

CHAPTER III

RESULTS



In Anaesthetized Rats:

Experiment 1. The Effects of ALKALOID on Systemic Blood Pressure and Heart Rate

The actions of ALKALOID were investigated in four groups of anaesthetized rats. An intravenous infusion of ALKALOID (slow bolus injection) caused dose-dependent decreases in both systolic and diastolic blood pressures. The higher doses, the longer depression was observed. As shown in Fig. 3, at the doses of 0.4, 0.8 and 1.6 mg/kg B.W. both systolic and diastolic blood pressures were similarly depressed. At the highest dose 3.2 mg/kg B.W., most of the results have shown a more depression in diastolic than systolic blood pressure. According to paired t-test, the decreases of blood pressure by ALKALOID were significantly different from control ($P < 0.001$). Collection of systolic, diastolic and mean blood pressures at each dose are shown in Table 1, 2 and 3 respectively and summarized in Fig. 4.

An intravenous infusion of ALKALOID elicited biphasic responses on heart rate. The responses were shown initial reduction followed by increase. This changes were significantly seen in higher doses (Table 4).

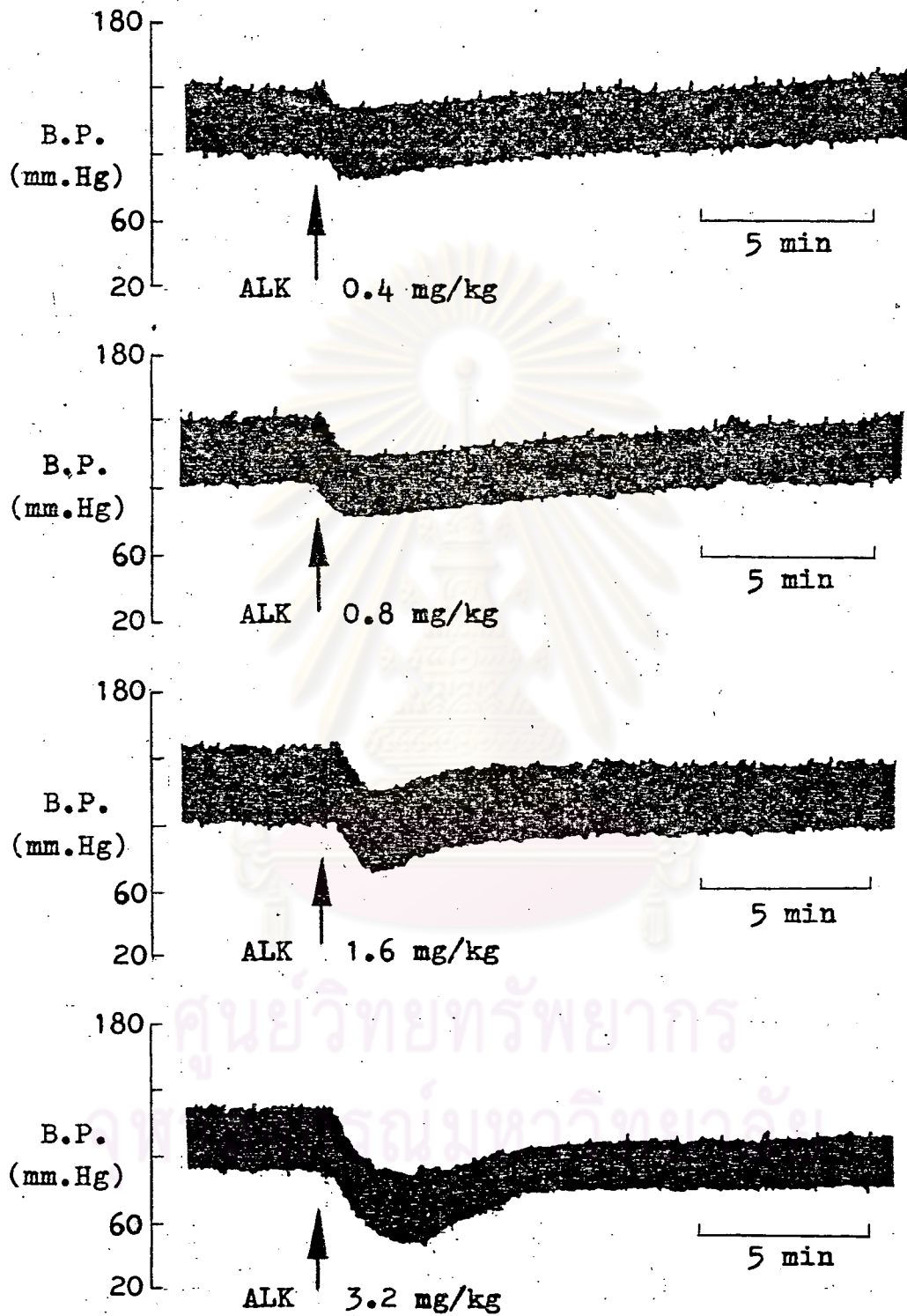


Figure 3. Records showing the effects of ALKALOID (ALK) in various doses on blood pressure in anaesthetized rats.

DOSE OF ALKALOID (mg/kg)	NO. OF RAT	SYSTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
0.4	11	150.91 ± 4.21	129.82 ± 3.52	13.77 ± 1.76 ***
0.8	14	146.86 ± 3.29	121.29 ± 3.29	17.36 ± 1.57 ***
1.6	12	145.50 ± 3.18	118.33 ± 2.67	18.41 ± 1.33 ***
3.2	14	153.57 ± 2.87	116.00 ± 2.49	24.42 ± 1.09 ***

Table 1. The effects of ALKALOID on systolic blood pressure in anaesthetized rats (mean ± S.E.M.).

*** P < 0.001, paired t-test.

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

DOSE OF ALKALOID (mg/kg)	NO. OF RAT	DIASTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
0.4	11	102.73 ± 3.93	83.46 ± 4.01	18.73 ± 2.55 ^{***}
0.8	14	106.71 ± 2.54	82.71 ± 3.72	23.20 ± 2.28 ^{***}
1.6	12	105.33 ± 1.48	76.17 ± 1.93	27.63 ± 1.73 ^{***}
3.2	14	105.43 ± 3.61	67.29 ± 3.80	36.72 ± 2.02 ^{***}

Table 2. The effects of ALKALOID on diastolic blood pressure in anaesthetized rats (mean ± S.E.M.).

*** P < 0.001, paired t-test

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

DOSE OF ALKALOID (mg/kg)	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
0.4	11	118.79 ± 3.77	98.01 ± 3.61	16.62 ± 2.07 ^{***}
0.8	14	120.57 ± 2.76	95.57 ± 3.36	20.79 ± 1.87 ^{***}
1.6	12	118.83 ± 1.78	90.22 ± 1.71	24.00 ± 1.31 ^{***}
3.2	14	121.48 ± 3.20	83.52 ± 3.02	31.39 ± 1.27 ^{***}

Table 3. The effects of ALKALOID on mean blood pressure in anaesthetized rats (mean ± S.E.M.). *** P < 0.001, paired t-test.

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

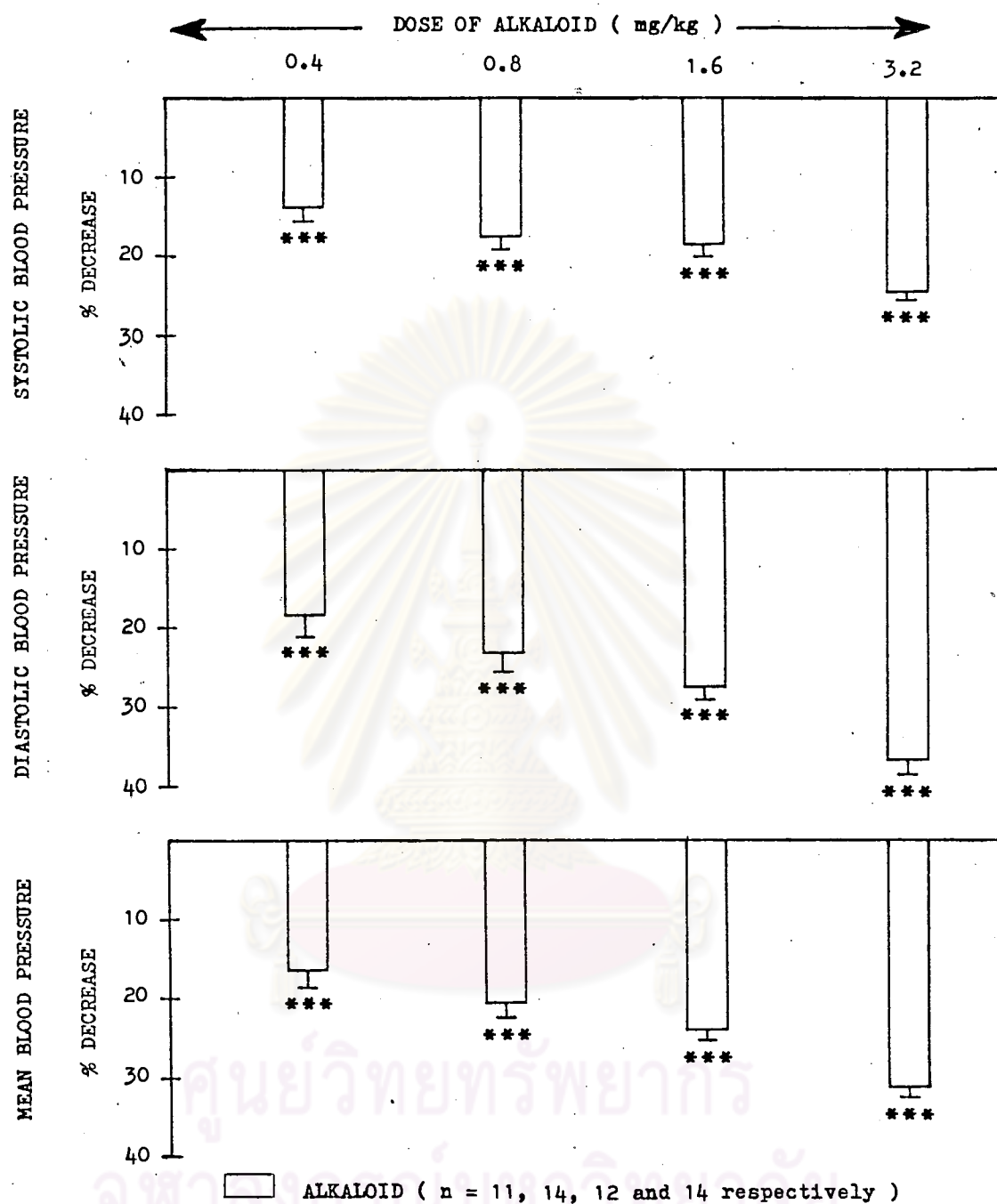


Figure 4. The effects of ALKALOID on blood pressures in anaesthetized rats. Bar graphs represent mean percent decrease + S.E.M. The ordinate scales are percent decrease in blood pressure; the abscissa scale is dose of ALKALOID in mg/kg B.W. *** $P < 0.001$, paired t-test

DOSE OF ALKALOID (mg/kg)	NO. OF RAT	HEART RATE (beats/min)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	
			★ INITIAL	● MAXIMUM
0.4	10	339.00 ± 12.43	337.00 ± 12.31	340.00 ± 12.57
0.8	11	340.91 ± 12.17	336.36 ± 11.99 *	349.00 ± 11.48 *
1.6	10	341.00 ± 9.94	336.00 ± 9.69 *	347.00 ± 10.33 *
3.2	13	347.69 ± 9.55	340.77 ± 10.33 *	353.08 ± 9.09 *

★ INITIAL = at initial hypotensive effect of ALKALOID

● MAXIMUM = at maximum hypotensive effect of ALKALOID

Table 4. The effects of ALKALOID on heart rate in anaesthetized rats (mean ± S.E.M.).

* Significant decrease from control, $P < 0.05$, paired t-test

* Significant increase from control, $P < 0.05$, paired t-test.

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

Experiment 2. The Effects of Beta-Adrenergic Blocking Drug (Propranolol) on the Hypotensive Effect of ALKALOID

Isoproterenol 1.0 $\mu\text{g}/\text{kg}$ B.W. decreased mean blood pressure 32.41 ± 5.43 percent, whilst ALKALOID 3.2 mg/kg B.W. decreased 31.39 ± 1.27 percent. This hypotensive effect of isoproterenol was completely blocked by propranolol 1.0 mg/kg B.W. (Fig. 5). Comparison of the effects of isoproterenol on mean blood pressure before and after propranolol blockade are shown in Table 5.

As shown in Fig. 6, administration of propranolol 1.0 mg/kg B.W. 5 min before ALKALOID could not significantly reduce the hypotensive effect of ALKALOID ($P > 0.05$, unpaired t-test). By the observation, some of the initial reduction in blood pressure at the doses of 1.6 and 3.2 mg/kg B.W. showed a shorter duration of the peak hypotension than the effect of ALKALOID alone.

Again as shown in Fig. 7, increasing the dose of propranolol to 2.0 mg/kg B.W. could not reduce the hypotensive effect of ALKALOID significantly. However, the sustained reduction of blood pressure by ALKALOID was observed at all doses. Summarization of the effects of ALKALOID on systolic, diastolic and mean blood pressures are illustrated in Table 6, 7, 8 and Fig. 8.

