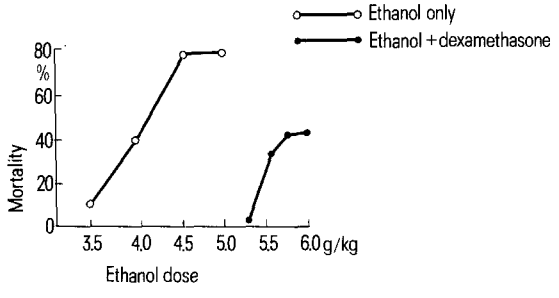
observed following drug(s) administration and the counts were made of the numbers that died or recovered after a 24-h-period.

Results. The figure shows that pretreatment with DXM shifted the mortality rate curve to the right. Maximum mortality rate using ethanol alone reached 80% using 4.5 or 5 g/kg of ethanol. Meanwhile the mortality rate was 40% using 5.75 g/kg or 6 g/kg of ethanol in animals pretreated with DXM 1 h prior the administration of ethanol. The difference in mortality rate between the 2 groups was found to be significant ($p<0.01$). It is of interest to note that using ethanol alone there was a linear relationship between the mortality rate and doses ranged between 3.5 and 4.5 g/kg. The same relationship was obvious in the DXM pretreated mice with higher levels of ethanol (between 5.25 and 5.75 g/kg). The figure also shows that no mortality in DXM pretreated animals, treated with 5.25 g/kg of ethanol. Lower dosage proved to be not lethal in the pretreated animals.

Discussion. It is clear from these results that DXM administration to mice prior to ethanol injection protected the animals from the lethal effect of ethanol. Since ethanol toxicity is directly related to respiratory depression[3], it might be suggested that DXM offers this protection through its central action. The central action of DXM was reported earlier[6]. The action of DXM might be brough about through the β-antagonist properties of the drug[5], by modulating brain biogenic amines concentrations. An inverse relationship has been found between circulating glucocorticoid levels and brain serotonin levels[7,8]. Meanwhile, ethanol effect has been known to be related to the increase of norepinephrine level in different brain regions[9].



The effect of dexamethasone pretreatment on ethanol toxicity. Each point represents the results on 12 animals.

1 Supported by a grant from U.S. National Aeronautics and Space Administration.
2 To whom request for reprints should be addressed.
3 A.A. Smith, K. Hyashida and Y. Kim, J. Pharm. Pharmac. *22*, 644 (1970).
4 K. Hyashida and A.A. Smith, J. Pharm. Pharmac. *23*, 718 (1971).
5 P. Fylling, Acta endocr. Copenh. *69*, 602 (1972).
6 K.F.A. Soliman and C.A. Walker, Experientia *33*, 400 (1977).
7 J. Vernikos-Danellis, P. Berger and J.D. Brachas, Prog. Brain Res. *39*, 301 (1973).
8 M.L. Simon and R. George, Neuroendocrinology *17*, 125 (1975).
9 A.T. Carlson, T. Magnusson, H. Sevenson and B. Waldeck, Psychopharmacologia *30*, 27 (1973).

N″-cyano-N-4-pyridyl-N′-1,2,2-trimethylpropylguanidine, monohydrate (P 1134): A new, potent vasodilator

E. Arrigoni-Martelli, Chr. Kaergaard Nielsen, U. Bang Olsen and H.J. Petersen[1]

Department of Pharmacology and Department of Chemistry, Leo Pharmaceutical Products, Ballerup (Denmark), 29 May 1979

Summary. N″-cyano-N-4-pyridyl-N′-1,2,2-trimethylpropylguanidine, monohydrate (P 1134) is a new agent which induces a marked and sustained hypotensive response in normotensive and renal, neurogenic, and spontaneously hypertensive rats, as well as in normotensive and renal hypertensive dogs. The overall potency of this compound is 2–3 times greater than that of hydralazine. The fall of blood pressure is accompanied by an increase in heart rate and cardiac output and a decrease in total peripheral resistance. The hypotensive effect appears to be due primarily to a direct relaxant effect on vascular smooth muscle.

Recent effort has concentrated on the development of agents which lower blood pressure by relaxation of vascular smooth muscle. The resultant vascular effect may correct the major hemodynamic disturbance in most hypertensive patients, i.e. the marked elevation of vascular resistance, without adding further abnormalities – as, for example, depression of cardiac output or impairment of sympathetic activity[2].

Following the observation some years ago that certain N-alkyl-N′-pyridylthioureas possess pronounced hypotensive activity[3] a series of N-alkyl-N″-cyano-N′-pyridylguanidines were synthesized[4]. P 1134, racemic N″-cyano-N-4-pyridyl-N′-1,2,2-trimethylpropylguanidine, monohydrate is a member of this series (figure 1). The present report describes its cardiovascular effects.

