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The role of endogenous bradykinin in blood pressure homeostasis in spontaneously hypertensive rats

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1 The role of endogenous bradykinin in mean arterial blood pressure (BP) homeostasis was studied in spontaneously hypertensive (SHR) and normotensive (WKY) rats by the use of a bradykinin B_2-receptor antagonist (BKant; Hoe 140, 11.6 $\mu g\ kg^{-1}$) and converting enzyme (kininase II) inhibitor (captopril, 10 mg). To obtain a response to captopril that was induced through inhibition of kinin-degradation only and not through inhibition of angiotensin II-formation, the studies were performed on binephrectomized male rats to eliminate the renin-angiotensin system.

2 The role of the nitric oxide (NO) and the adrenergic systems were evaluated by the use of NO-synthase inhibitor (L-NAME, 0.3 $g\ kg^{-1}$) and phentolamine (2 $mg\ kg^{-1}$), respectively.

3 The rats were anaesthetized and pretreated with two injections of vehicle (PBS) or drugs spaced 5 min apart: PBS+PBS; BKant+PBS; PBS+L-NAME; BKant+L-NAME; or phentolamine+L-NAME. All rats were given captopril 15 min later. Time-control groups were treated with L-NAME but not captopril.

4 In WKY rats, captopril did not significantly alter BP in any of the groups. In the SHR-PBS+PBS group, on the other hand, captopril induced an immediate fall in BP ($\Delta BP = -23 \pm 4$ mmHg, $P < 0.0017$) which was completely blocked by BKant ($\Delta BP = 2 \pm 2$ mmHg) ($P < 0.0011$). L-NAME did not significantly alter the immediate hypotensive response to captopril but disclosed a later hypertensive reaction. In L-NAME+BKant-treated rats, both the hypotensive response and the late hypertension was abolished. In rats treated with phentolamine+L-NAME, the immediate fall in BP was not different from the controls whereas the late hypertension was absent.

5 BKant itself had no effect on basal BP in either WKY or SHR even when a 10 times higher dose was tested in a separate set of experiments. This was true also for conscious, nonnephrectomized SHR rats.

6 It was concluded that endogenous production of bradykinin was demonstrable through kininase II-inhibition in hypertensive but not in normotensive rats. However, this endogenous bradykinin did not play a role in basal BP homeostasis. The captopril-induced hypotension depended on kinin but, under the present conditions, not on NO as a mediator. The fall in BP induced a compensatory adrenergic hypertensive response which was revealed when the continuous NO-synthesis was blocked by L-NAME.

Keywords: Bradykinin; bradykinin antagonist; converting enzyme inhibitor; captopril; NO; sympathetic nervous system; blood pressure; hypertension

Introduction

Bradykinin is a vasodilator peptide and a potent hypotensive agent. Breakdown of this peptide is predominantly catalyzed by converting enzyme/kininase II (E.C. 3.4.15.1) which also catalyzes the conversion of the pressor substance angiotensin I to angiotensin II (Yang *et al.*, 1970). Inhibitors of this enzyme (CEI) are widely used drugs in treatment of hypertensive disease. The role of kinins in blood pressure (BP) homeostasis and in the hypotensive effect of CEI, is still a matter for debate. However, bradykinin antagonists or antibodies to kinin have been shown to attenuate the hypotensive response to CEI not only in spontaneously hypertensive rats (SHR) (Cachofeiro *et al.*, 1992) but also in other models of hypertension (two-kidney, one clip, Benetos *et al.*, 1986a; and aortic ligation between both renal arteries, Carretero *et al.*, 1981; Carbonell *et al*, 1988). Moreover, CEI decreased BP in hypertensive subjects with normal and low renin activity (Gavras *et al.*, 1978). In normotensive, sodium-replete, human subjects a decrease in BP was observed after CEI but not after renin-inhibition (Kiowski *et al.*, 1992). Furthermore, high doses of a bradykinin antagonist have been shown to increase basal BP in normotensive rats (Benetos *et al.*, 1986b), although CEI did not induce hypotension in normotensive rats (Cachofeira *et al.*, 1992). These studies suggest a role for bradykinin in arterial blood pressure (BP) homeostasis in hypertensive disease and possibly also in the normotensive condition.