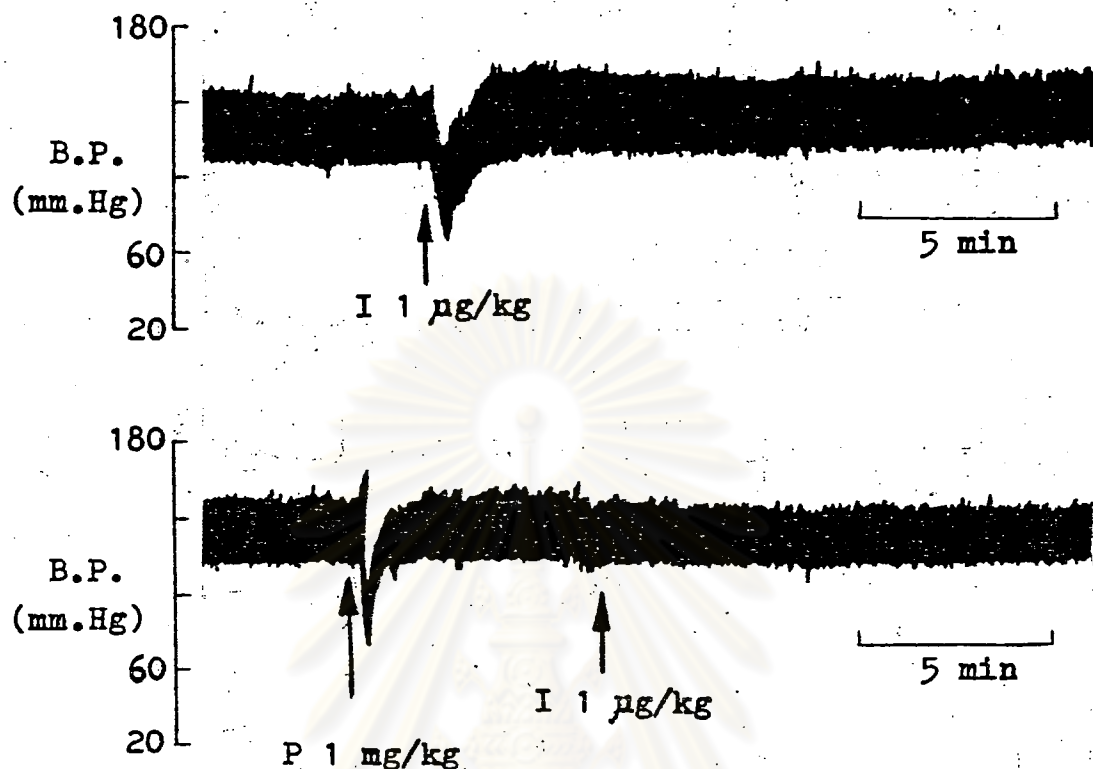


Figure 5. Records showing the effects of isoproterenol (I) 1 $\mu\text{g}/\text{kg}$ on blood pressure before and after propranolol (P) 1 mg/kg blockade for 5 min in anaesthetized rats.

SUBSTANCE	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ISOPROTERENOL INFUSION	AFTER ISOPROTERENOL INFUSION	% DECREASE
ISOPROTERENOL 1.0 $\mu\text{g}/\text{kg}$	4	120.67 \pm 4.13	82.00 \pm 9.02	32.41 \pm 5.43
ISOPROTERENOL 1.0 $\mu\text{g}/\text{kg}$ AFTER PROPRANOLOL 1.0 mg/kg	4	118.84 \pm 4.44 (122.17 \pm 4.17)	118.84 \pm 4.44	0.00 \pm 0.00 ***

Table 5. Comparison of the effects of isoproterenol on mean blood pressure before and after propranolol blockade in anaesthetized rats (mean \pm S.E.M.). Number in parenthesis represents mean blood pressure before propranolol administration. *** $P < 0.001$, unpaired t-test

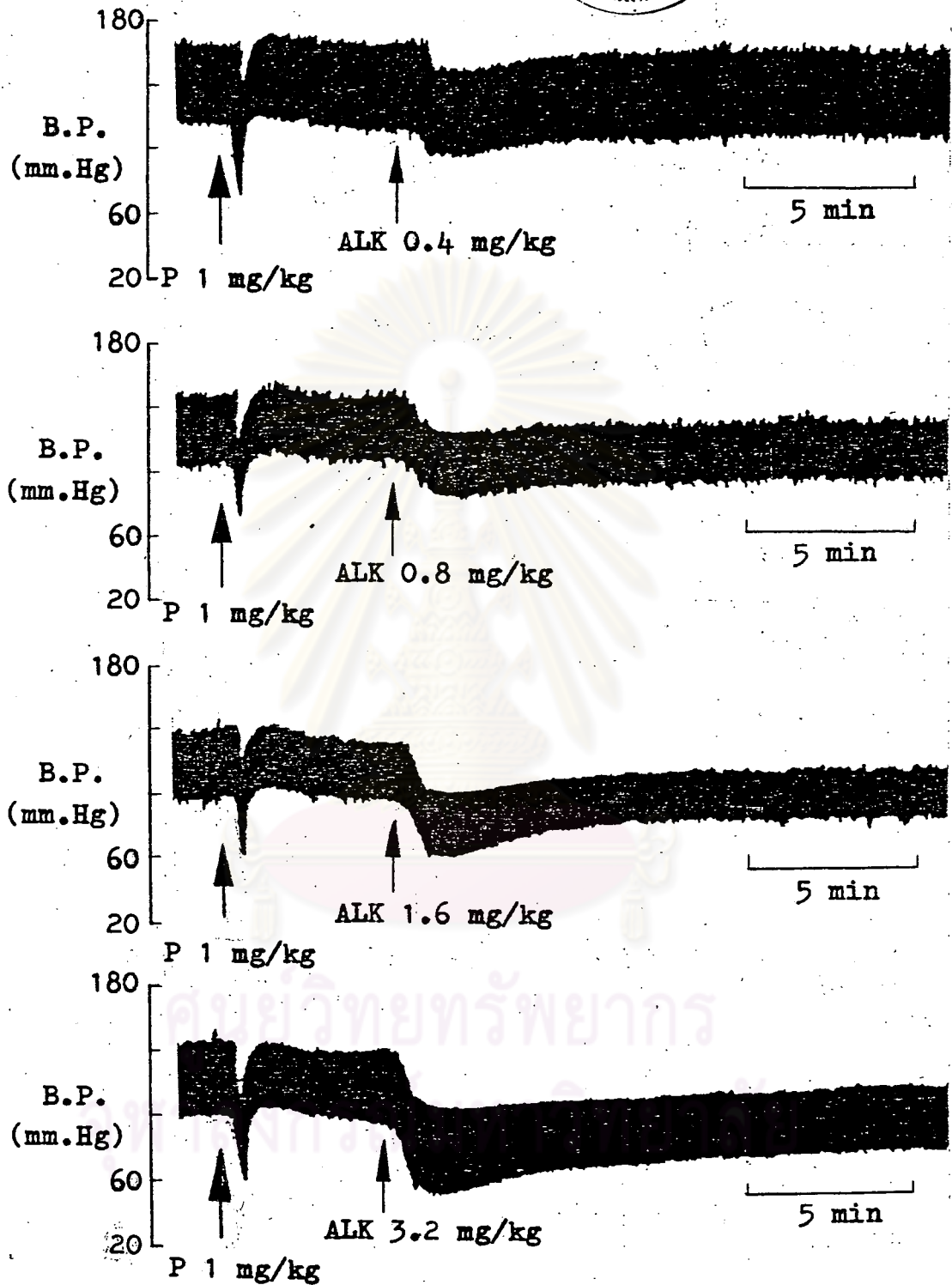


Figure 6. Records showing the effects of ALKALOID (ALK) in various doses on blood pressure after propranolol (P) 1 mg/kg blockade for 5 min in anaesthetized rats.

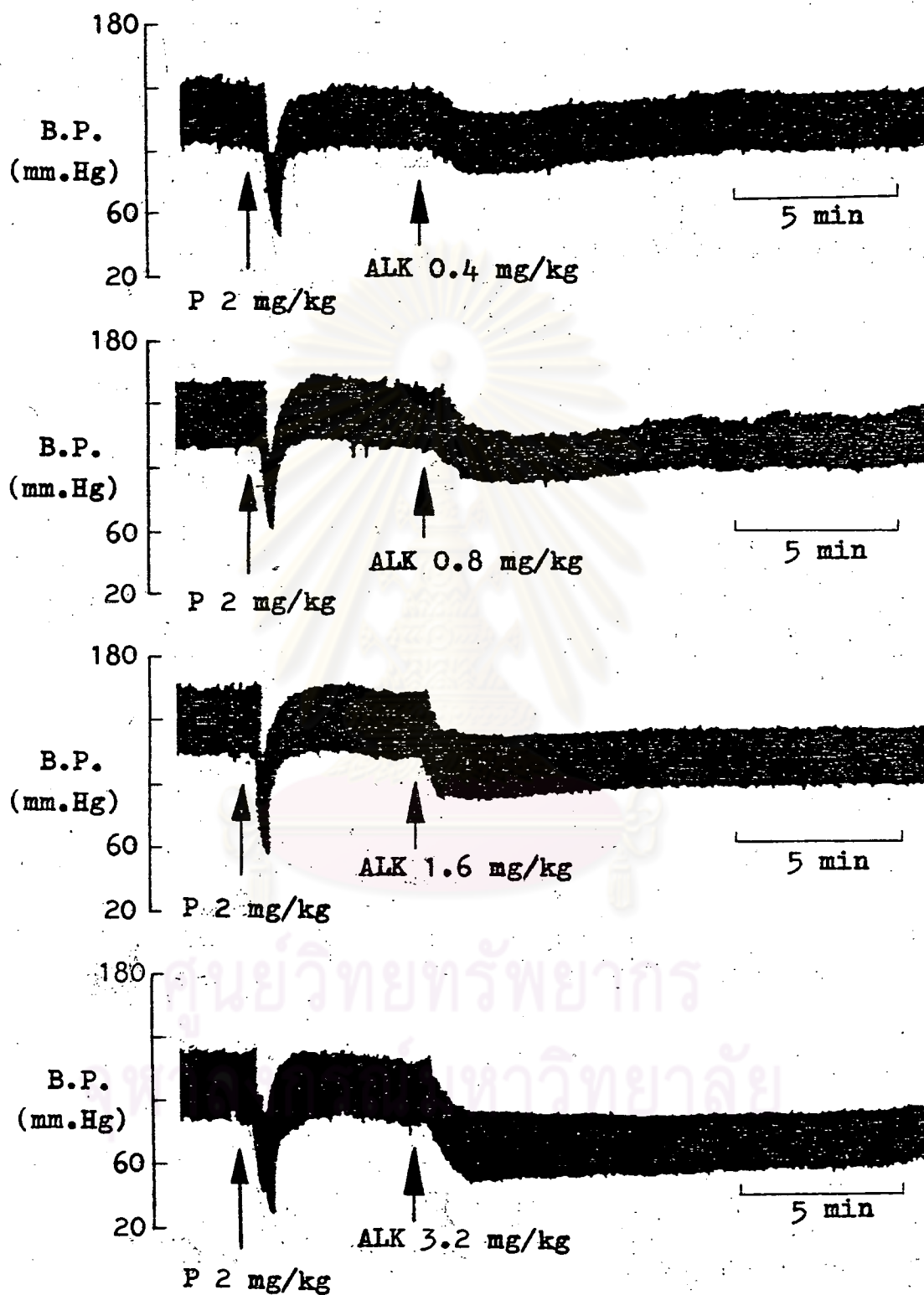


Figure 7. Records showing the effects of ALKALOID (ALK) in various doses on blood pressure after propranolol (P) 2 mg/kg blockade for 5 min in anaesthetized rats.

SUBSTANCE (mg/kg)	NO. OF RAT	SYSTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	150.91 ± 4.21	129.82 ± 3.52	13.77 ± 1.76
ALKALOID 0.4 AFTER PROPRANOLOL 1.0	9	141.56 ± 4.11 (144.89 ± 3.71)	124.44 ± 3.12	11.98 ± 0.86
ALKALOID 0.4 AFTER PROPRANOLOL 2.0	6	134.33 ± 4.30 (141.00 ± 3.17)	120.33 ± 3.44	10.34 ± 0.98
ALKALOID 0.8	14	146.86 ± 3.29	121.29 ± 3.29	17.36 ± 1.57
ALKALOID 0.8 AFTER PROPRANOLOL 1.0	7	140.29 ± 1.41 (145.43 ± 2.65)	116.29 ± 2.24	17.12 ± 1.19
ALKALOID 0.8 AFTER PROPRANOLOL 2.0	6	147.33 ± 3.75 (157.00 ± 3.79)	120.67 ± 3.49	16.90 ± 1.32
ALKALOID 1.6	12	145.50 ± 3.18	118.33 ± 2.67	18.41 ± 1.33
ALKALOID 1.6 AFTER PROPRANOLOL 1.0	7	144.29 ± 6.60 (144.57 ± 5.10)	115.43 ± 5.73	20.16 ± 1.00
ALKALOID 1.6 AFTER PROPRANOLOL 2.0	6	145.33 ± 2.62 (151.33 ± 2.04)	118.00 ± 2.68	18.77 ± 1.68
ALKALOID 3.2	14	153.57 ± 2.87	116.00 ± 2.49	24.42 ± 1.09
ALKALOID 3.2 AFTER PROPRANOLOL 1.0	6	136.00 ± 6.61 (144.67 ± 4.15)	106.67 ± 2.86	21.11 ± 2.08
ALKALOID 3.2 AFTER PROPRANOLOL 2.0	6	146.00 ± 6.85 (147.00 ± 4.52)	114.33 ± 4.69	21.53 ± 1.69

Table 6. Comparison of the effects of ALKALOID on systolic blood pressure before and after propranolol blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent systolic blood pressure before propranolol administration.

