

회주 하대정맥에서 퓨린수용체활성화에 의한 혈관반응

1 2  
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## Vascular Reactivity by Purinoceptor Activation in Rat Inferior Vena Cava

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## ABSTRACT

**Background :** Extracellular ATP, released from platelets and nerve endings, plays significant roles in the regulation of circulation. The effects of ATP depend on the location of the vessels and the species of experimental animals. Until now, studies were limited to arteries, so we compared the effects of ATP in rat vena cava with those in the aorta and attempted to identify the characteristics of their receptors. **Methods :** Vascular rings were isolated from the rat inferior vena cava and descending thoracic aorta. Endothelial cells were preserved or removed by gentle rubbing. The isometric contractions were recorded on polygraph using a force transducer. **Results :** In the vena cava ring precontracted by 100 nM norepinephrine (NE), ATP elicited relaxations in a dose-dependent manner. These effects were abolished by removal of the endothelium or pretreatment with a nitric oxide synthase inhibitor. Relaxations to ATP in the vena cava ( $EC_{50}$  : 9.9  $\mu$ M) were less potent than those in the aorta (1.7  $\mu$ M). The relative order of potencies was ADP > ATP > AMP > adenosine, but the maximal relaxation to ADP was smaller than to ATP. ATP-induced vasorelaxation was blocked by suramin, a nonselective antagonist for P<sub>2</sub> purinoceptor and reactive blue-2, a P<sub>2Y</sub> blocker. At basal tension, ATP contracted the vena cava dose-dependently and these effects were potentiated by endothelium-removal. Contractions in the vena cava were also less potent than in the aorta, and the order of potencies was , -MeATP > UTP > ATP > ADP > AMP = adenosine. ATP-induced vasoconstriction was blocked by suramin and , -MeATP, a desensitizing antagonist of P<sub>2X</sub> purinoceptor, and potentiated by pretreatment with UTP. **Conclusion :** These results suggest that ADP and ATP acts on P<sub>2Y1</sub>- and P<sub>2Y2</sub>-purinoceptor in the endothelium, respectively and induces vasorelaxation of the vena cava, which is mediated by nitric oxide. Since ATP and UTP induced vasoconstriction in endothelium-denuded condition, it may be mediated by the activation of the P<sub>2X</sub> and P<sub>2Y4,6</sub> purinoceptor on smooth muscles, respectively.

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**KEY WORDS :** Adenosine 5'-triphosphate (ATP) · , -methylene adenosine 5'-triphosphate (, -MeATP) · Inferior vena cava · Purinoceptor · Vasorelaxation · Vasoconstriction.

서 룰

1)2)

3)4)

5)

6)

adenine	adenosine
: 2000 1 10	
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: (0371) 741 - 0291 .	: (0371) 745 - 6461 E - mail : jwlee@wonju.yonsei.ac.kr

