

신성고혈압쥐에서 Norepinephrine에 의한 혈관수축에 있어 Tyrosine Kinase의 역할

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Role of Tyrosine Kinases in Norepinephrine-Induced Vascular Contraction in Renal Hypertensive Rats

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ABSTRACT

Background and Objectives : Protein tyrosine kinases appear to be involved in the signal transduction mechanisms, which result in vascular smooth muscle contraction, as well those required in cell growth. The present study was conducted to examine the role of tyrosine kinases in the norepinephrine-induced vascular smooth muscle contraction of isolated aortae from two-kidney, one clip (2K1C) hypertensive rats. **Materials and Methods** : 2K1C hypertension was made by clipping the left renal artery of the rats, with age-matched rats receiving a sham treatment serving as controls. Thoracic aortae denuded of endothelium were mounted in tissue baths to measure the isometric tension. **Results** : The putative tyrosine kinase inhibitors, genistein and tyrphostin 25, significantly inhibited the contractile responses of the aorta to norepinephrine in the control rats, but not in the 2K1C rats. The protein tyrosine phosphatase inhibitor, sodium orthovanadate, selectively potentiated the contractile response to norepinephrine, but only in the controls. Genistein, tyrphostin 25 and sodium orthovanadate did not affect KCl-induced vascular contractions in either the 2K1C or the controls. The vascular contraction elicited by phorbol 12, 13 dibutyrate, in the presence and absence of genistein, did not alter in either the 2K1C or the controls. **Conclusion** : These findings indicate that protein tyrosine kinases participate in the norepinephrine-induced contraction of rat aortic smooth muscle, where the role is attenuated in 2K1C renal hypertension. (**Korean Circulation J 2002;32 (10):894-901**)

KEY WORDS : Protein-tyrosine kinase ; Muscle, smooth, vascular ; Norepinephrine ; Hypertension renal.

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서 론
 sine kinase 가
 two - kidney, one clip(2K1C)
 tyrosine kinase
 가
 phospholipase C(PLC)가
 phosphoinositide(PI) 가
 가 가
 protein kinase C(PKC)가
 1)2)
 tyrosine kinase
 3)
 4) tyrosine kinase
 5 - 7)
 norepinephrine
 PI
 8) norepinephrine
 가
 9) norepinephrine tyro-
 sine kinase
 norepinephrine ty-
 rosine kinase
 tyrosine kinase 가 nore-
 pinephrine 가
 가 6)10)11) norepinephrine
 phenylephrine tyrosine ki-
 nase 4)11)
 (hyperplasia)
 (hypertrophy)
 12) (spontaneously hypert-
 ensive rats, SHR)
 13)
 12)
 tyrosine kinase
 가
 norepinephrine tyro-

실험동물
 2K1C
 150 200 g
 (Sprague - Dawley,) thiopental(40 mg/kg,
 IP) 0.2 mm
 silver clip
 1
 (Harvard 10766)
 4
 160 mmHg
 clip
 (sham - clipped)
 혈관의 등장성 장력기록
 2 3 mm
 15 mL
 37 95% O₂ 5% CO₂
 (Grass,
 FT03) Polygraph(Grass,
 Model 79)
 NaCl 118.3, KCl 4.7, NaHCO₃ 25, MgCl₂ 1.2, KH₂
 PO₄ 1.2, CaCl₂ 2.5, glucose 11.1 mM pH
 7.4 2 g
 1.5 2
 40 mM KCl(KCl NaCl
)
 norepinephrine(1 u M)

acetylcholine(10 uM) , ST. Louis) . Genistein tyr-
phostin PDB dimethylsulfoxide(DMSO)
3 no-
repinephrine
ascorbic acid(2 mg/mL) 가 .
DMSO 0.04% .