SUBSTANCE (mg/kg)	NO. OF RAT	DIASTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	102.73 ± 3.93	83.46 ± 4.01	18.73 ± 2.55
ALKALOID 0.4 AFTER PROPRANOLOL 1.0	9	101.33 ± 3.54 (106.67 ± 3.38)	85.56 ± 3.07	15.53 ± 1.03
ALKALOID 0.4 AFTER PROPRANOLOL 2.0	6	96.00 ± 4.84 (101.33 ± 3.49)	82.33 ± 4.01	14.51 ± 1.11
ALKALOID 0.8	14	106.71 ± 2.54	82.71 ± 3.72	23.20 ± 2.28
ALKALOID 0.8 AFTER PROPRANOLOL 1.0	7	100.29 ± 1.82 (105.43 ± 1.29)	76.00 ± 2.72	24.17 ± 2.56
ALKALOID 0.8 AFTER PROPRANOLOL 2.0	6	104.33 ± 4.39 (109.33 ± 3.37)	81.00 ± 5.88	22.81 ± 2.68
ALKALOID 1.6	12	105.33 ± 1.48	76.17 ± 1.93	27.63 ± 1.73
ALKALOID 1.6 AFTER PROPRANOLOL 1.0	7	103.71 ± 4.56 (106.29 ± 3.36)	80.00 ± 4.59	23.09 ± 1.79
ALKALOID 1.6 AFTER PROPRANOLOL 2.0	6	102.67 ± 1.43 (109.00 ± 1.23)	80.00 ± 3.76	23.35 ± 3.33
ALKALOID 3.2	14	105.43 ± 3.61	67.29 ± 3.80	36.72 ± 2.02
ALKALOID 3.2 AFTER PROPRANOLOL 1.0	6	97.33 ± 6.80 (101.67 ± 5.07)	63.00 ± 4.37	35.22 ± 1.36
ALKALOID 3.2 AFTER PROPRANOLOL 2.0	6	102.00 ± 5.32 (106.67 ± 3.67)	70.33 ± 7.16	31.84 ± 3.84

Table 7. Comparison of the effects of ALKALOID on diastolic blood pressure before and after propranolol blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent diastolic blood pressure before propranolol administration.

SUBSTANCE (mg/kg)	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	118.79 ± 3.77	98.01 ± 3.61	16.62 ± 2.07
ALKALOID 0.4 AFTER PROPRANOLOL 1.0	9	114.74 ± 3.70 (119.41 ± 3.38)	98.52 ± 2.99	14.07 ± 0.87
ALKALOID 0.4 AFTER PROPRANOLOL 2.0	6	108.78 ± 4.64 (114.55 ± 3.25)	95.00 ± 3.77	12.58 ± 1.01
ALKALOID 0.8	14	120.57 ± 2.76	95.57 ± 3.36	20.79 ± 1.87
ALKALOID 0.8 AFTER PROPRANOLOL 1.0	7	113.62 ± 1.50 (118.76 ± 1.16)	89.43 ± 2.26	21.26 ± 1.87
ALKALOID 0.8 AFTER PROPRANOLOL 2.0	6	118.67 ± 4.15 (125.22 ± 3.32)	94.78 ± 4.78	20.31 ± 1.60
ALKALOID 1.6	12	118.83 ± 1.78	90.22 ± 1.71	24.00 ± 1.31
ALKALOID 1.6 AFTER PROPRANOLOL 1.0	7	117.24 ± 5.21 (119.05 ± 3.79)	91.81 ± 4.95	21.87 ± 1.41
ALKALOID 1.6 AFTER PROPRANOLOL 2.0	6	118.00 ± 1.95 (123.11 ± 1.80)	92.67 ± 3.35	21.47 ± 2.59
ALKALOID 3.2	14	121.48 ± 3.20	83.52 ± 3.02	31.39 ± 1.27
ALKALOID 3.2 AFTER PROPRANOLOL 1.0	6	110.22 ± 6.73 (116.00 ± 4.62)	77.56 ± 3.82	29.35 ± 1.38
ALKALOID 3.2 AFTER PROPRANOLOL 2.0	6	116.67 ± 5.63 (120.11 ± 3.81)	85.00 ± 6.13	27.47 ± 2.29

Table 8. Comparison of the effects of ALKALOID on mean blood pressure before and after propranolol blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent mean blood pressure before propranolol administration.

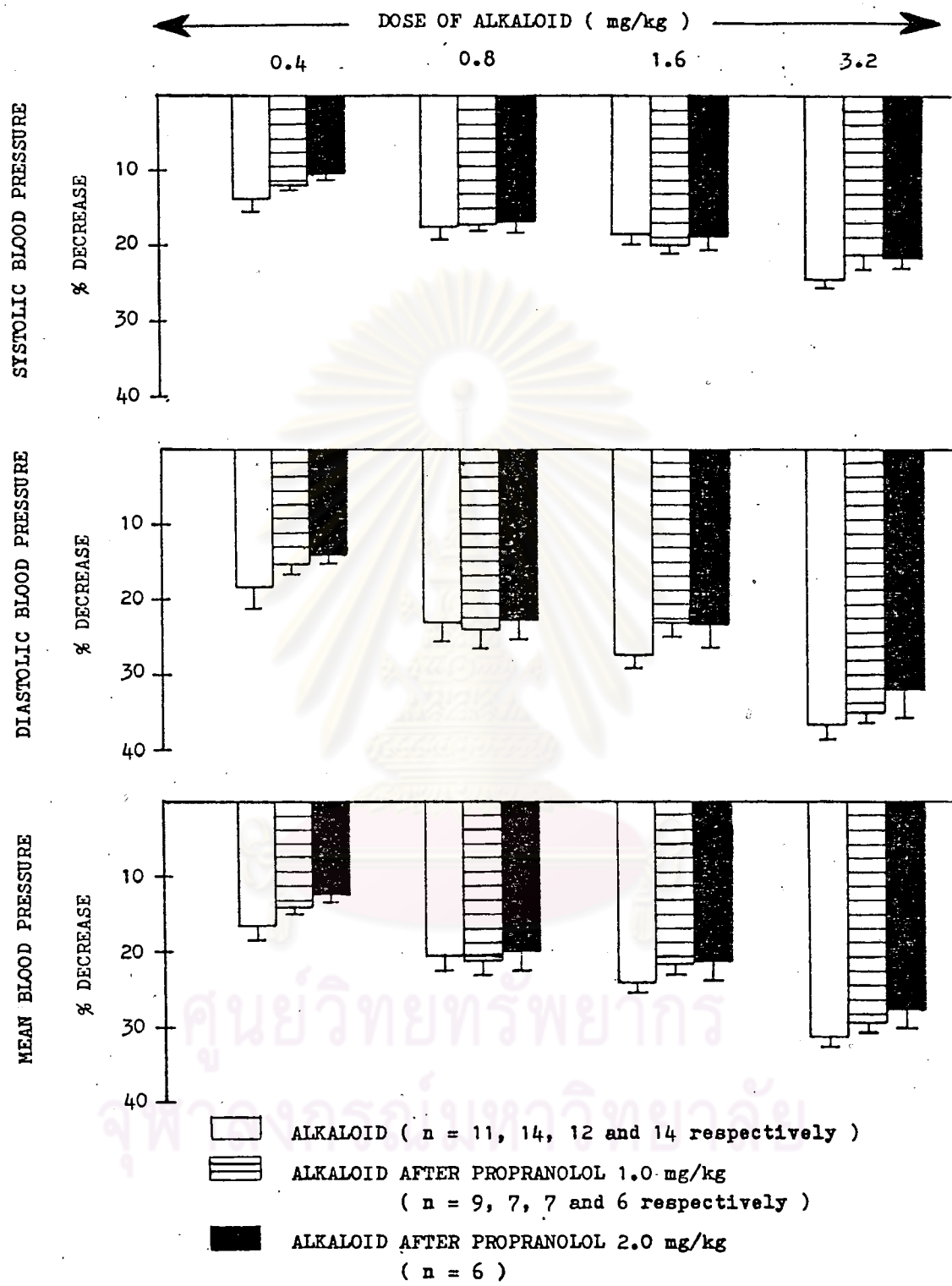


Figure 8. Comparison of the effects of ALKALOID on blood pressures before and after propranolol blockade in anaesthetized rats. Bar graphs represent mean percent decrease + S.E.M. The ordinate scales are percent decrease in blood pressure; the abscissa scale is dose of ALKALOID in mg/kg B.W.

Experiment 3. The Effects of Selective Antihistaminic Drug (H_1 -and H_2 -Receptor Antagonists) on the Hypotensive Effect of ALKALOID

Histamine 10 $\mu\text{g}/\text{kg}$ B.W. caused a marked falls in systemic blood pressure, as shown in Fig. 9. This hypotensive effect was abolished by administration of mepyramine 10 mg/kg B.W. plus cimetidine 20 mg/kg B.W. 5 min before addition of histamine. The data obtained from this experiment are summarized in Table 9.

Administration of the mepyramine 10 mg/kg B.W. plus cimetidine 20 mg/kg B.W. 5 min before ALKALOID showed no significant reduction in hypotensive effect of ALKALOID ($P > 0.05$, unpaired t-test). Records of some experiments are shown in Fig. 10. The effects of ALKALOID before and after these two antihistamine blockade are illustrated in Table 10, 11, 12 and Fig. 11.

Experiment 4. The Effects of Cholinergic Blocking Drug (Atropine) on the Hypotensive Effect of ALKALOID

An intravenous infusion of acetylcholine 1.2 $\mu\text{g}/\text{kg}$ B.W. elicited a marked hypotensive effect. This effect was abolished by prior administration of atropine 0.2 mg/kg B.W. 5 min. Again as shown in Fig. 12, the hypotensive effect of acetylcholine 2.0 $\mu\text{g}/\text{kg}$ B.W. was also abolished by prior administration of atropine 0.3 mg/kg B.W. Summarization of these results are presented in Table 13.

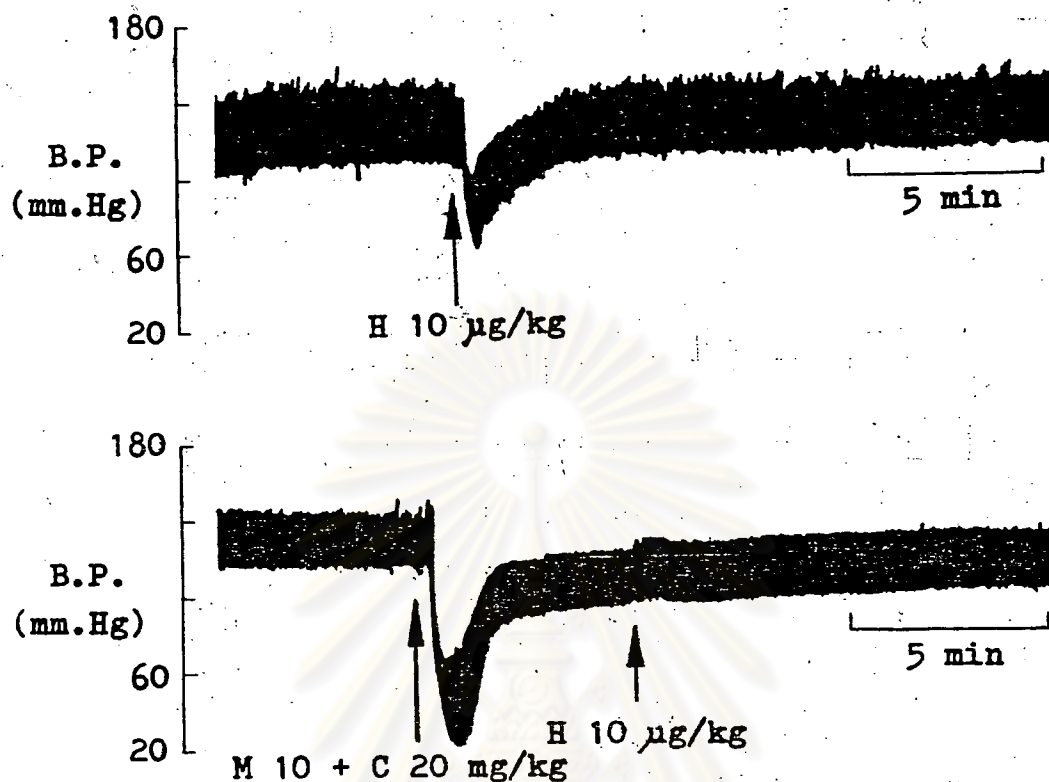


Figure 9. Records showing the effects of histamine (H) 10 $\mu\text{g}/\text{kg}$ on blood pressure before and after mepyramine (M) 10 mg/kg plus cimetidine (C) 20 mg/kg blockade for 5 min in anaesthetized rats.

SUBSTANCE	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE HISTAMINE INFUSION	AFTER HISTAMINE INFUSION	% DECREASE
HISTAMINE 10 $\mu\text{g}/\text{kg}$	5	121.60 \pm 3.62	71.20 \pm 6.74	41.85 \pm 3.79
HISTAMINE 10 $\mu\text{g}/\text{kg}$ AFTER MEPYRAMINE 10 mg/kg PLUS CIMETIDINE 20 mg/kg	5	99.46 \pm 5.53 (114.13 \pm 4.34)	99.46 \pm 5.53	0.00 \pm 0.00 ***

Table 9. Comparison of the effects of histamine on mean blood pressure before and after mepyramine plus cimetidine blockade in anaesthetized rats (mean \pm S.E.M.). Number in parenthesis represents mean blood pressure before mepyramine plus cimetidine administration.

