentan Hoffmann - La -  
Roche Dr. Martine Clozel  
연구배경 :  
(endothelin ; ET)  
ET  
가  
ET 가  
방 법 :  
Wistar rat( ; 260 400g) pento - barbital  
(50mg/kg ; )  
, (10ml/kg, 60/min)  
6 - 0 . 1  
2  
Evans blue(1% w/v)  
, triphenyltetrazolium chl -  
oride(1%)  
/ .

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