분석 및 통계
± norepi-
nephrine 40 mM KCl
% EC₅₀ pD₂ (the mean
of the negative log molar concentration)
ANOVA(analysis of variance)
unpaired t - test p<0.05

phostin Biomol (genistein ty-
, Plymouth
Meeting) Sigma (

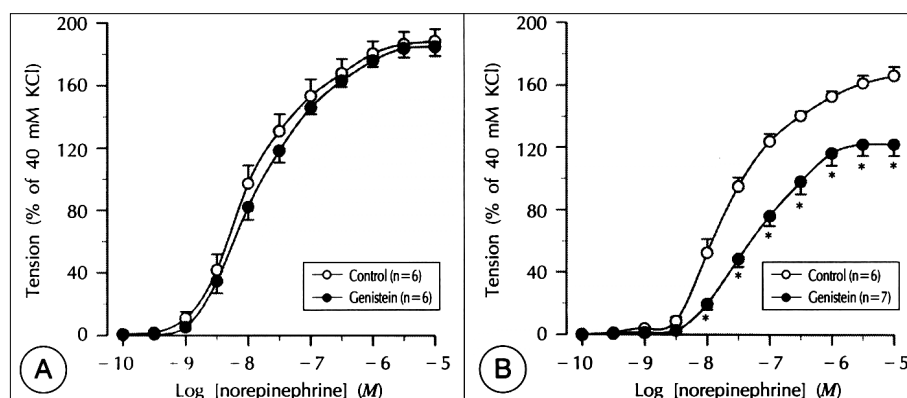


Fig. 1. Effects of genistein on concentration-response curves for norepinephrine in isolated aorta from (A) 2K1C and (B) sham-clipped rats. * : p<0.05, compared with corresponding control values. n : indicates number of rings.

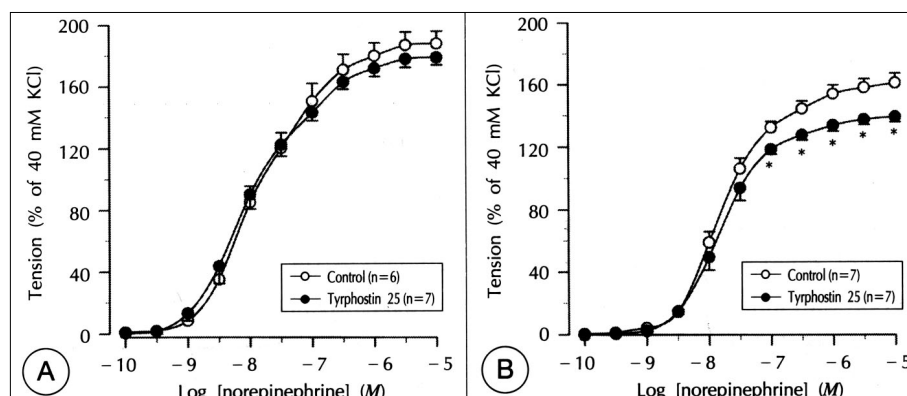


Fig. 2. Effects of tyrphostin 25 on concentration-response curves for norepinephrine in isolated aorta from (A) 2K1C and (B) sham-clipped rats. * : p<0.05, compared with corresponding control values.