*** $P < 0.001$, unpaired t-test

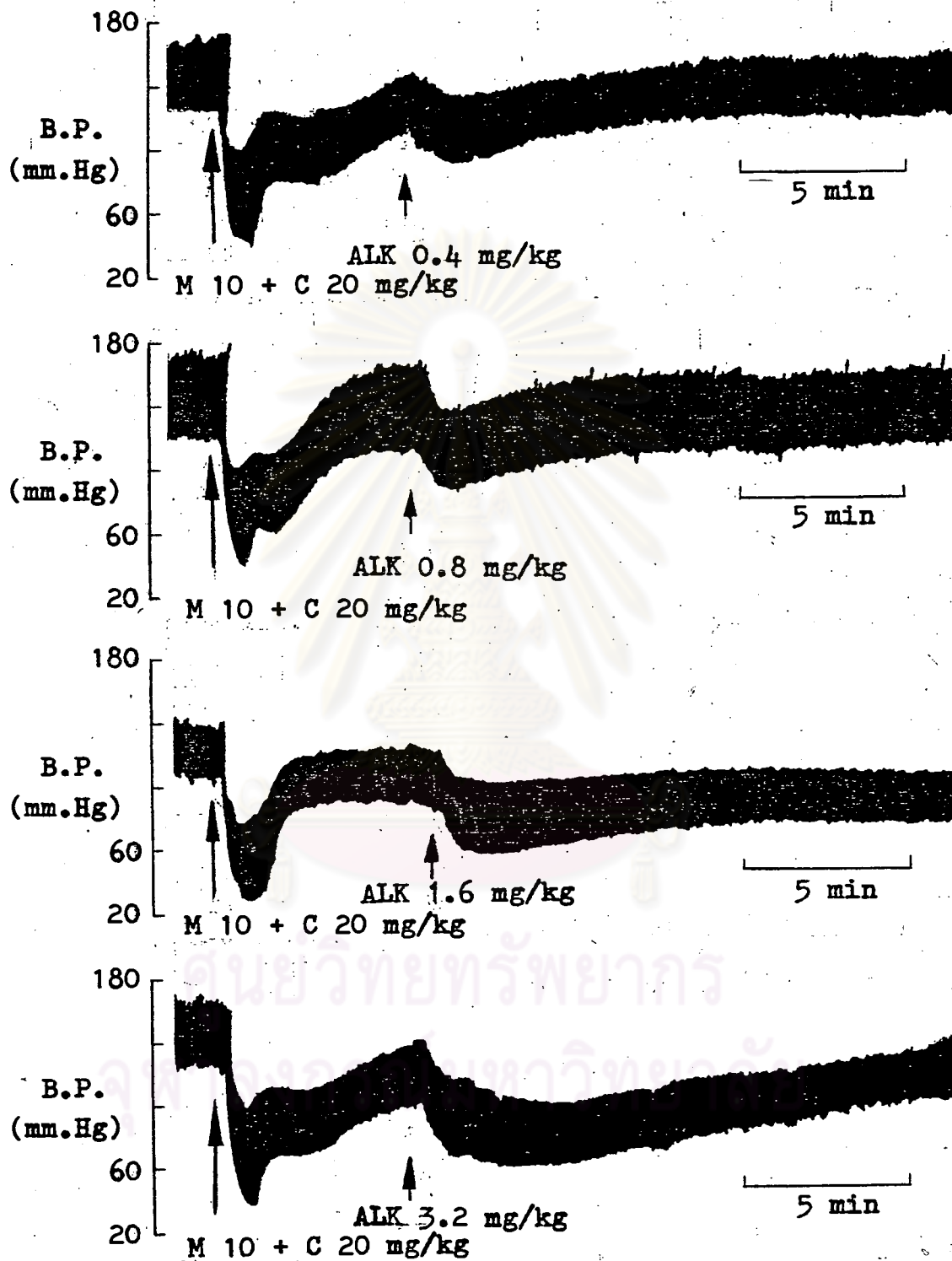


Figure 10. Records showing the effects of ALKALOID (ALK) in various doses on blood pressure after mepyramine (M) 10 mg/kg plus cimetidine (C) 20 mg/kg blockade for 5 min in anaesthetized rats.

SUBSTANCE (mg/kg)	NO. OF RAT	SYSTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	150.91 ± 4.21	129.82 ± 3.52	13.77 ± 1.76
ALKALOID 0.4 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	142.40 ± 9.42 (150.00 ± 4.09)	124.40 ± 6.69	12.21 ± 1.91
ALKALOID 0.8	14	146.86 ± 3.29	121.29 ± 3.29	17.36 ± 1.57
ALKALOID 0.8 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	132.20 ± 4.45 (148.80 ± 4.45)	115.20 ± 4.58	13.54 ± 1.55
ALKALOID 1.6	12	145.50 ± 3.18	118.33 ± 2.67	18.41 ± 1.33
ALKALOID 1.6 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	144.80 ± 8.32 (152.40 ± 1.60)	122.80 ± 5.21	14.90 ± 1.86
ALKALOID 3.2	14	153.57 ± 2.87	116.00 ± 2.49	24.42 ± 1.09
ALKALOID 3.2 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	139.60 ± 7.27 (150.40 ± 6.26)	108.40 ± 4.91	22.09 ± 2.38

Table 10. Comparison of the effects of ALKALOID on systolic blood pressure before and after mepyramine plus cimetidine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent systolic blood pressure before mepyramine plus cimetidine administration.

SUBSTANCE (mg/kg)	NO. OF RAT	DIASTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	102.73 ± 3.93	83.46 ± 4.01	18.73 ± 2.55
ALKALOID 0.4 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	105.20 ± 5.63 (112.00 ± 2.19)	86.80 ± 5.53	17.60 ± 1.74
ALKALOID 0.8	14	106.71 ± 2.54	82.71 ± 3.72	23.20 ± 2.28
ALKALOID 0.8 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	96.40 ± 2.71 (109.60 ± 4.11)	74.80 ± 3.97	22.47 ± 3.17
ALKALOID 1.6	12	105.33 ± 1.48	76.17 ± 1.93	27.63 ± 1.73
ALKALOID 1.6 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	100.40 ± 10.91 (108.00 ± 3.19)	74.40 ± 9.58	26.44 ± 2.62
ALKALOID 3.2	14	105.43 ± 3.61	67.29 ± 3.80	36.72 ± 2.02
ALKALOID 3.2 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	95.20 ± 5.15 (107.60 ± 4.91)	57.60 ± 2.13	38.62 ± 4.69

Table 11. Comparison of the effects of ALKALOID on diastolic blood pressure before and after mepyramine plus cimetidine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent diastolic blood pressure before mepyramine plus cimetidine administration.

SUBSTANCE (mg/kg)	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	118.79 ± 3.77	98.01 ± 3.61	16.62 ± 2.07
ALKALOID 0.4 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	117.60 ± 6.62 (124.67 ± 2.10)	99.33 ± 5.68	15.47 ± 1.71
ALKALOID 0.8	14	120.57 ± 2.76	95.57 ± 3.36	20.79 ± 1.87
ALKALOID 0.8 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	108.67 ± 3.11 (122.67 ± 4.16)	82.27 ± 3.77	18.79 ± 2.42
ALKALOID 1.6	12	118.83 ± 1.78	90.22 ± 1.71	24.00 ± 1.31
ALKALOID 1.6 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	115.20 ± 9.97 (123.33 ± 2.59)	90.53 ± 8.29	21.42 ± 1.97
ALKALOID 3.2	14	121.48 ± 3.20	83.52 ± 3.02	31.39 ± 1.27
ALKALOID 3.2 AFTER MEPYRAMINE 10 PLUS CIMETIDINE 20	5	110.00 ± 5.79 (121.84 ± 5.28)	74.53 ± 1.30	31.60 ± 3.30

Table 12. Comparison of the effects of ALKALOID on mean blood pressure before and after mepyramine plus cimetidine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent mean blood pressure before mepyramine plus cimetidine administration.

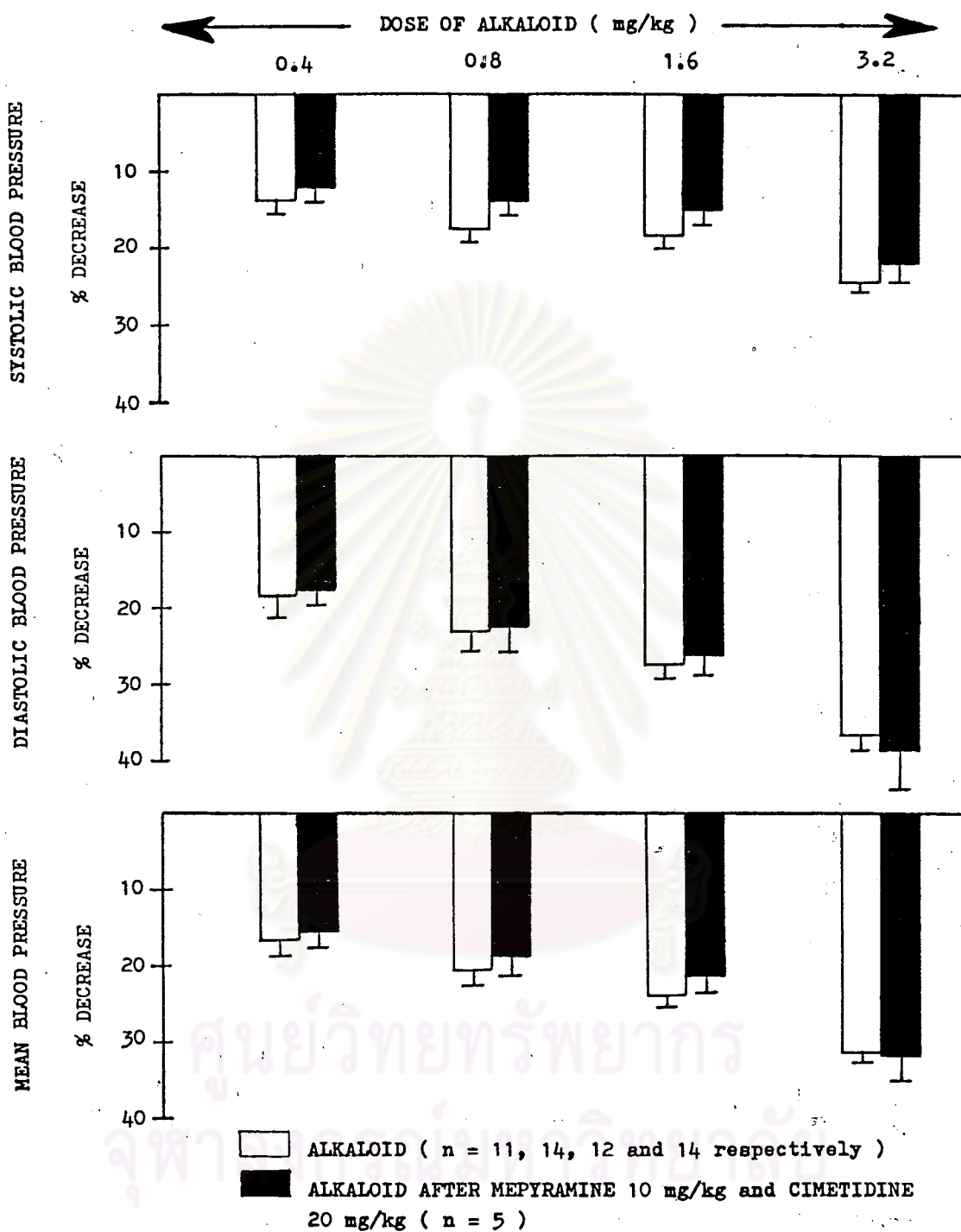


Figure 11. Comparison of the effects of ALKALOID on blood pressures before and after mepyramine plus cimetidine blockade in anaesthetized rats. Bar graphs represent mean percent decrease + S.E.M. The ordinate scales are percent decrease in blood pressure; the abscissa scale is dose of ALKALOID in mg/kg B.W.

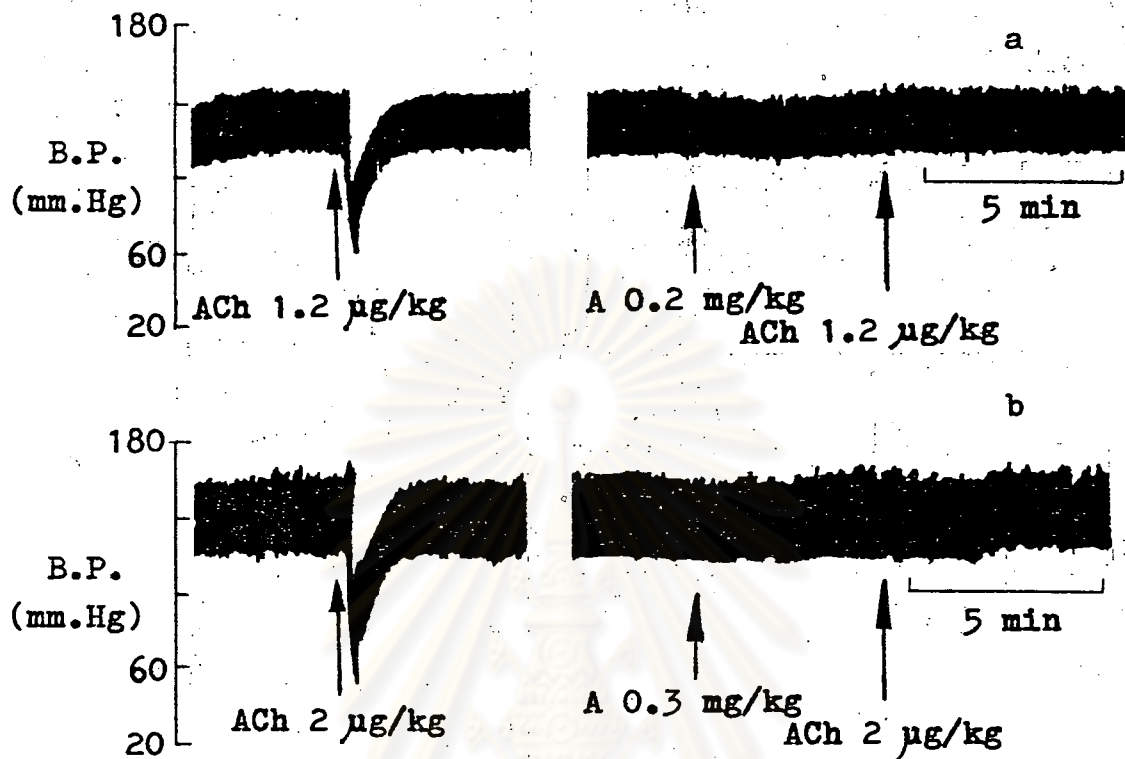


Figure 12. Records showing the effects on blood pressure in anaesthetized rats of

- a. Acetylcholine (ACh) 1.2 $\mu\text{g}/\text{kg}$ before and after atropine (A) 0.2 mg/kg blockade for 5 min.
- b. Acetylcholine (ACh) 2.0 $\mu\text{g}/\text{kg}$ before and after atropine (A) 0.3 mg/kg blockade for 5 min.