Several *in vitro* studies demonstrate the release of nitric oxide (NO) in response to bradykinin (Kelm & Schrader, 1988; O'Shaughnessy *et al.*, 1992). Moreover, the hypotensive effect of bradykinin injected intravenously has been shown to be abbreviated in rats pretreated with NO-synthase inhibitors, and it was therefore suggested that NO acts as a mediator in the late phase of bradykinin-induced hypotension (Rees *et al.*, 1990). However, we have recently shown that in normotensive rats this abbreviation was dependent on a compensatory adrenergic activation which after NO-synthase inhibitor no longer was counteracted by the continuous NO-production (Bjørnstad-Østensen & Berg, 1994). In spontaneously hypertensive rats, endothelial cell function is impaired or altered as compared to normotensive rats (Dzau, 1989; Burnstock, 1990). It is therefore possible that the interplay between the kinin- and the NO-systems may be different in the two strains of rats.

In the present study, we wished to evaluate the role of endogenous bradykinin in maintaining basal BP in normotensive and spontaneously hypertensive rats. Effects of the endogenous kinin system were either blocked or enhanced by the use of a bradykinin B_2 receptor antagonist or CEI, respectively. To obtain an animal model where the response to CEI

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was induced through kinin only and not through inhibition of angiotensin II formation, the studies were performed on male rats where the renin-angiotensin system had been eliminated by binephrectomy 24 h before the experiments (Ørstavik *et al.*, 1982). The role of NO as a mediator was studied by the use of a NO-synthase inhibitor. In addition, the importance of adrenergic activation in compensating for a kinin-induced fall in BP was evaluated by α_1- and α_2-adrenoceptor blockade.

Methods

Surgical procedures

Male Wistar Kyoto normotensive rats (WKY) and spontaneously hypertensive rats (SHR) (270–350 g body wt.) fed on a conventional rat diet (0.7% NaCl) were anaesthetized (Equthisin; 113 mg kg^{-1} chloralhydrate and 26 mg kg^{-1} pentobarbitone, i.p.), flank incisions were made, and both kidneys were removed. Nephrectomy (Nx) was performed in order to prevent a hypotensive response to CEI due to inhibition of the renin-angiotensin system. Twenty-four hours later, the animals were anaesthetized with pentobarbitone (50 mg kg^{-1}, i.p.) and tracheotomized. Unless otherwise indicated, all drugs were dissolved in phosphate-buffered saline (PBS; 0.01 M Na-phosphate, pH 7.4, 0.14 M NaCl) and administered as bolus injections (20 s, 0.6 ml kg^{-1}) through a catheter in the femoral vein. The catheter was flushed with 0.1 ml PBS after each injection. Arterial BP was recorded throughout the experiment through a heparinized catheter in the femoral artery by a Statham strain-gauge transducer connected to a computer for storage and computation of data. BP-data are presented as mean arterial BP.

Experimental design

Protocol 1: The effect of bradykinin antagonist on basal BP in WKY and SHR rats WKY and SHR rats ($n=6-8$ per group) were nephrectomized, and 24 h later anaesthetized and prepared as described above. BP was recorded before and after intravenous administration of Hoe 140, either as one injection (11.6 μg kg^{-1}) or two injections (116 μg kg^{-1}, 5 min apart). For the latter WKY and SHR group, BP was monitored for another 15 min. The response to the antagonist was compared with that of sham-injection (PBS) controls in protocol 2 (WKY1/SHR1). In addition, the BP-response to Hoe 140 (2×116 μg kg^{-1}, 5 min apart as above) was measured in conscious, nonNx SHR rats 2 h after cannulation of the femoral artery and vein during Equthisin-anaesthesia. The rats were kept in restriction cages after they recovered consciousness and during the experiment. The lower dose of Hoe 140 was chosen in accordance with that used by Wirth *et al.* (1991).