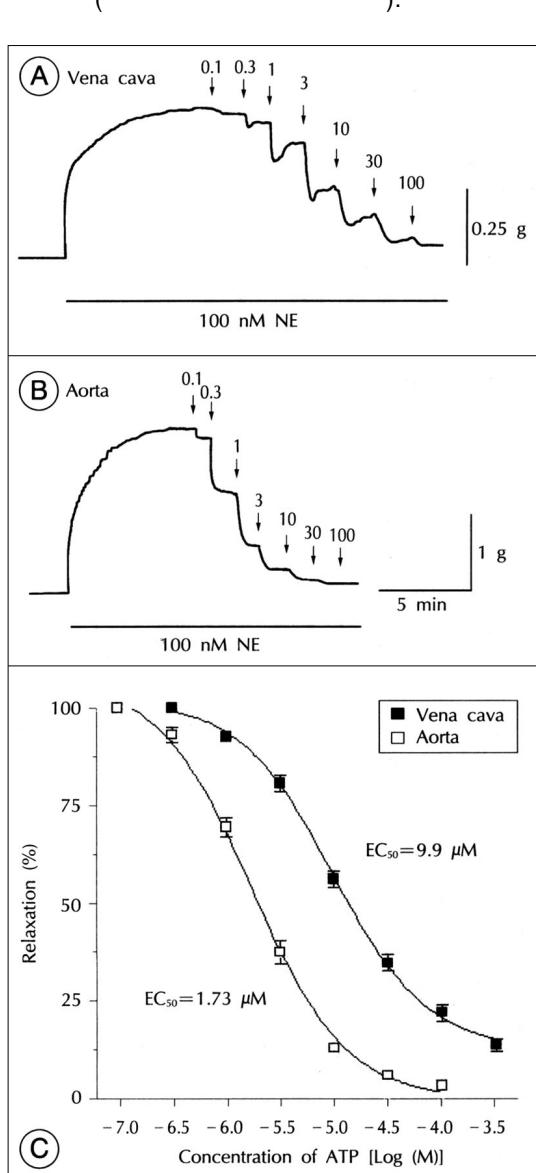
enosine , ad - CaCl<sub>2</sub> 1.91, KH<sub>2</sub>PO<sub>4</sub> 1.19, MgSO<sub>4</sub> 1.44, NaHCO<sub>3</sub>  
 P<sub>2</sub> P<sub>1</sub> ATP 24.8 glucose 5.5 mM , 95% O<sub>2</sub> - 5% CO<sub>2</sub>  
 P<sub>1</sub> A<sub>1</sub>, A<sub>2</sub>, A<sub>3</sub> , P<sub>2</sub> P<sub>2X1-7</sub> P<sub>2Y1-11</sub> pH 7.4 . Ad-  
 9)10) enosine(AD), adenosine 5' - monophosphate(AMP),  
 ATP adenine adenosine 5' - diphosphate(ADP), adenosine 5' - triphosphate(ATP), -methylene adenosine 5' - triphosphate( - MeATP), uridine 5' - triphosphate(UTP), theophylline, reactive blue - 2(RB - 2), N - nitro - L - arginine methylester(L - NAME), acetylcholine(ACh) norepinephrine(NE) Sigma  
 P<sub>2X</sub> , 11 - 13) suramin RBI (Research Bi-  
 ATP P<sub>2Y</sub> ochemicals International, MA, USA)  
 가 , Adenosine theophylline DMSO ,  
 P<sub>2Y</sub> 1 N NaOH pH가 7.0  
 20

8)15)16) 대정맥 또는 대동맥 적출 및 장력측정  
 clin nitric oxide(NO) sodium thiopental(50 mg/kg)  
 , prostacy - heparin(500 IU/kg)  
 17)18)

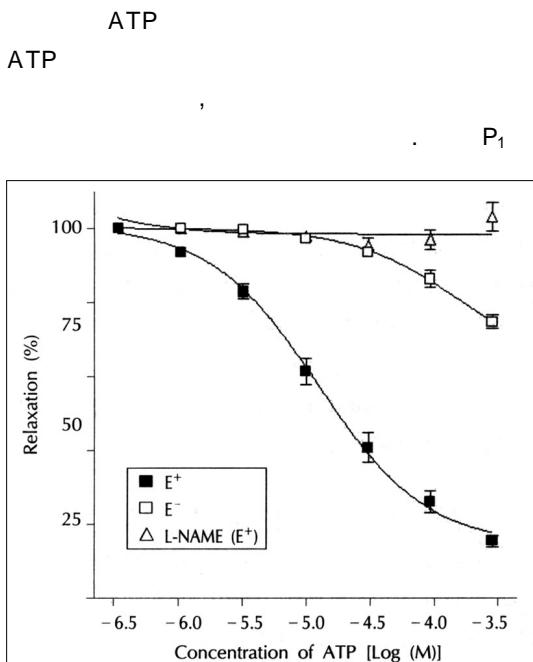
adenine ATP 5 mm KRB  
 adenine , , , , , , , , , , , ,  
 , , , , , , , , , , , , , , , ,  
 대상 및 방법 KRB organ bath L  
 대상 및 방법 가 stainless steel  
 실험 동물 force transducer(Grass FT03C)  
 300 gm (Sprague - (Grass Model 7E) . KRB 37  
 Dawley rat) . (95% O<sub>2</sub> - 5% CO<sub>2</sub>)  
 , , 0.5 gm,  
 실험용액 및 약물 1 gm , 60  
 Krebs - Ringer bicarbonate(KRB) KRB  
 , NaCl 117, KCl 4.7, 3 4 , Furchgott