ศูนย์วิทยุทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

SUBSTANCE	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ACETYLCHOLINE INFUSION	AFTER ACETYLCHOLINE INFUSION	% DECREASE
ACETYLCHOLINE 1.2 µg/kg	5	123.20 ± 2.01	76.26 ± 1.44	38.04 ± 1.47
ACETYLCHOLINE 1.2 µg/kg AFTER ATROPINE 0.2 mg/kg	5	122.53 ± 2.63 (126.67 ± 2.00)	122.53 ± 2.63	0.00 ± 0.00
ACETYLCHOLINE 2.0 µg/kg	5	121.87 ± 5.66	65.87 ± 2.54	46.21 ± 1.84
ACETYLCHOLINE 2.0 µg/kg AFTER ATROPINE 0.3 mg/kg	5	119.07 ± 6.09 (120.93 ± 6.30)	119.07 ± 6.09	0.00 ± 0.00

Table 13. Comparison of the effects of acetylcholine on mean blood pressure before and after atropine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent mean blood pressure before atropine administration. *** P<0.001, unpaired t-test

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

As shown in Fig. 13, prior administration of atropine 0.2 mg/kg B.W. 5 min could partially reduce the hypotensive effect of ALKALOID. By observation, only ALKALOID 3.2 mg/kg B.W. after atropine produced shorter duration of the maximum reduction than ALKALOID alone.

Again as illustrated in Fig. 14, atropine 0.3 mg/kg B.W. could partially reduce the hypotensive effect of ALKALOID at all doses significantly. The reduction in blood pressure showed slightly decreased and then sustained. Summarization of the effects of ALKALOID on systolic, diastolic and mean blood pressures from each dose before and after atropine blockade are shown in Table 14, 15, 16 and Fig. 15.

Experiment 5. The Effects of Ganglionic Blocking Drug (Hexamethonium) on the Hypotensive Effect of ALKALOID

An intravenous infusion 3.5 mg/kg B.W. of hexamethonium caused a dramatic reduction in systemic blood pressure. This reduction was gradually recovered to control level within 15-20 min after administration. Prior administration of hexamethonium 3.5 mg/kg B.W. 3 min, the hypotensive effect of ALKALOID dose 0.8 mg/kg B.W. was variable reduction. However, the percentage changes of these reductions were relatively depended on the pressure at that period. The effects of ALKALOID 1.6 mg/kg B.W., at period of 3, 5 and 10 min after hexamethonium showed that hexamethonium could reduce the hypotensive effect of

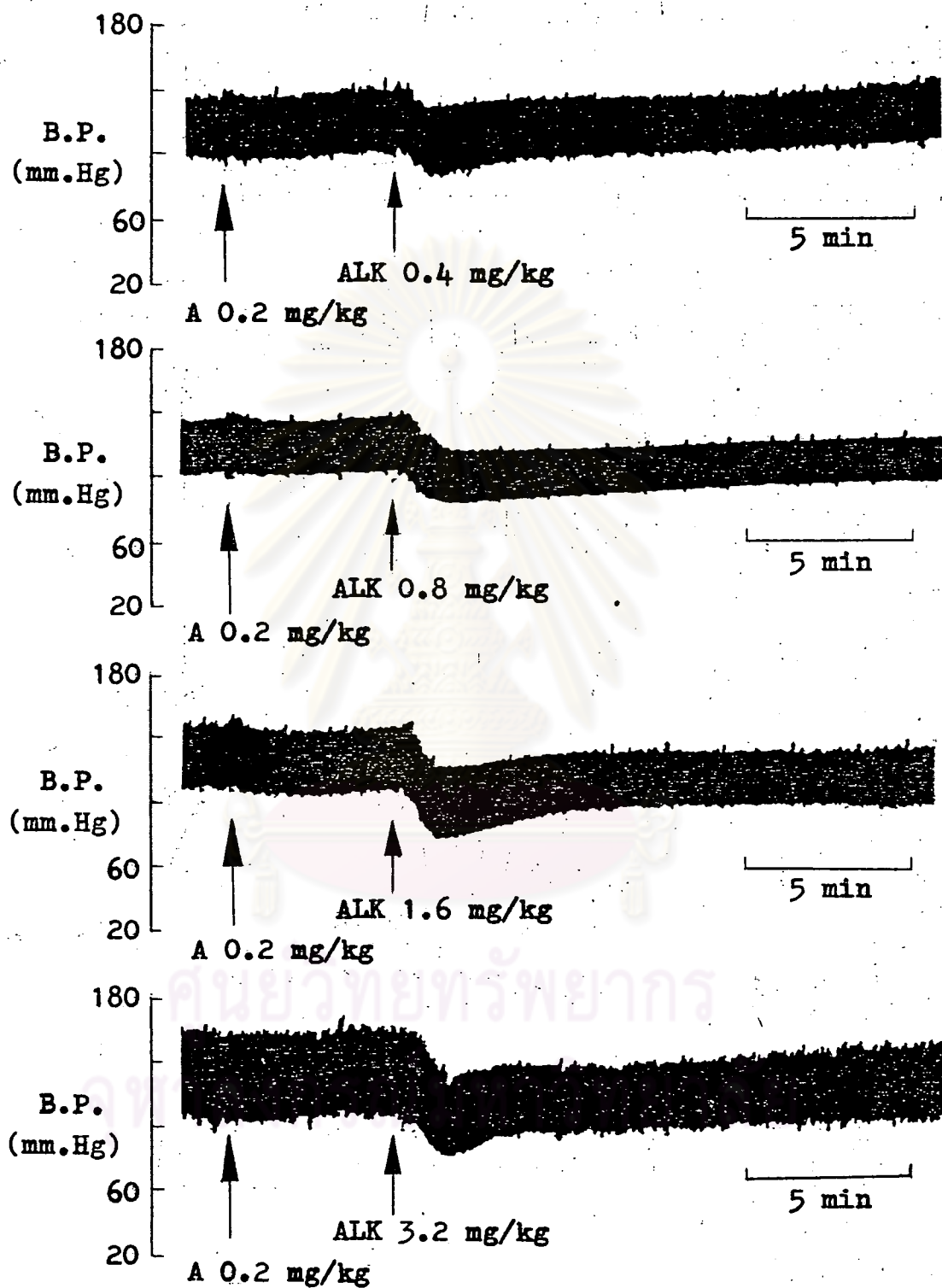


Figure 13. Records showing the effects of ALKALOID (ALK) in various doses on blood pressure after atropine (A) 0.2 mg/kg blockade for 5 min in anaesthetized rats.

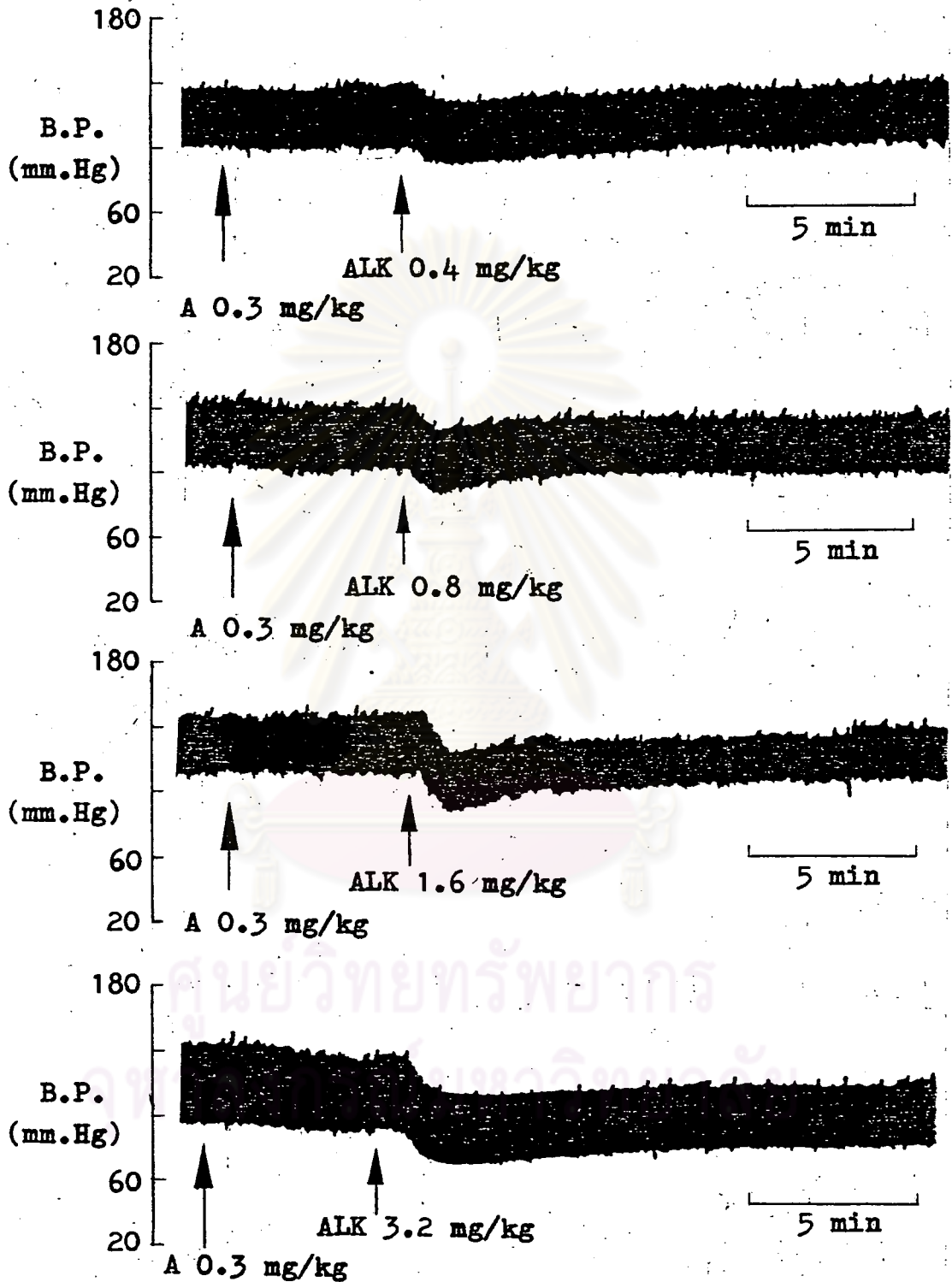


Figure 14. Records showing the effects of ALKALOID (ALK) in various doses on blood pressure after atropine (A) 0.3 mg/kg blockade for 5 min in anaesthetized rats.

SUBSTANCE (mg/kg)	NO. OF RAT	SYSTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	150.91 ± 4.21	129.82 ± 3.52	13.77 ± 1.76
ALKALOID 0.4 AFTER ATROPINE 0.2	9	146.44 ± 3.26 (147.33 ± 3.88)	131.33 ± 2.52	10.22 ± 1.23
ALKALOID 0.4 AFTER ATROPINE 0.3	8	149.00 ± 3.05 (150.50 ± 3.37)	136.00 ± 2.93	8.85 ± 1.17
ALKALOID 0.8	14	146.86 ± 3.29	121.29 ± 3.29	17.36 ± 1.57
ALKALOID 0.8 AFTER ATROPINE 0.2	8	144.25 ± 3.08 (146.75 ± 3.09)	124.25 ± 3.53	13.90 ± 1.35
ALKALOID 0.8 AFTER ATROPINE 0.3	11	148.91 ± 3.63 (150.55 ± 3.57)	128.55 ± 3.28	12.95 ± 1.21
ALKALOID 1.6	12	145.50 ± 3.18	118.33 ± 2.67	18.41 ± 1.33
ALKALOID 1.6 AFTER ATROPINE 0.2	11	144.73 ± 2.99 (146.91 ± 3.15)	118.36 ± 3.41	18.19 ± 1.72
ALKALOID 1.6 AFTER ATROPINE 0.3	9	147.78 ± 4.70 (148.00 ± 4.00)	126.22 ± 2.97	14.38 ± 1.05
ALKALOID 3.2	14	153.57 ± 2.87	116.00 ± 2.49	24.42 ± 1.09
ALKALOID 3.2 AFTER ATROPINE 0.2	9	144.00 ± 1.89 (147.11 ± 2.34)	116.00 ± 2.24	19.43 ± 1.25
ALKALOID 3.2 AFTER ATROPINE 0.3	9	146.00 ± 3.83 (149.11 ± 2.59)	122.89 ± 4.08	15.83 ± 1.79

Table 14. Comparison of the effects of ALKALOID on systolic blood pressure before and after atropine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent systolic blood pressure before atropine administration. * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$, unpaired t-test.