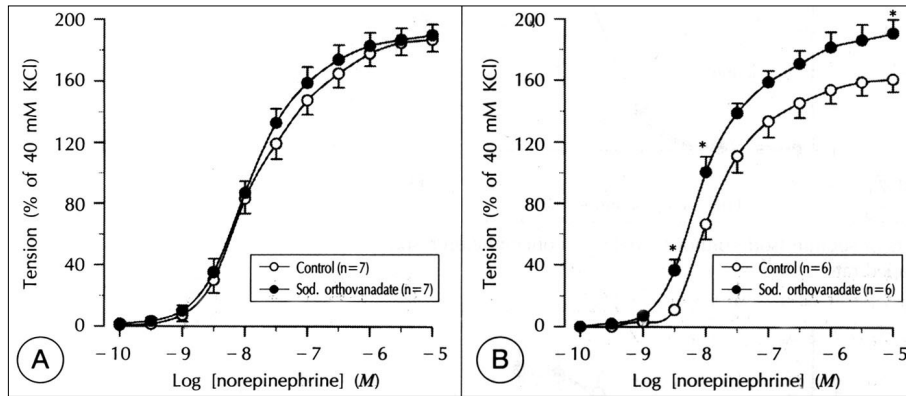


Fig. 3. Effects of sodium (sod.) orthovanadate on concentration-response curves for norepinephrine in isolated aorta from (A) 2K1C and (B) sham-clipped rats. * : $p < 0.05$, compared with corresponding control values.

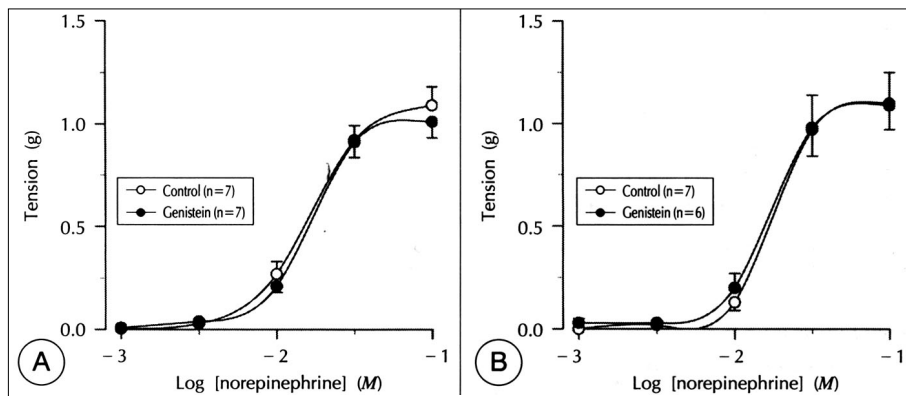


Fig. 4. Effects of genistein on concentration-response curves for KCl in isolated aorta from (A) 2K1C and (B) sham-clipped rats.

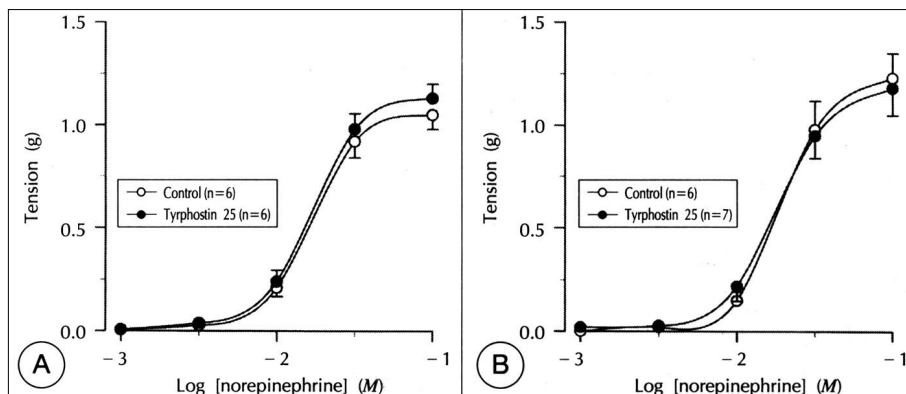


Fig. 5. Effects of tyrphostin 25 on concentration-response curves for KCl in isolated aorta from (A) 2K1C and (B) sham-clipped rats.