M ACh	80%	<sup>19)</sup> 100 nM NE (E <sup>+</sup> ) , (E <sup>-</sup> )	가 1 가 가	ATP
<b>자료 분석</b>				
<b>퓨린수용체 활성화에 의한 장력변화 측정</b>				
NE		100 nM , ATP, ADP,	Student paired t - test	p 0.05
AMP, AD		100 nM 1 mM	가	결 과
NE 정맥 및 동맥에서 ATP에 의한 혈관이완반응				
50% (EC <sub>50</sub> )		100 nM NE ATP KCl(40 mM)	Fig. 1A B 100 nM NE 가 , ATP	가 ,
NO	L - NAME(1 mM)	NO가 ,	EC <sub>50</sub> (9.9 μM) 가 (1.7 μM) 30 μM , 300 μM	100% 가 80% (Fig. 1C).
<b>퓨린수용체 차단제에 의한 영향</b>				
ATP		내피세포 제거 및 NO 생성 차단에 의한 영향		
, 100 μM ATP	가	NE P <sub>1</sub> theophylline(50 μM), P <sub>2</sub> suramin(1 mM), P <sub>2X</sub> μM), <sup>20)21)</sup> P <sub>2Y</sub> 20 ,	ATP , 30 μM L - NAME	가
UTP(50 μM)		- MeATP(100 RB - 2(10 μM) ATP P <sub>2Y2</sub> /P <sub>2Y4</sub> ATP ADP(EC <sub>50</sub> ; 0.52 μM)	NO (Fig. 2).	
(desensitization) <sup>22)</sup>		NE adenosine 30 nM 가 ADP(EC <sub>50</sub> ; 0.52 μM)	ATP, ADP, AMP 1 mM AMP(153 M)	
ATP		2 mM ATP	ATP	

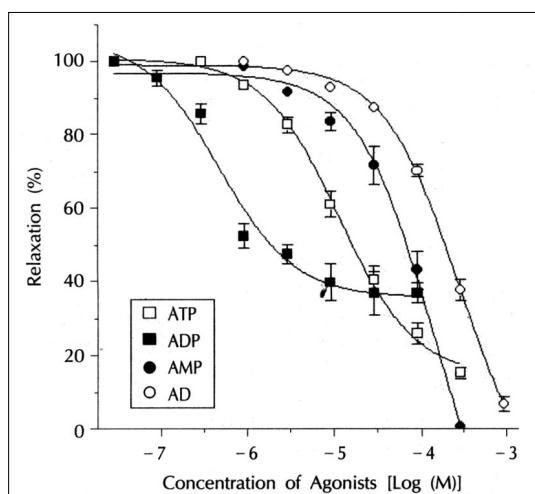
adenosine(309  $\mu$ M)  
ATP(9.9  $\mu$ M)  
(Fig. 3). UTP



**Fig. 1.** Comparison of ATP-induced vasorelaxation in NE-precontracted rat inferior vena cava and aorta. Following precontraction with NE (100 nM), vasorelaxation was induced by cumulative application of ATP (0.1  $\mu$ M - 100  $\mu$ M) in inferior vena cava (a) and aorta (b) with intact endothelium. (c) Concentration-response curves represent relaxation (%) from NE-induced contraction. Data are presented as mean  $\pm$  SEM from six experiments.



**Fig. 2.** Effects of ATP on NE-precontracted rat inferior vena cava with or without endothelium in presence of L-NAME. Following precontraction with NE (100 nM), vasorelaxation was induced by cumulative application of ATP (0.1  $\mu$ M - 100  $\mu$ M). Concentration-response curves represent relaxation (%) from NE-induced contraction. Data are presented as mean  $\pm$  SEM from eight experiments.