SUBSTANCE (mg/kg)	NO. OF RAT	DIASTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	102.73 ± 3.93	83.46 ± 4.01	18.73 ± 2.55
ALKALOID 0.4 AFTER ATROPINE 0.2	9	105.78 ± 1.90 (106.00 ± 1.83)	91.56 ± 2.16	13.47 ± 1.12
ALKALOID 0.4 AFTER ATROPINE 0.3	8	108.00 ± 3.19 (108.50 ± 2.66)	96.00 ± 3.95	11.30 ± 1.40 *
ALKALOID 0.8	14	106.71 ± 2.54	82.71 ± 3.72	23.20 ± 2.28
ALKALOID 0.8 AFTER ATROPINE 0.2	8	106.25 ± 3.03 (107.00 ± 3.29)	87.50 ± 3.29	17.72 ± 1.48
ALKALOID 0.8 AFTER ATROPINE 0.3	11	107.46 ± 2.40 (107.09 ± 2.48)	90.36 ± 2.57	15.96 ± 1.23 *
ALKALOID 1.6	12	105.33 ± 1.48	76.17 ± 1.93	27.63 ± 1.73
ALKALOID 1.6 AFTER ATROPINE 0.2	11	106.36 ± 2.92 (107.64 ± 3.15)	78.91 ± 4.48	26.12 ± 2.95
ALKALOID 1.6 AFTER ATROPINE 0.3	9	108.22 ± 4.48 (107.77 ± 4.29)	88.44 ± 3.71	18.24 ± 0.94 ***
ALKALOID 3.2	14	105.43 ± 3.61	67.29 ± 3.80	36.72 ± 2.02 *
ALKALOID 3.2 AFTER ATROPINE 0.2	9	102.67 ± 2.75 (104.44 ± 2.51)	74.00 ± 3.82	28.18 ± 2.34
ALKALOID 3.2 AFTER ATROPINE 0.3	9	106.67 ± 2.49 (109.11 ± 1.57)	86.67 ± 3.15	18.87 ± 1.58 ***

Table 15. Comparison of the effects of ALKALOID on diastolic blood pressure before and after atropine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent diastolic blood pressure before atropine administration. * P < 0.05 *** P < 0.001, unpaired t-test.

SUBSTANCE (mg/kg)	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.4	11	118.79 ± 3.77	98.01 ± 3.61	16.62 ± 2.07
ALKALOID 0.4	9	119.33 ± 1.99	104.81 ± 1.94	12.14 ± 1.05
AFTER ATROPINE 0.2		(119.78 ± 2.00)		*
ALKALOID 0.4 AFTER ATROPINE 0.3	8	121.67 ± 2.84 (122.50 ± 2.56)	109.33 ± 3.47	10.29 ± 1.27
ALKALOID 0.8	14	120.57 ± 2.76	95.57 ± 3.36	20.76 ± 1.87
ALKALOID 0.8 AFTER ATROPINE 0.2	8	118.92 ± 2.94 (120.25 ± 3.13)	99.75 ± 3.30	16.19 ± 1.30
ALKALOID 0.8 AFTER ATROPINE 0.3	11	121.28 ± 2.71 (121.58 ± 2.69)	103.39 ± 2.48	14.71 ± 1.08
ALKALOID 1.6	12	118.83 ± 1.78	90.22 ± 1.71	24.00 ± 1.31
ALKALOID 1.6 AFTER ATROPINE 0.2	11	119.15 ± 2.86 (120.73 ± 3.05)	90.73 ± 4.21	22.86 ± 2.33
ALKALOID 1.6 AFTER ATROPINE 0.3	9	121.41 ± 4.40 (121.19 ± 4.06)	101.04 ± 3.30	16.66 ± 0.74
ALKALOID 3.2	14	121.48 ± 3.20	83.52 ± 3.02	31.39 ± 1.27
ALKALOID 3.2 AFTER ATROPINE 0.2	9	116.44 ± 2.29 (119.33 ± 2.42)	88.00 ± 3.19	24.54 ± 1.82
ALKALOID 3.2 AFTER ATROPINE 0.3	9	119.78 ± 2.84 (122.44 ± 1.73)	98.74 ± 3.36	17.64 ± 1.55

Table 16. Comparison of the effects of ALKALOID on mean blood pressure before and after atropine blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent mean blood pressure before atropine administration. * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$, unpaired t-test

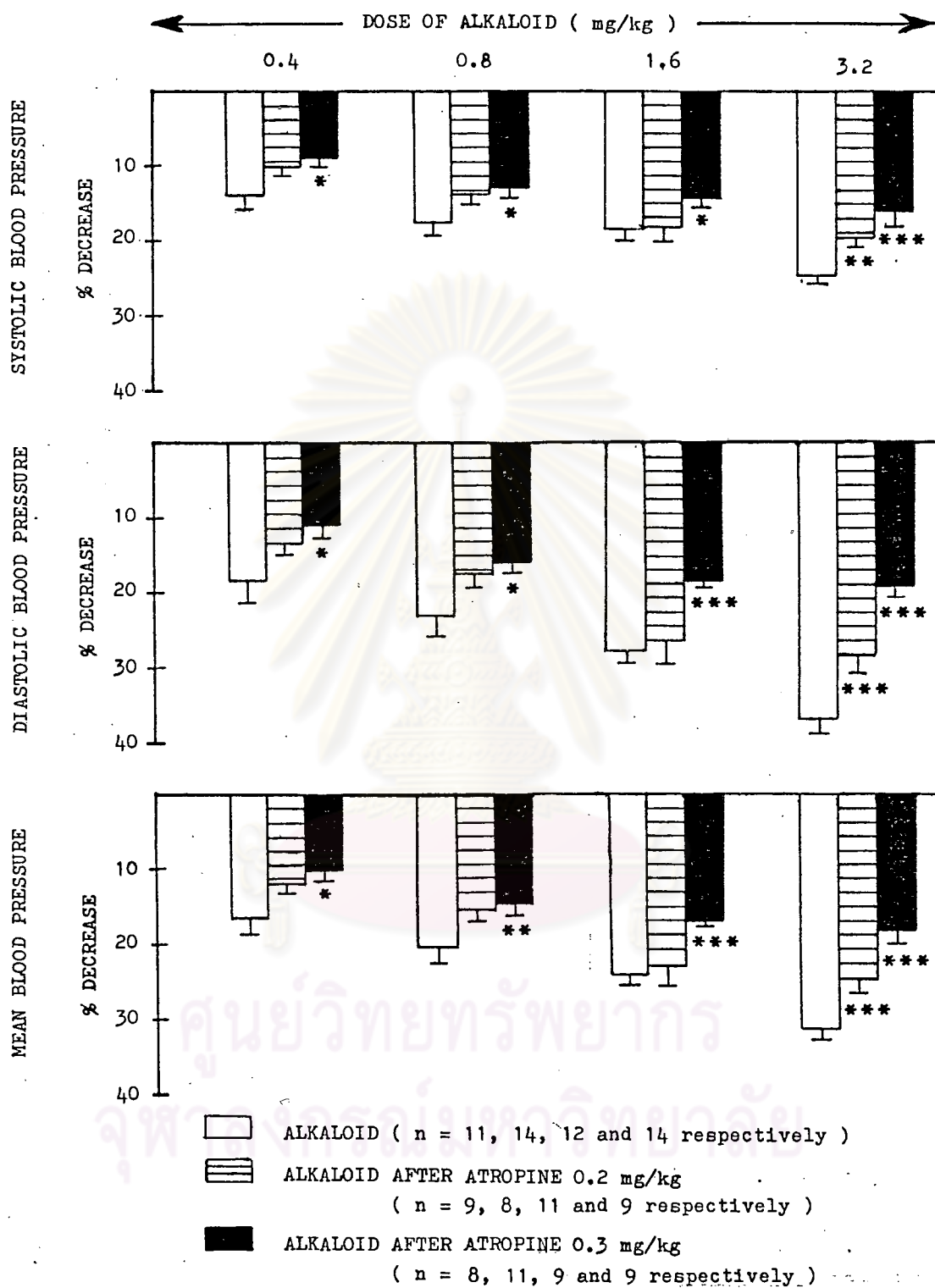


Figure 15. Comparison of the effects of ALKALOID on blood pressures before and after atropine blockade in anaesthetized rats. Bar graphs represent mean percent decrease + S.E.M. The ordinate scales are percent decrease in blood pressure; the abscissa scale is dose of ALKALOID in mg/kg B.W. * $P < 0.05$ ** $P < 0.01$ *** $P < 0.001$, unpaired t-test.

ALKALOID. Again the percentage changes of these reductions were depended on the pressure level at that period. The percent changes on systemic blood pressure are summarized in Table 17, 18, 19 and Fig. 17.

Experiment 6. The Effects of ALKALOID on Norepinephrine Releasing Sympathomimetic Amine (Tyramine)

Intravenous infusion of tyramine 0.3 mg/kg B.W. elicited an increase in mean blood pressure, as shown in Fig. 18 and Table 20. Administration of ALKALOID 3.2 mg/kg B.W. caused a dramatic reduction in systemic blood pressure which could be reversed immediately after infusion of tyramine (Fig. 18). Comparison of the effects of tyramine on mean blood pressure before and after ALKALOID blockade were not statistically significance ($P > 0.05$, unpaired t-test) (Fig. 19 Table 20).

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

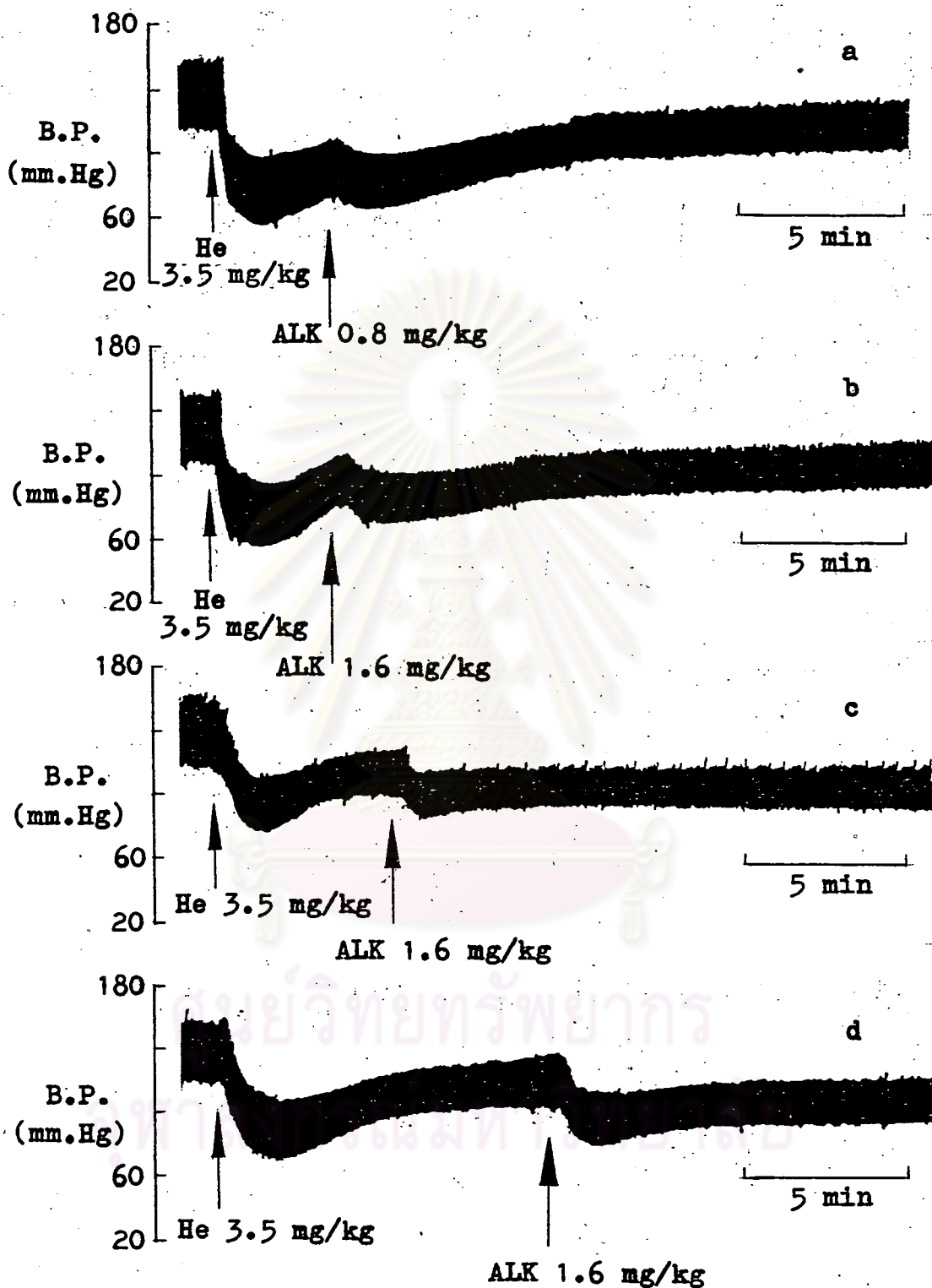


Figure 16. Records showing the effects of ALKALOID (ALK) on blood pressure after hexamethonium (He) 3.5 mg/kg blockade for 3 min (a and b), 5 min (c), 10 min (d) in anaesthetized rats.

SUBSTANCE (mg/kg)	NO. OF RAT	SYSTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.8	14	146.86 ± 3.29	121.29 ± 3.29	17.36 ± 1.57
ALKALOID 0.8 AFTER HEXAMETHONIUM 3.5 3 MIN	6	113.00 ± 2.62 (143.67 ± 3.80)	102.33 ± 2.44	9.42 ± 0.98
ALKALOID 1.6	12	145.50 ± 3.18	118.33 ± 2.67	18.41 ± 1.33
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 3 MIN	5	116.80 ± 1.02 (150.80 ± 3.71)	103.20 ± 0.80	11.63 ± 0.58
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 5 MIN	5	116.80 ± 3.45 (144.40 ± 3.59)	104.40 ± 3.59	10.63 ± 1.38
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 10 MIN	5	127.60 ± 5.94 (143.20 ± 6.24)	109.20 ± 7.35	14.68 ± 2.08

Table 17. Comparison of the effects of ALKALOID on systolic blood pressure before and after hexamethonium blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent systolic blood pressure before hexamethonium administration.