Protocol 2: The effect of L-NAME, bradykinin-antagonist, and phentolamine on the BP-response to CEI in Nx WKY and SHR rats The rats were divided into groups of corresponding WKY and SHR ($n=6-8$ per group), and treated with two injections 5 min apart containing: (1) PBS followed by PBS, respectively (the control groups; WKY1/SHR1), (2) bradykinin antagonist (Hoe 140, 11.6 μg kg^{-1}) and PBS (WKY2/SHR2), (3) PBS and L-NAME (0.3 g kg^{-1}) (WKY3/SHR3), (4) bradykinin antagonist and L-NAME (WKY4/SHR4), or (5) the α_1 and α_2-adrenoceptor antagonist, phentolamine (2 mg kg^{-1}) followed by L-NAME (WKY5/SHR5). Fifteen minutes later all groups received the CEI, captopril (10 mg in 0.5 ml 0.45% NaCl, i.v.). Time-control groups (WKY6/SHR6) were treated with PBS and L-NAME at the same time intervals but did not receive captopril. The doses of L-NAME, phentolamine, and captopril were the same as in previous studies in rats (Ørstavik & Gautvik, 1978; Ørstavik *et al.*, 1982; Bjørnstad-Østensen & Berg, 1994), where they were found to induce an increase in blood pressure, prevent α-adrenergic salivary gland secretion, or induce a kinin-dependent increase in salivary gland blood flow, respectively.

Materials

The following drugs were used: *N*-ω-nitro-L-arginine methyl ester (L-NAME) (Sigma Chemical Co., St. Louis, MO, U.S.A.); phentolamine (Ciba-Geigy, Basel, Switzerland), heparin (Nycomed, Oslo, Norway), D-Arg[Hyp3,Thi5,D-Tic7, Oic8] bradykinin (Hoe 140) (kindly supplied by Hoechst AG, Frankfurt (M), Germany), captopril (a kind gift from Squibb, Princeton, NJ, U.S.A.), pentobarbitone sodium and Equthisin (The National Hospital, Oslo, Norway).

Statistics

The results are expressed as mean ± s.e.mean. To determine if changes in BP in response to the bradykinin antagonist in Protocol 1 were significant, one-sample Student's *t* tests were applied. Group differences in ΔBP were tested by two-sample *t* tests. To determine differences in BP in response to pretreatment in Protocol 2, two-sample *t* tests were used between groups at the same time, and one-sample *t* tests for the difference between BP before versus after pretreatment within groups. To analyze the effect of captopril on changes in BP, Analysis of Variance and Covariance with repeated measures (ANOVA) (BMDP Statistical Software, BMDP Statistical Software Inc., Cork, Ireland) was used, first as over-all tests for all WKY or SHR groups ($P<0.05$), respectively, then for individual groups or between groups. When a development over time was detected for a group, significant changes were located with Student's one-sample *t* tests. The ANOVA-intergroup comparisons were made between the control groups (WKY1/SHR1) and the experimental groups, as well as between the L-NAME-only groups (WKY3/SHR3) and the other L-NAME-treated groups (WKY4-6/SHR4-6). When differences or interactions between groups were detected, Student's two-sample *t* tests were used to locate differences. When more than one comparison was made, the *P*-value limit was corrected by the Bonferroni adjustment.

Results

Protocol 1: The effect of Hoe 140 on basal BP in WKY and SHR rats

The bradykinin B_2-antagonist, Hoe 140, by itself did not induce alterations of basal BP in either normotensive or hypertensive anaesthetized, Nx rats (Table 1). This was also true when the dose of antagonist was increased ten times. We also observed no significant change in BP in response to the high dose of bradykinin antagonist in conscious, nonNx SHR rats (Table 1).

Protocol 2: The effect of L-NAME, bradykinin-antagonist, and phentolamine on the BP-response to CEI in Nx WKY and SHR rats

In SHR, inhibition of NO-synthesis by L-NAME induced a fall in BP followed by a sustained increase in BP, although at the time when captopril was given, the rise in BP had not yet reached its maximum as shown by the L-NAME-time-controls not given captopril (Figure 1, Table 3). ΔBP in response to L-NAME (0–25 min) was in SHR6, +31 ± 5 mmHg ($P<0.0001$). In WKY, the same pattern was observed; however, BP, 25 min after L-NAME, was not different from that prior to L-NAME (WKY6: +2 ± 8 mmHg, NS). Pretreatment with the bradykinin antagonist did not alter the response to L-NAME in either WKY or SHR (WKY3 and 4: ΔBP = 10 ± 8 and 5 ± 10 mmHg, respectively, and 42 ± 4 and 39 ± 4 mmHg in SHR3 and 4, NS). Phentolamine induced a sustained reduction in BP in both WKY and SHR (ΔBP = −38 ± 7 and −37 ± 7 mmHg in WKY5 and SHR5, respectively, $P<0.004$).