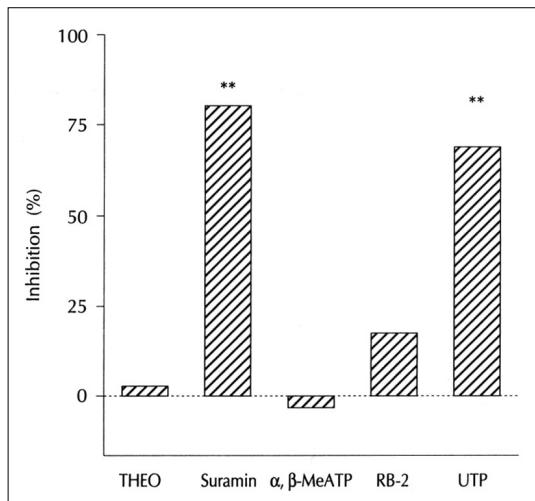


**Fig. 3.** Effects of adenine nucleotides and adenosine on NE-precontracted rat inferior vena cava with intact endothelium. Following precontraction with NE (100 nM), vasorelaxation was induced by cumulative application of either adenine nucleotides or adenosine (0.03  $\mu$ M - 1000  $\mu$ M). Concentration-response curves represent relaxation (%) in NE-induced contraction. Data are presented as mean  $\pm$  SEM from six experiments.

theophylline(50  $\mu$ M) ATP  
 $(2.8 \pm 1.9\%), P_2$   
suramin(1 mM)  $P_{2Y2}$   
 $(80.0 \pm 2.0\%)$ .  
/P<sub>2Y4</sub> UTP(50  $\mu$ M)  $P_{2Y}$   
RB-2(10  $\mu$ M) 69.6  $\pm$  8.0%  
18.0  $\pm$  3.0%  $P_{2X}$   
, - MeATP(100  $\mu$ M)  
(- 2.9  $\pm$  9.2%) (Fig. 4).

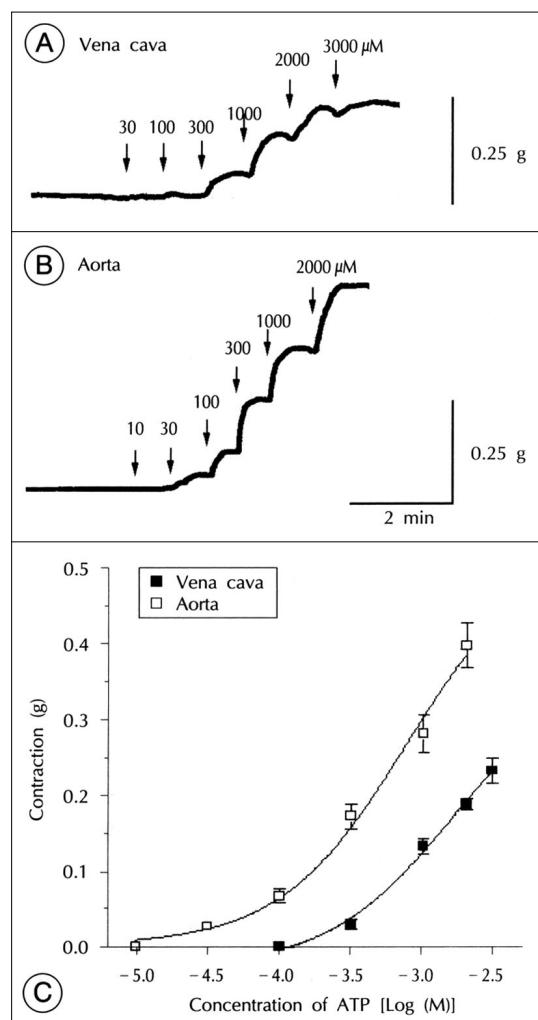
정맥 및 동맥에서 ATP에 의한 혈관수축반응  
ATP 10  $\mu$ M  
M 3 mM  
Fig. 5 A B

,  
(Fig. 5). ATP  
, 가  
NO 가  
가 (Fig. 6).