SUBSTANCE (mg/kg)	NO. OF RAT	DIASTOLIC BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.8	14	106.71 ± 2.54	82.71 ± 3.72	23.20 ± 2.28
ALKALOID 0.8 AFTER HEXAMETHONIUM 3.5 3 MIN	6	84.00 ± 3.31 (107.67 ± 2.22)	73.67 ± 3.16	12.24 ± 1.92
ALKALOID 1.6	12	105.33 ± 1.48	76.17 ± 1.93	27.63 ± 1.73
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 3 MIN	5	86.40 ± 1.72 (114.40 ± 2.31)	72.00 ± 2.45	16.70 ± 1.88
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 5 MIN	5	92.40 ± 3.54 (103.20 ± 5.38)	79.20 ± 3.61	14.26 ± 2.43
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 10 MIN	5	96.00 ± 6.47 (105.60 ± 6.51)	78.00 ± 8.70	19.54 ± 3.94

Table 18. Comparison of the effects of ALKALOID on diastolic blood pressure before and after hexamethonium blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent diastolic blood pressure before hexamethonium administration.

จุฬาลงกรณ์มหาวิทยาลัย



SUBSTANCE (mg/kg)	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE ALKALOID INFUSION	AFTER ALKALOID INFUSION	% DECREASE
ALKALOID 0.8	14	120.57 ± 2.76	95.57 ± 3.36	20.79 ± 1.87
ALKALOID 0.8 AFTER HEXAMETHONIUM 3.5 3 MIN	6	93.67 ± 2.94 (119.67 ± 2.67)	83.22 ± 2.82	11.11 ± 1.50
ALKALOID 1.6	12	118.83 ± 1.78	90.22 ± 1.71	24.00 ± 1.31
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 3 MIN	5	96.53 ± 1.29 (126.27 ± 2.71)	82.40 ± 1.81	14.65 ± 1.33
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 5 MIN	5	100.54 ± 3.38 (117.20 ± 4.74)	87.60 ± 3.52	12.86 ± 1.96
ALKALOID 1.6 AFTER HEXAMETHONIUM 3.5 10 MIN	5	106.53 ± 6.18 (118.13 ± 6.41)	88.40 ± 8.19	17.56 ± 3.16

Table 19. Comparison of the effects of ALKALOID on mean blood pressure before and after hexamethonium blockade in anaesthetized rats (mean ± S.E.M.). Numbers in parentheses represent mean blood pressure before hexamethonium administration.

จุฬาลงกรณ์มหาวิทยาลัย

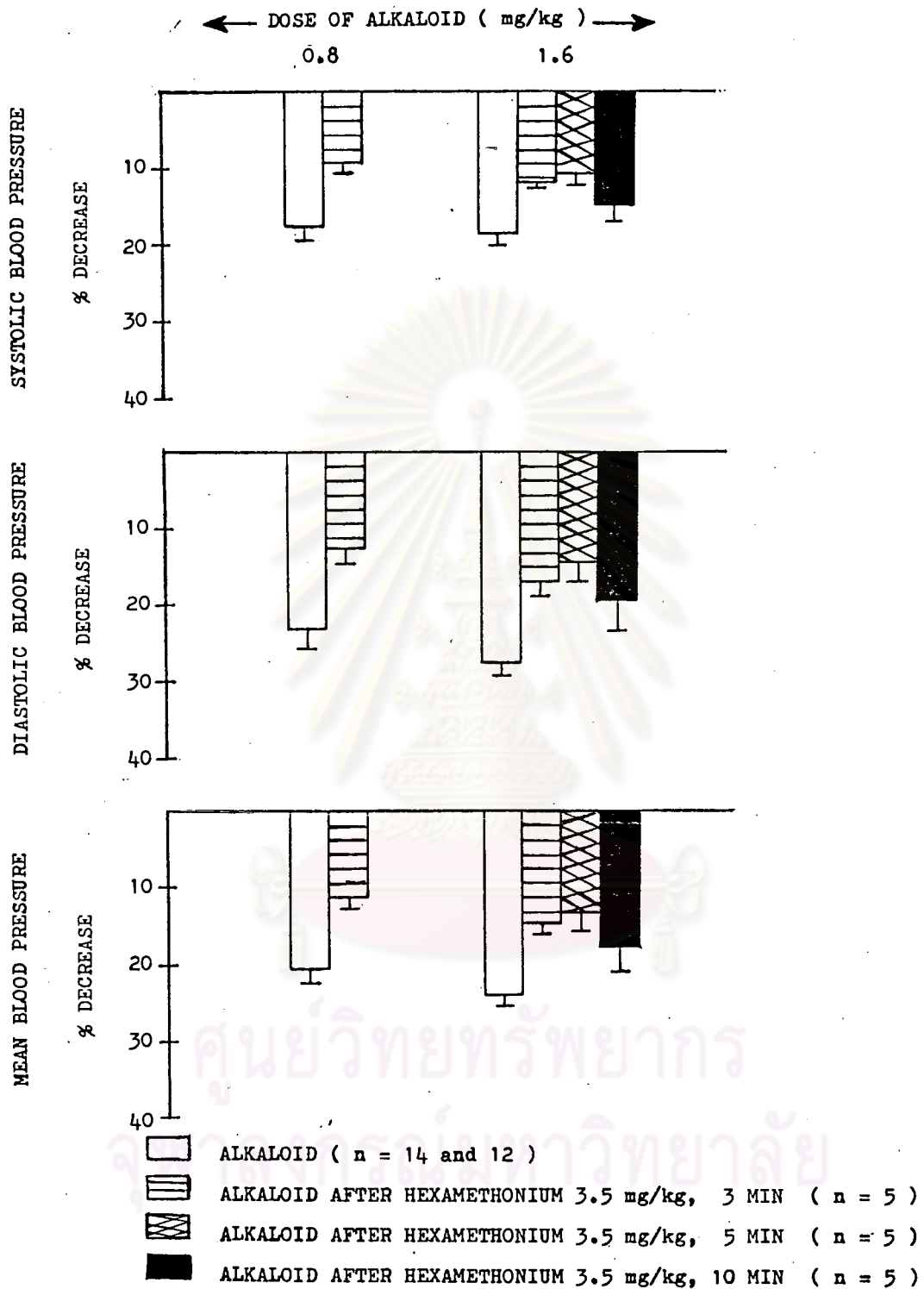


Figure 17. Comparison of the effects of ALKALOID on blood pressures before and after hexamethonium blockade in anaesthetized rats. Bar graphs represent mean percent decrease + S.E.M. The ordinate scales are percent decrease in blood pressure; the abscissa scale is dose of ALKALOID in mg/kg B.W.

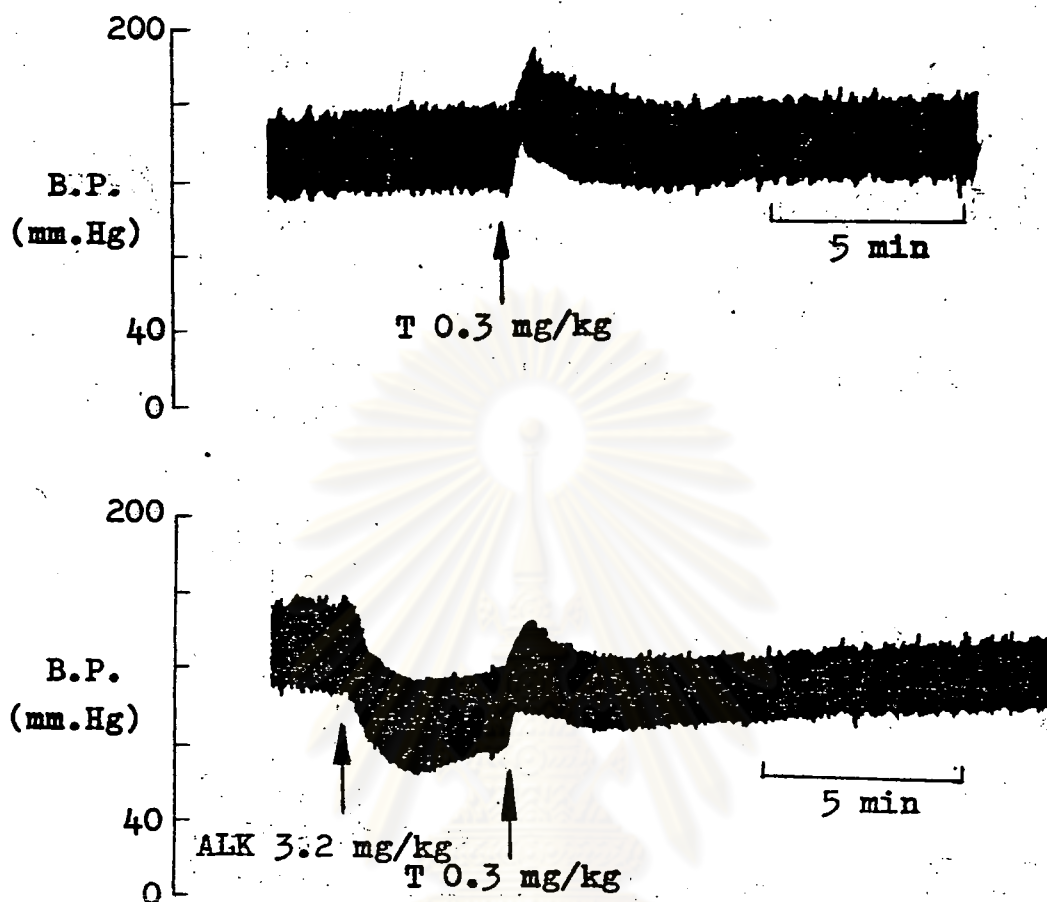


Figure 18. Records showing the effects of tyramine (T) 0.3 mg/kg on blood pressure before and after ALKALOID (ALK) 3.2 mg/kg blockade for 3 min in anaesthetized rats.

ศูนย์วิทยุทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

SUBSTANCE (mg/kg)	NO. OF RAT	MEAN BLOOD PRESSURE (mm.Hg)		
		BEFORE TYRAMINE INFUSION	AFTER TYRAMINE INFUSION	% INCREASE
TYRAMINE 0.3	9	124.22 ± 3.31	150.30 ± 4.72	20.90 ± 0.99
TYRAMINE 0.3 AFTER ALKALOID 3.2, 3 MIN	9	97.85 ± 2.20 (115.78 ± 2.82)	116.22 ± 3.06	18.76 ± 1.56

Table 20. Comparison of the effects of tyramine on mean blood pressure before and after ALKALOID blockade in anaesthetized rats (mean ± S.E.M.). Number in parenthesis represents mean blood pressure before ALKALOID administration.

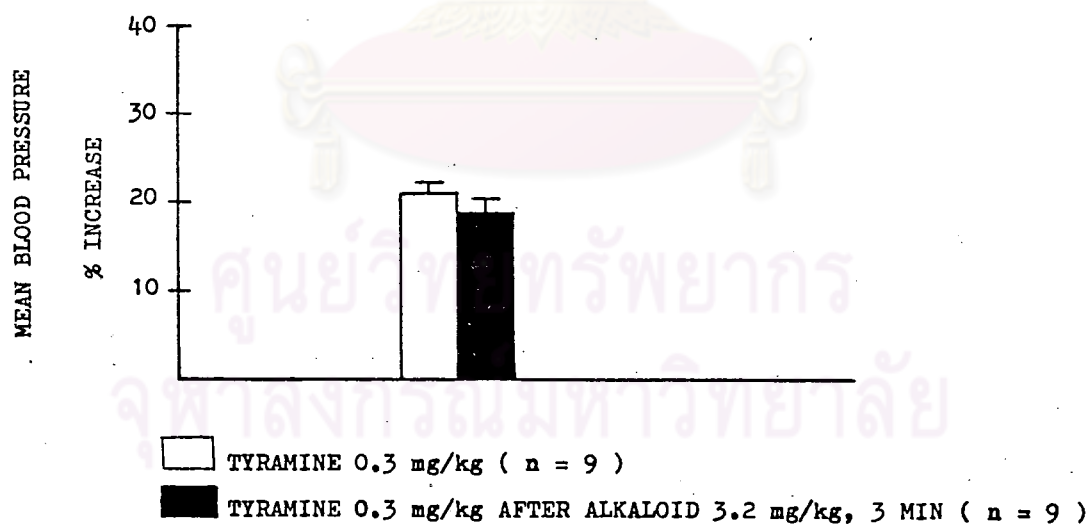


Figure 19. Comparison of the effects of tyramine on mean blood pressure before and after ALKALOID blockade in anaesthetized rats. Bar graphs represent mean percent increase + S.E.M. The ordinate scale is percent increase in mean blood pressure.

In Isolated Rat Atrial Strips:

Experiment 7. The Effects of ALKALOID on the Isolated Right Atrial Strips

Contractile response of the right atria when exposed to various concentration of ALKALOID (0.02, 0.04, 0.08 and 0.13 mg/ml) were measured in Locke solution for a period of 30 min after addition of each concentration of ALKALOID to the bathing fluid. As shown in Fig. 20, ALKALOID caused significant ($P < 0.05$, paired t-test) negative chronotropic effect which was found that the degree of these effect was related to the increased doses. In this experiment a dose of 0.13 mg/ml induced cardiac arrhythmias in 3 from 7 of right atrial strips.

Experiment 8. The Effects of ALKALOID on the Isolated Left Atrial Strips

ALKALOID (0.02, 0.04, 0.08 and 0.13 mg/ml) caused slight variable depressed on myocardial contractile tension in electrical paced left atrial strips of the rats. At 30 min after addition, maximum effects obtained from each dose of ALKALOID were not difference from the others, as illustrated in Fig. 21.