Table 1 Changes in BP in response to bradykinin antagonist

Rats		*Drug injected*	n	*ΔBP* (mmHg) *5 min after (1) injection*	*15 min after (2) injection*
WKY Nx	Anaesthetized	PBS	8	1±1	3±1
		Hoe 140 (11.6 $\mu g\,kg^{-1}$)	6	0±1	Not done
		Hoe 140 (116 $\mu g\,kg^{-1}$)	6	−2±0	5±2
SHR Nx	Anaesthetized	PBS	7	2±2	2±2
		Hoe 140 (11.6 $\mu g\,kg^{-1}$)	6	0±1	Not done
		Hoe 140 (116 $\mu g\,kg^{-1}$)	6	−3±2	−1±2
SHR nonNx	Conscious	Hoe 140 (116 $\mu g\,kg^{-1}$)	6	−5±3	0±2

One-sample *t* tests were used to establish the significance of changes in BP (ΔBP) after control injections with vehicle (PBS) or with bradykinin antagonist (Hoe 140) within the Nx WKY or SHR groups, respectively (P value limit = 0.010), and within the nonNx, conscious SHR group (P value limit = 0.025). Two-sample *t* tests were used to establish differences between the control and the two antagonist groups at the same time (P value limit = 0.0167). No significant differences were discovered. *n* = number of rats per group.

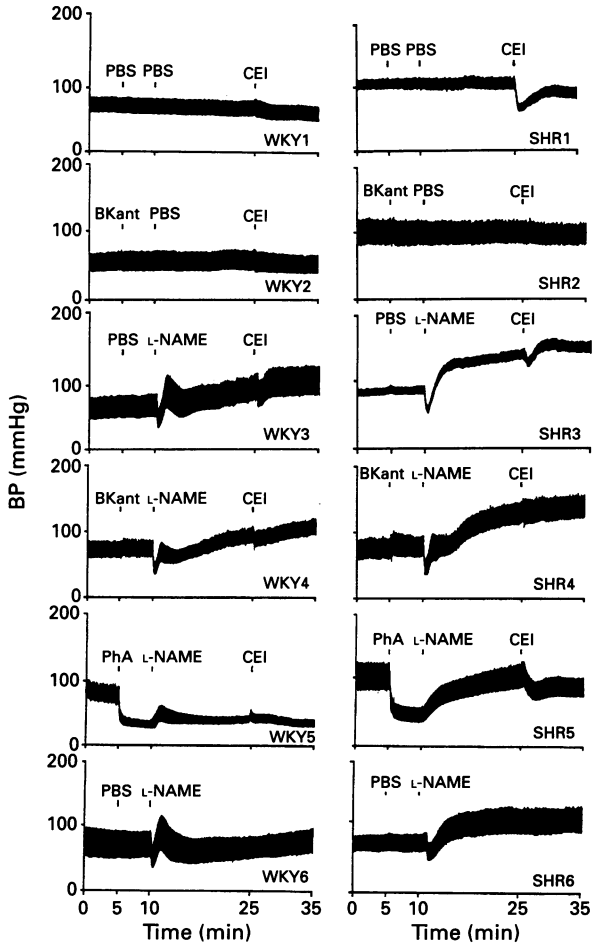

Figure 1 Typical recordings of the BP-response in normotensive (WKY1-6) and hypertensive (SHR1-6) rats pretreated with vehicle (PBS), bradykinin-antagonist (BKant), L-NAME, or phentolamine (PhA), alone or in combination. All groups subsequently received captopril (CEI) except the time control group (WKY6/SHR6) which received L-NAME but not captopril.

The BP values resulting from pretreatment with these drugs alone or in combination, and thus the BP prior to administration of captopril, are given in Table 2. Typical recordings are shown in Figure 1.