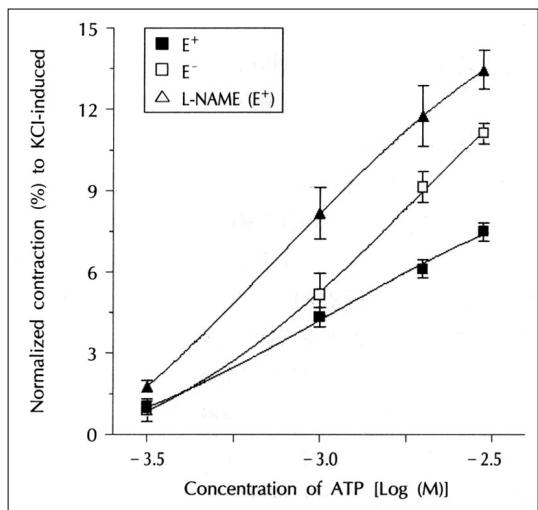


**Fig. 4.** Effects of purinergic agonists and antagonists on ATP-induced relaxations in NE-precontracted rat inferior vena cava with intact endothelium. Following pretreatment with theophylline (THEO, 50  $\mu$ M), suramin (1 mM), RB-2 (10  $\mu$ M), -MeATP (100  $\mu$ M) or UTP (50  $\mu$ M) for 20 min, ATP (100 nM)-induced relaxation (%) was acquired in NE (100 nM)-precontracted preparation. The value below zero indicates more contraction than control. Data are presented as mean  $\pm$  SEM from six experiments and denotes  $p < 0.01$ .

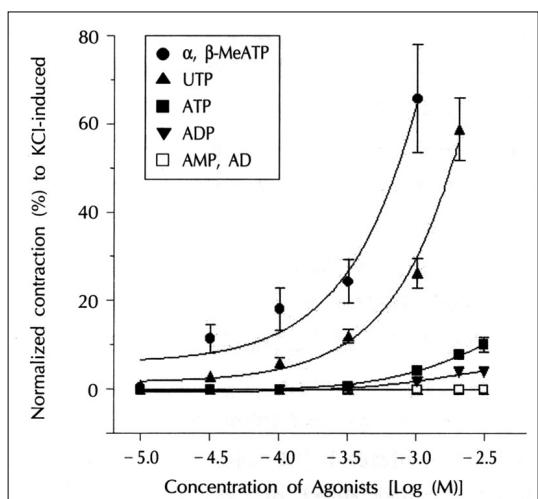
여러 뉴클레오티드 및 adenosine에 의한 정맥 수축반응  
, - MeATP, UTP, ATP,  
ADP, AMP adenosine 10  $\mu$ M 3 mM  
가  
, - MeATP 가  
UTP, ATP, ADP , AMP  
adenosine  
ATP  
(Fig. 7).



**Fig. 5.** Comparison of ATP-induced vasoconstriction in rat inferior vena cava and aorta as basal tension. Vasoconstriction was induced by cumulative application of ATP (10  $\mu$ M - 3 mM) in inferior vena cava (a) and aorta (b) with intact endothelium. (c) Cumulative concentration-response curves represent changes in vascular tension. Data are presented as mean  $\pm$  SEM from five experiments.

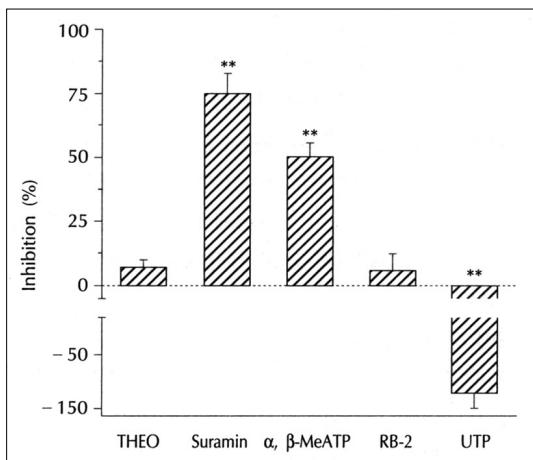


**Fig. 6.** Comparison of ATP-induced contraction in rat inferior vena cava with or without endothelium in presence of L-NAME. ATP (300  $\mu$ M - 2 mM) was added cumulatively in inferior vena cava. Contraction (%) was normalized to the maximal tension developed by high KCl (40 mM). Data are presented as mean  $\pm$  SEM from five experiments.