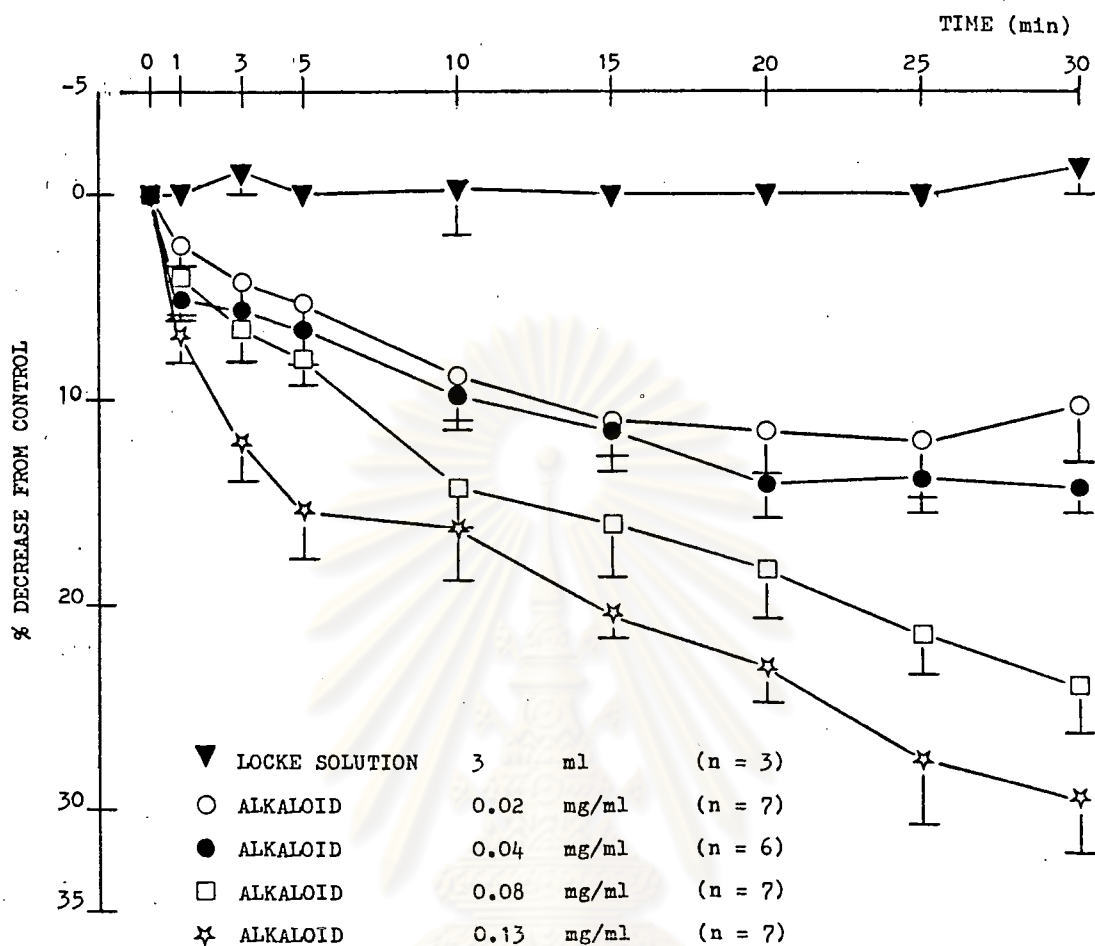


Figure 20. The concentration response curves for negative chronotropic effect of ALKALOID on isolated right atrial strips of the rats. Each point is the mean + S.E.M. The ordinate scale is percent decrease in heart rate; the abscissa scale is the time in minute.

จุฬาลงกรณ์มหาวิทยาลัย

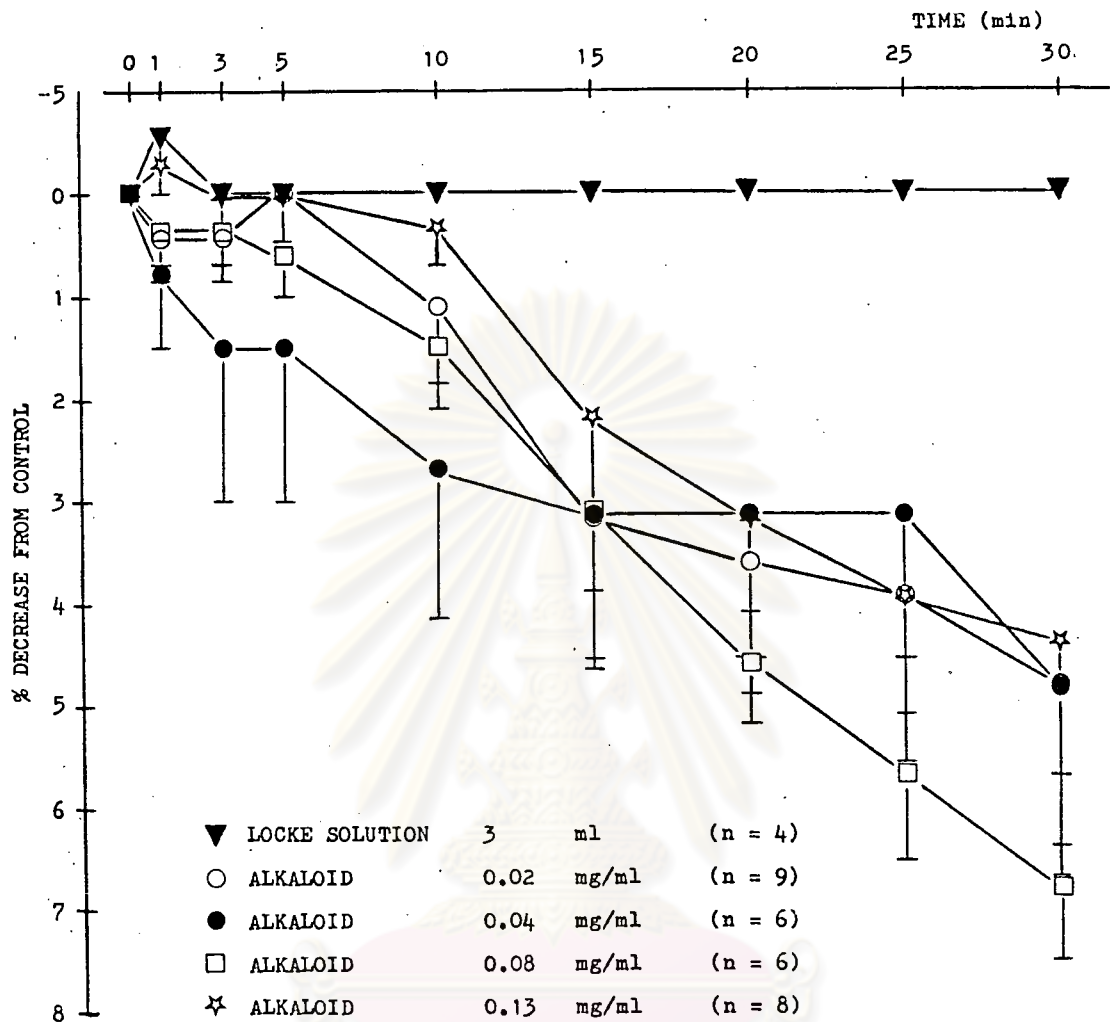


Figure 21. The effects of ALKALOID on inotropic response of the rats isolated left atrial strips. Each point is the mean + S.E.M. The ordinate scale is percent decrease in contractile tension; the abscissa scale is the time in minute.

Experiment 9. The Effects of Cholinergic Blocking Drug (Atropine) on Negative Chronotropic Effect of ALKALOID

The negative chronotropic effect of ALKALOID 0.08 mg/ml on spontaneous beating right atrial strips of the rats was not significantly inhibited by prior administration of atropine 0.3 μ g/ml to the bath fluid 5 min ($P > 0.05$, unpaired t-test), as shown in Fig. 22. The same concentration of atropine could abolish the negative chronotropic effect of acetylcholine 0.5 μ g/ml significantly ($P < 0.001$, unpaired t-test) which was considerably more potent than ALKALOID 0.08 mg/ml in decreasing heart rate, as shown in Fig. 23.

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

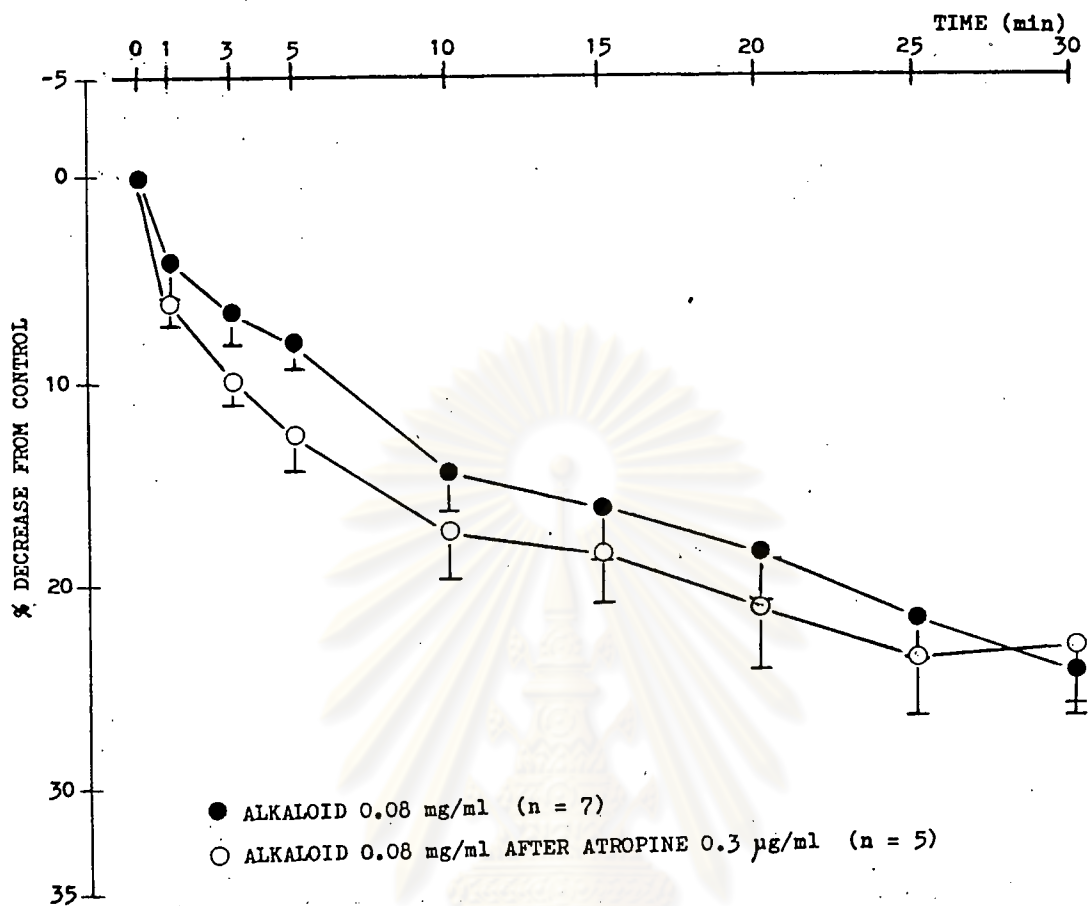


Figure 22. The negative chronotropic effect of ALKALOID before and after atropine blockade in isolated right atrial strips of the rats.

ศูนย์วิทยุทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย

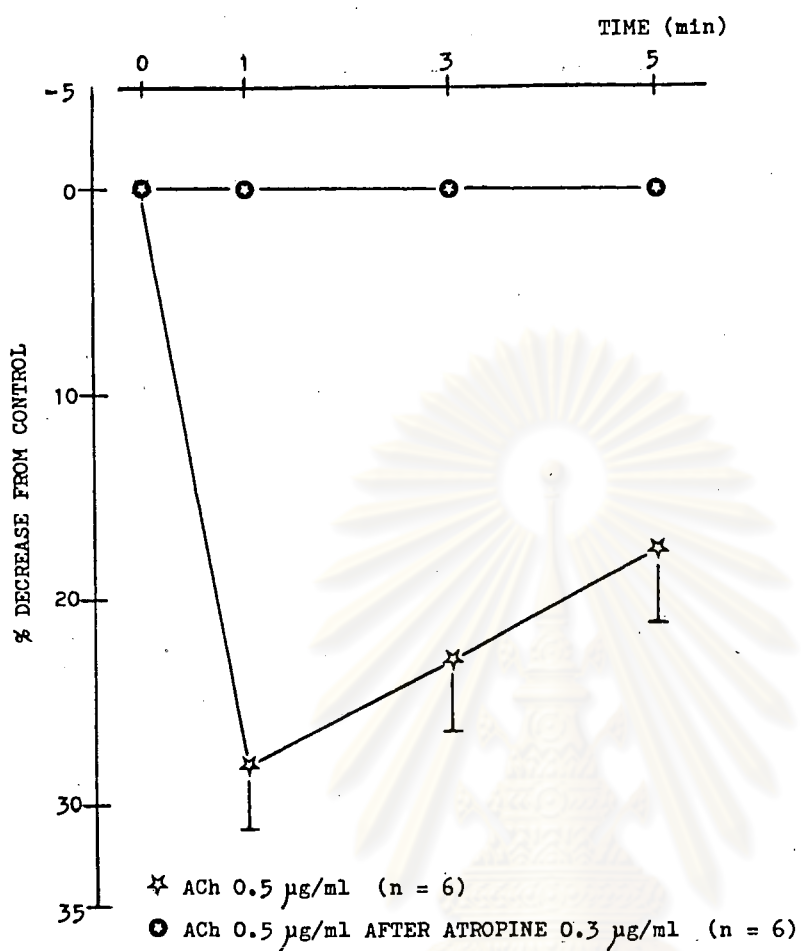


Figure 23. The inhibition effect of atropine on negative chronotropic response to acetylcholine in isolated right atrial strips of the rats.

ศูนย์วิทยทรัพยากร
จุฬาลงกรณ์มหาวิทยาลัย