Table 3 shows the changes in BP in response to captopril in the WKY and SHR groups. In WKY, no significant change in BP was observed immediately (1 min) after captopril in any of the seven groups (NS). However, 10 min after administration of captopril, an elevated BP was observed in WKY3 and 4 as well as in the L-NAME-time control group not given captopril (WKY6) ($P<0.0007$ compared to WKY1). The changes in BP in the L-NAME-only group (WKY3) and in the bradykinin antagonist + L-NAME group (WKY4) were not different from that of the time control group WKY6) (NS). The rise in BP due to L-NAME during the CEI-observation period was completely abolished by pretreatment with phentolamine (WKY5) ($P<0.0022$ compared to WKY3).

On the other hand, in the SHR1-control group, captopril induced an immediate (within 1 min) fall in BP ($P<0.0017$) which returned towards the preinjection level within the 10 min observation period ($P=0.0029$) (Figure 1, Table 3). After pretreatment with the bradykinin antagonist (SHR2), no change in BP was observed immediately or 10 min after the injection of captopril (NS) ($P<0.0011$ and NS compared to $SHR1_{1\text{ and }10\text{ min}}$, respectively). After pretreatment with L-NAME (SHR3), captopril induced a significant immediate fall in BP ($P=0.0045$) which was lower than, although not significantly different from that of SHR1 ($P=0.12$). After 10 min, BP in SHR was significantly increased ($P<0.0007$) and was higher ($P<0.004$) than that in the L-NAME time control group not given captopril (SHR6) ($P<0.002$). After pretreatment with the bradykinin antagonist in combination with L-NAME (SHR4), no immediate fall was observed (NS) ($P<0.0064$ compared to SHR1 and 3), and the late hypertension was not different from that of the L-NAME time control (NS). $BP_{10\text{ min}}$ in the SHR4 group was also lower than that in the SHR3 L-NAME group ($P<0.0043$). In rats treated with phentolamine and L-NAME (SHR5), the immediate fall in BP in response to captopril ($P<0.0018$) was not different from that in the SHR1-control or the SHR3-L-NAME groups (NS), whereas the late hypertension seen in the L-NAME-only group (SHR3) was abolished ($P<0.0001$ and NS compared to SHR3 and 1, respectively).

Discussion

In the present study we found that the converting enzyme/kininase II inhibitor captopril induced a kinin-dependent hypotensive response in hypertensive but not in normotensive rats. The role of kinin in this response was established by the use of 24 h binephrectomized rats and the bradykinin B_2-receptor antagonist, Hoe 140. Twenty-four hour binephrectomized rats were used in order to eliminate the renin-angiotensin system and thus obtain a response to captopril which would be purely dependent on preventing degradation of endogenous kinins and not through inhibition of angiotensin II-formation. Since the hypotensive effect of an angiotensin II receptor antagonist or renin antibodies was absent

Table 2 BP before and after pretreatment in WKY and SHR rats

			BP (mm Hg)		
Group	n	*Pretreatment*	*Before pretreatment*		*After pretreatment*
WKY1	8	PBS + PBS	63±4		68±3
WKY2	6	BKant + PBS	68±7		70±4
WKY3	7	PBS + L-NAME	66±4		78±10
WKY4	6	BKant + L-NAME	76±8	*	95±15
WKY5	6	PhA + L-NAME	75±9	*	53±6
WKY6	6	PBS + L-NAME	70±7		64±6
SHR1	7	PBS + PBS	84±5		87±5
SHR2	6	BKant + PBS	86±7		86±5
SHR3	6	PBS + L-NAME	81±5	***	122±7**
SHR4	6	BKant + L-NAME	89±10	***	128±8**
SHR5	6	PhA + L-NAME	95±7		89±11
SHR6	8	PBS + L-NAME	77±7	**	101±4

Two-sample *t* tests were used to establish differences in BP prior to administration of captopril (i.e., 15 min after second pretreatment injection) in the WKY and SHR experimental groups as compared to the corresponding control groups (WKY1 and SHR1, respectively) (after values) (*P* value limit = 0.01). Changes in BP before versus after pretreatment within groups were tested with one-sample *t* tests (between values) (*P* value limit = 0.05). For these comparisons, significant differences other than those indicated, were not found. $*P<0.05$, $**P<0.0083$, $***P<0.0008$.