Materials and methods. Blood pressure was measured in conscious rats by an indirect method, by application to the tail of the animals of a Gartner cuff connected to an 8000 BP recorder (W+W Electronic, Basel).

In conscious dogs the following parameters were measured: a) Blood pressure, by an indirect method using ultrasonic Doppler detection of caudal artery wall motion during deflation of an occlusive cuff with a manometer (Roche Arteriosonde 1010). Heart rate was simultaneously recorded. b) Cardiac output, according to the thermodilution method of Ganz et al.[5] by means of an Edwards Labs. model 9500 computer with a Swan-Ganz thermodilution catheter No. 93-118-7F inserted in a jugular vein. c) From the cardiac output (=CO, l/min) and the mean arterial blood pressure (=MABP, mmHg) mean total peripheral resistance (=MTPR, mmHg/l/min) was calculated by the formula: MTPR=MABP/CO. d) Renal blood flow, by an indirect method, i.e. the clearance of para-amino-hippuric acid (PAH) and inulin, according to standard methods. e) Plasma renin activity, by radioimmunoassay using a modification of the technique of Haber et al.[6].

In anaesthetized dogs (Na pentobarbital 30 mg/kg i.v.) the blood pressure was measured by means of a Statham

transducer (P23 D6) inserted in a carotid artery and the renal, mesenteric, and femoral blood flows were measured by means of electromagnetic flowmeters (Nycotron) connected to a Grass 7C poligraph. The cardiac output was measured as previously described.

Experimental renal hypertension was produced in rats (Sprague-Dawley, Charles River, female, b.wt = 180–200 g) by the method of Grollman[7] and in dogs (mongrel, both sexes) by the method of Goldblatt et al.[8]. Neurogenic hypertension was produced in rats (Sprague-Dawley, female, b.wt = 200–220 g) by the method of Krieger et al.[9]. Genetic hypertensive rats (female, b.wt = 180–200 g) g) were of the Wistar-Okamoto strain.

Results and discussion. The table summarizes the hypotensive effects observed in conscious normotensive and in conscious renal, neurogenic, and spontaneously hypertensive rats administered a single oral dose of 2.5 mg/kg of either P 1134 or hydralazine. The greater effectiveness of P 1134 in comparison with hydralazine in hypertensive rats reached the level of significance ($p < 0.01$). For both compounds the peak effect was observed 2 h after treatment. The duration of the hypotensive effect of P 1134 exceeded 12 h and that of hydralazine never reached 8 h. The heart rate was found to be increased in normotensive rats by P 1134 and hydralazine 21% and 27% respectively. In renal and spontaneously hypertensive rats P 1134 increased the heart rate 15% and 18% and hydralazine 21% and 20%. In neurogenic hypertensive rats the observed increases were 7% and 9% in P 1134 and hydralazine treated animals, respectively.

Daily doses of 0.5, 1.25, and 2.5 mg/kg of P 1134 and 1.25, 2.5, and 5.0 mg/kg of hydralazine were administered orally for 5 consecutive days to groups of 10 spontaneously hypertensive rats. The results are shown in figure 2. Within the dose ranges tested there is a linear dose-response for both compounds on the 3rd and 5th day of treatment. The statistical analysis of the results shows that the 4 linear regressions are significant ($p < 0.001$). There is no significant difference between the slopes of the 2 regression lines

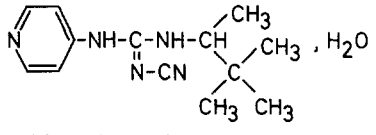


Fig. 1. Structural formula of P 1134.

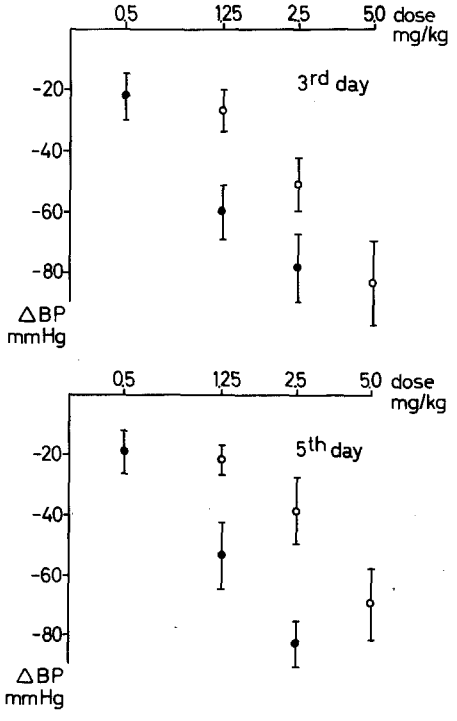


Fig. 2. Dose related decrease of the blood pressure in spontaneously hypertensive rats on the 3rd and 5th day of treatment with P 1134 (closed symbols) and hydralazine (open symbols) (mean ± SD, 10 rats each group).