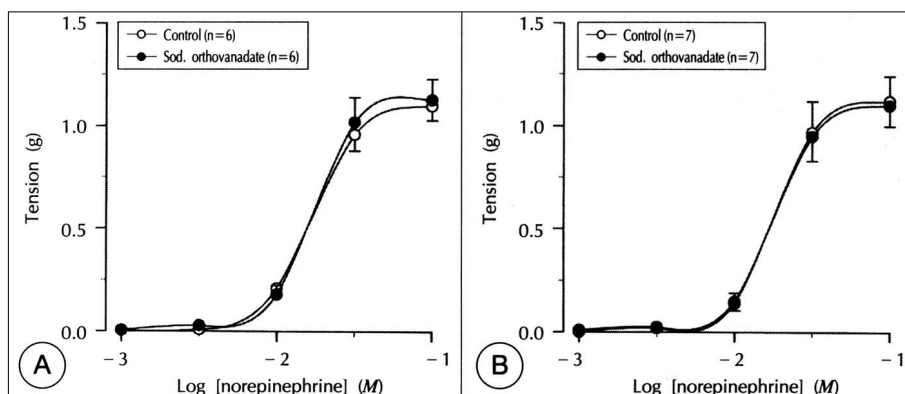


Fig. 6. Effects of sodium (sod.) orthovanadate on concentration-response curves for KCl in isolated aorta from (A) 2K1C and (B) sham-clipped rats.

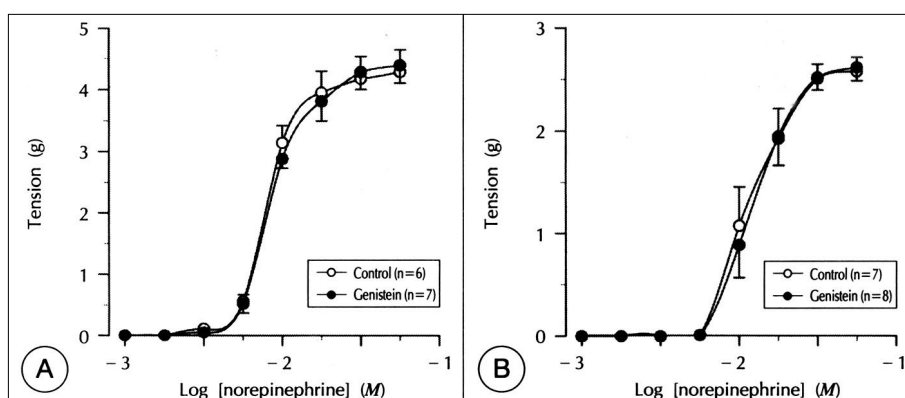


Fig. 7. Effects of genistein on concentration-response curves for phorbol 12, 13 dibutyrate (PDB) in isolated aorta from (A) 2K1C and (B) sham-clipped rats.

(968 ± 25 mg)	가	2K1C	norepinephrine	가 21.3 ± 4.5%
Norepinephrine	2K1C			
가	18.5 ± 2.7%	(Fig. 3).	KCl	2K1C
Norepinephrine	tyrosine kinase			
genistein	2K1C			
	norepinephrine 10 ⁻⁸ M		(1.10 ± 0.08 g)	(1.08 ± 0.05 g)
가 29.2 ± 3.8%	(Fig. 1).		Genistein	tyrphostin 25
tyrosine kinase	tyrphostin 25		ium orthovanadate	2K1C
2K1C	norepinephrine		KCl	(Fig. 4 - 6).
	10 ⁻⁷ M		PKC	PDB
	genistein		가 63 ± 8.2%	가 Genistein 2K1C
(Fig. 2). Tyrosine phosphatase	sodium orthovanadate		PDB	(Fig. 7).