**Fig. 7.** Effects of adenine nucleotides and adenosine on basal tension of rat inferior vena cava. Contraction was induced by a cumulative application of different purinergic receptor agonists ( $\alpha$ ,  $\beta$ -MeATP, UTP, adenine nucleotides and adenosine (10  $\mu$ M - 3 mM)) from endotheliumdenuded preparation. Contraction (%) was normalized to the maximal tension developed by high KCl (40 mM). Data are presented as mean  $\pm$  SEM from six experiments.

ophylline (50  $\mu$ M) (5.4  $\pm$  2.9%), suramin(1 mM ; 74.1  $\pm$  7.4%) , - Me -



**Fig. 8.** Effects of purinergic agonists and antagonists on ATP-induced contraction in rat inferior vena cava. Following pretreatment with theophylline (THEO, 50  $\mu$ M), suramin (1 mM), RB-2 (10  $\mu$ M),  $\alpha$ ,  $\beta$ -MeATP (100  $\mu$ M) and UTP (50  $\mu$ M) for 20 min., then ATP (100  $\mu$ M) was applied to endotheliumdenuded preparation. Result represents as percentage changes of ATP-induced contraction. The value below zero indicates a further contraction than those in the absence of purinergic agonists. Data are presented as mean  $\pm$  SEM from six experiments and \*\* denotes  $p < 0.01$ .

ATP (100  $\mu$ M ; 48.7  $\pm$  3.1%)  
 RB-2(10  $\mu$ M) ATP  
 (9.3  $\pm$  7.5%), UTP  
 가 (- 127.7  $\pm$  25.5%) (Fig. 8).

고 안  
 ATP norepinephrine(NE)  
 vesicle ATP↑ 600 mM  
 23 - 25)

13)26) ATP↑  
 ,  
 ATP  
 가  
 14)27)28)

ATP

NO가	35(36)
endothelium derived relaxing factor(EDRF) ATP가	P <sub>2Y</sub>
EDRF , EDRF가 oxide(NO) cyclooxygenase prostacyclin prostacyclin L - NAME	(agonist sensitivity) EDRF ATP ADP nitric NO UTP 18)31) P <sub>2Y2</sub> /P <sub>2Y4</sub> , NO UTP <sup>22)</sup> ATP ATP 가
NO Adenine ADP AMP adenosine ATP AMP adenosine ATP P <sub>1</sub>	ADP P <sub>2Y1</sub> ATP UTP P <sub>2Y2</sub> 가 60% ATP adenosine P <sub>1</sub> ( ). Adenosine NO synthase inhibitor 15) 가
P <sub>1</sub> P <sub>2</sub> , -methylene adenosine 5' - triphosphate( - MeATP) <sup>20)21)32)</sup> reactive blue - 2 <sup>33)</sup> UTP <sup>22)34)</sup>	theophylline suramin ATP , ATP 가 P <sub>2</sub> . P <sub>2</sub> P <sub>2X</sub> NO 가 가 ATP , P <sub>2Y</sub> 18.0%, ATP 69.6%가 P <sub>2Y</sub> IP <sub>3</sub> adenosine Ca <sup>2+</sup> 38) ATP

theophylline

RB - 2<sup>33)</sup> P<sub>2Y1</sub> P<sub>2Y1</sub>

$P_2$		suramin
$P_{2X}$		
,	- MeATP <sup>20)(21)</sup>	
가	.	ATP
,	- MeATP	
UTP	ATP	가
.	.	$P_2$
		ATP
$P_{2X}$		,
		UTP
	ATP	
$P_{2Y2}$	uridine	
$P_{2Y4}$	$P_{2Y6}$	
.	UTP	ATP
가	가	

가 가  
 ATP shock  
 가 ,  
 39)  
 adenosine 40)  
 ATP  
 가  
 ATP

요약

### 연구목적 :

adenosine 5' - triphosphate(ATP)

중심 단어 : Adenosine 5' - triphosphate (ATP) . ,  
 - methylene adenosine 5' - triphosphate ( , , -  
 MeATP) . . . . .

ATP

가

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