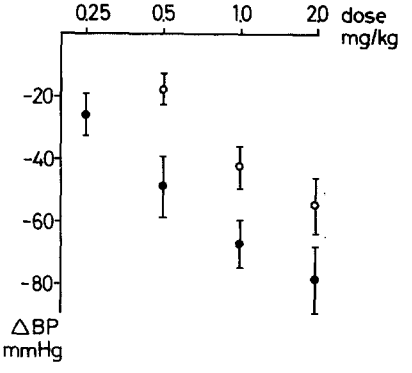


Fig. 3. Dose related decrease of the blood pressure in conscious renal hypertensive dogs treated with P 1134 (closed symbols) and hydralazine (open symbols) (mean ± SD, 5 dogs each group).

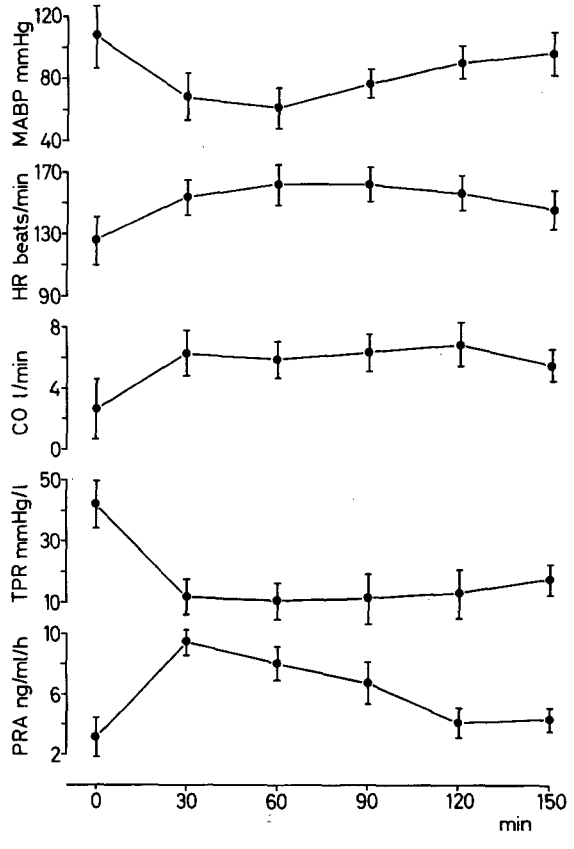


Fig. 4. Effect of P 1134, 0.5 mg/kg i.v. on mean arterial blood pressure (MABP), heart rate (HR), cardiac output (CO), total peripheral resistance (TPR), and plasma renin activity (RA) in conscious dogs (mean ± SD, 4 dogs).

Hypotensive action of P 1134 and hydralazine, 2.5 mg/kg p. o., in conscious rats, 10 animals per group

	P 1134 Mean blood pressure (mmHg) (mean ± SD)		Hydralazine Mean blood pressure (mmHg) (mean ± SD)	
	Before treatment	Peak fall	Before treatment	Peak fall
Normotensive	125± 7	13± 8	128± 10	24± 9
Spontaneously hypertensive	218± 11	98± 12	207± 12	62± 14
Renal hypertensive	177± 13	61± 11	181± 9	30± 12
Neurogenic hypertensive	167± 12	49± 11	173± 14	27± 9

on either the 3rd or 5th day of treatment. The potency ratio between P 1134 and hydralazine is 2.19 (1.95–2.73) on the 3rd day and 2.81 (2.42–3.36) on the 5th day.

Doses between 0.25 and 2.0 mg/kg of P 1134 and hydralazine orally administered to groups of 5 conscious renal hypertensive dogs caused a dose related depressor response (figure 3). Under these experimental conditions P 1134 was about 3 times more active than hydralazine. The blood pressure recorded 8 h after doses of P 1134 greater than 1 mg/kg was still lower than the control values. At that time the hypotensive effect of 2 mg/kg of hydralazine had vanished. The heart rate was found to be increased at the 2 highest doses of P 1134 and hydralazine 58% and 67%, and 72% and 83%, respectively.

Doses of 0.5 and 1.0 mg/kg of P 1134 were orally administered daily for 5 consecutive days to 4 renal hypertensive dogs. The blood pressure was recorded on the 1st and the 5th day just prior to the dosing and then hourly until the 6th h after dosing. No significant difference was observed in the intensity and duration of the hypotensive effects as assessed on the 1st and the 5th day of treatment. The renal clearances of PAH and inulin increased 41.5± 7.3% and 21.2± 3.4% respectively (mean ± SD in 4 normotensive conscious dogs) during the 1st h following the i.v. administration of 0.5 mg/kg of P 1134 and then returned to the range of control values. Higher doses of this compound causing fall of the blood pressure greater than 40–50 mmHg resulted in a decrease of renal clearance. These changes are most likely dependent on the autoregulatory capacity of the kidney and suggest that homeostatic circulatory reflexes remain fully functional.