Table 1. Effects of genistein, tyrphostin 25 and sodium orthovanadate on NE, KCl and PDB-induced contractions in isolated aorta from sham-clipped and two-kidney, one clip (2K1C) rats

Treatment	Sham-clipped	2K1C
NE (control)	7.60±0.12	7.86±0.12
Genistein+NE	7.24±0.07*†	7.79±0.09
NE (control)	7.73±0.09	7.87±0.13
Tyrphostin 25+NE	7.72±0.08	7.89±0.08
NE (control)	7.77±0.10	7.81±0.11
Sod. orthovanadate+NE	7.92±0.07	7.83±0.12
KCl (control)	1.74±0.02	1.82±0.05
Genistein+KCl	1.81±0.05	1.80±0.02
KCl (control)	1.74±0.03	1.76±0.04
Tyrphostin 25+KCl	1.76±0.04	1.78±0.03
KCl (control)	1.76±0.06	1.75±0.02
Sod. orthovanadate+KCl	1.75±0.05	1.76±0.05
PDB (control)	7.85±0.13	8.13±0.09
Genistein+PDB	7.80±0.10	8.08±0.11

Data are mean ± SE of pD₂ values for six to eight separate experiments. pD₂ denotes EC₅₀ values which are expressed as the negative log molar concentration. NE : norepinephrine, Sod : sodium, PDB : phorbol 12, 13 dibutyrate, * : p<0.05, † : p<0.05, compared with corresponding control or 2K1C values, respectively.

2K1C
EC₅₀ Table 1
norepinephrine
genistein
EC₅₀
(Table 1).
고 찰
norepinephrine
PLC PI 가
8)
norepinephrine
tyrosine
kinase 14)15) genistein
tyrphostin 25 nor-
epinephrine tyro-
sine kinase
genistein ATP
14) tyrphostin tyrosine kinase
가 (pseudosubstrate) 15)

tyrosine kinase
norepinephrine genistein ty-
rphostin EC₅₀
genistein 10)
가 norepinephrine
tyrosine phosphatase tyrosine ki-
nase 16)17) sod-
ium orthovanadate ty-
rosine kinase norepinephrine
tyrosine kinase (specificity)
phostin genistein tyr-
myosin light chain kinase⁶⁾
protein kinase A⁶⁾¹⁴⁾¹⁵⁾ PKC¹⁴⁾¹⁵⁾
tyrosine kinase
ge-
nistein 2K1C PKC
PDB
sodium orthovanadate
tyrosine phosphatase
16 - 18)
genistein tyrphostin 25 sodium orthov-
anadate가 KCl
2K1C
norepinephrine
tyrosine kinase
Norepinephrine
tyrosine kinase 10)
6)
ifeddine 11) tyrosine kinase Sa-
tyrphostin norepinephrine genistein
10) Saif-
eddine 11) tyrosine kinase 10
Abebe 10)
가 30

가

12)

tyrosine kinase

13)

가 24)

2K1C

12)

가

2K1C

가

hrine

25)

tyrosine kinase

norepinep-

tyrosine kinase

19)

norepinephrine

PDB

EC₅₀

2K1C

가

가

20)

deoxycorticosterone acetate

21)22)

(DOCA) - salt

PI

20)21)

PKC

2K1C

가

가

가

가

KCl

가

2K1C

23)

rosine kinase

2K1C

ty-

tyrosine kinase

norepinephrine

tyrosine kinase

요 약

배경 및 목적 :

Norepinephrine

tyrosine kinase

가

tyrosine kinase

two - kidney, one clip(2K1C)

가

방 법 :

clip

norepinephrine

tyrosine kinase

genistein

tyrphostin 25

tyrosine phosphatase

sodium orthovanadate

ty-

angio-

Malloy 19)

tyrphostin 25

tyrphostin 25

가

tyrphostin

24)

genistein

serotonin

가 DOCA -

22)

salt

2K1C

genistein

tyrpho-

Sodium or-

thovanadate norepinephrine 2K1C
 . Genistein tyrphostin 25 so-
 dium orthovanadate 2K1C
 KCl . Protein kin-
 ase C(PKC) phorbol 12,13 - dibutyrate
 (PDB) 2K1C ge-
 nistein .

결 론 :

norepinephrine
 tyrosine kinase
 2K1C .

중심 단어 : ; ;
 ; .

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