In conscious normotensive dogs the fall of the blood pressure is the result of a marked decrease of peripheral resistance and it is associated with an increase of heart rate, cardiac output, and plasma renin activity (figure 4). These increases are most likely compensatory reactions to the fall of the blood pressure and are characteristic of a general systemic vasodilator drug[10–12].

In anaesthetized dogs the i.v. administration of doses between 0.25 and 1.0 mg/kg of P 1134 caused decreases of blood pressure similar in magnitude and duration to those observed in conscious animals. However, the barbiturate anaesthesia blunted, as expected, the increase of heart rate and cardiac output. Direct measurement of renal blood flow gave a picture similar to that obtained in conscious animals. Femoral and mesenteric blood flow were only marginally affected. Different degrees of vasodilation in the various vascular beds have been reported with hydralazine[13,14] and hydralazine derivatives[15].

The hypotensive activity of P 1134 is not modified by agents which block beta-adrenergic, cholinergic, or histaminergic receptors; thus it does not act via these receptor mechanisms. Preliminary findings obtained in in vitro experiments support the hypothesis that the hypotensive effect of P 1134 is mainly due to relaxation of peripheral vascular smooth muscles.

Consideration has been given to the possibility that the 2 enantiomers of P 1134 have different hypotensive potency. A separation of the isomers has been achieved[16] and the 2 enantiomers have been tested for acute toxicity in mice and hypotensive activity in spontaneously hypertensive rats in comparison with the racemate used in the experiments previously reported. No difference in toxicity was observed. The hypotensive effect of 4 different doses of each of the 3 compounds (6 rats/dose) has been evaluated according to the formula

$$\frac{(BP_{T0}-BP_{T2})+(BP_{T0}-BP_{T4})}{2}=\Delta \text{ mm Hg where}$$

BP_{T0} = blood pressure mm Hg

before drug, BP_{T2} = blood pressure mmHg 2 h after drug, BP_{T4} = blood pressure mmHg 4 h after drug. The dose of each compound causing a decrease of the blood pressure of 30 mmHg has been calculated and the following values have been found:

P 1134, racemic = 0.36 mg/kg p.o.
P 1134, (−)-enantiomer = 0.15 mg/kg p.o.
P 1134, (+)-enantiomer = 0.64 mg/kg p.o.

Toxicological evaluation of P 1134 (racemic) demonstrated that the drug was nontoxic in the rat and the dog after prolonged administration at dose levels far in excess of those required for hypotensive activity.

In hypertensive patients single doses of P 1134 up to 25 mg afforded substantial and prolonged (> 6 h) reduction of systolic and diastolic blood pressure with a concomitant pattern of hemodynamic changes typical for a peripheral vasodilator agent[17].

1 Acknowledgment. The helpful comments and advice of Dr W.O. Godtfredsen are gratefully acknowledged.
2 J. Koch-Weser, Arch. intern. Med. *133*, 1017 (1974).
3 H.J. Petersen, German Patent offenleg. 2557438, 1976.
4 H.J. Petersen, C. Kaergaard-Nielsen and E. Arrigoni-Martelli, J. med. Chem. *21*, 773 (1978).
5 A.W. Ganz, Am. J. Cardiol. *29*, 241 (1972).
6 E. Haber, T. Koerner, L.B. Page and B. Kliman, J. clin. Endocr. Metab. *29*, 1349 (1979).
7 A. Grollmann, Proc. Soc. exp. Biol. Med. *57*, 102 (1944).
8 H. Goldblatt, J. Lynch, R.F. Hanzan and W.W. Summerville, J. exp. Med. *59*, 347 (1934).
9 A.H. Krieger, Circulation Res. *15*, 511 (1964).
10 S. Mellander and R. Johansson, Pharmac. Rev. *20*, 117 (1968).
11 H. Brunner, P.R. Hedwall and M. Meier, Br. J. Pharmac. *30*, 123 (1967).
12 A.A. Rubin, L. Zitowitz and L. Hausler, J. Pharmac. exp. Ther. *140*, 46 (1963).
13 H.J. Bein, F. Gross, J. Tripod and R. Meier, Schweiz. med. Wschr. *83*, 336 (1953).
14 F. Gross, J. Druey and R. Meier, Experientia *6*, 11 (1950).
15 C. Carpi and L. Dorigotti, Br. J. Pharmac. *52*, 459 (1974).
16 The (+)- and (−)-forms were obtained by fractionated crystallization of the D(−)- and (+)-tartrates, respectively, from 80% EtOH. $[\alpha]_D^{20}$: +135° and −135° (c = 1, EtOH).
17 T. Hilden, personal communication.