PBS, sham-injection with phosphate-buffered saline; BKant, bradykinin antagonist (Hoe 140, 11.6 $\mu g\,kg^{-1}$); PhA, phentolamine (2 $mg\,kg^{-1}$); L-NAME, NO-synthase inhibitor (0.3 $g\,kg^{-1}$). *n* = number of rats per group.

Table 3 Changes in BP in response to captropril in WKY and SHR rats

			ΔBP (mm Hg) *after captopril*	
Group	*Pretreatment*	n	*1 min*	*10 min*
WKY1	PBS + PBS	8	−4±1	−3±2
WKY2	BKant + PBS	6	−2±1	−2±1
WKY3	PBS + L-NAME	7	−4±2	12±2*†
WKY4	BKant + L-NAME	6	−2±2	10±2*†
WKY5	PhA + L-NAME	6	4±1	0±2‡
WKY6	PBS + L-NAME, no CEI	6	0±0	8±1*†
SHR1	PBS + PBS	7	−23±4*	−11±4
SHR2	BKant + PBS	6	1±1†	−3±1
SHR3	PBS + L-NAME	6	−14±3*	18±2*†
SHR4	BKant + L-NAME	6	1±1†‡	9±2*†‡
SHR5	PhA + L-NAME	6	−11±5*	−12±3*‡
SHR6	PBS + L-NAME, no CEI	8	0±0†‡	6±1*†‡

The table shows the maximum change in BP, which occurred within 1 min, and ΔBP 10 min after administration of captopril in normotensive (WKY1–5) and hypertensive (SHR1–5) rats pretreated as indicated. The time control group (WKY6/SHR6) received PBS + L-NAME but not captopril, and shows the effect of L-NAME alone during the same time interval. Overall tests (ANOVA) indicated differences as well as interactions ($P<0.0001$) within both the WKY and SHR groups. A development over time was observed for WKY3, 4 and 6 ($P<0.0001$) and for SHR1, 3–6 ($P<0.0027$) (*P* value limit = 0.0083). For these groups, one-sample *t* tests located significant changes in BP as indicated (*) (*P* value limit = 0.025). When ANOVA intergroup comparisons between the control groups (WKY1/SHR1) and the experimental groups (†), as well as between the L-NAME-only group (WKY3/SHR3) and the other L-NAME-treated groups (WKY4–6/SHR4–6) (‡) had revealed significant differences or interaction (*P* value limit = 0.0063), differences were located at 1 and 10 min as indicated by two-sample *t* tests (†, ‡ < *P* value limit = 0.025). For these comparisons, significant differences other than those indicated, were not found.

PBS, sham-injection with phosphate-buffered saline; BKant, bradykinin antagonist (Hoe 140, 11.6 $\mu g\,kg^{-1}$); PhA, phentolamine (2 $mg\,kg^{-1}$); L-NAME, NO-synthase inhibitor (0.3 $g\,kg^{-1}$). *n* = number of rats per group.

after nephrectomy in SHR rats (Inagami *et al.*, 1991), the lack of any residual response to captopril after pretreatment with a bradykinin antagonist not only confirmed the role of kinin in the observed hypotensive response to captopril, but also showed that elimination of the renin-angiotensin system had indeed been achieved. The present results are in complete agreement with studies performed on non-Nx, anaesthetized rats where the hypotensive effect of captopril was partially reduced by a kinin antagonist in spontaneously hypertensive but not normotensive rats (Cachofeira *et al.*, 1992).

Since captopril did not induce any response in normotensive but a clear hypotension of −23±4 mmHg in hypertensive Nx rats, a response which was totally abolished by pretreatment with the bradykinin antagonist, we concluded that endogenous bradykinin-production was enhanced in hypertensive but not in normotensive rats. Tissue kallikrein originating from organs other than the kidneys such as the submandibular glands (Berg *et al.*, 1985) or kallikrein located within the blood vessels (Saed *et al.*, 1990), may be responsible for this kinin-production. However, since we observed no effect on basal BP upon administration of the bradykinin receptor antagonist either in anaesthetized, Nx WKY and SHR rats or in conscious, nonNx SHR rats, it could also be concluded that this endogenous bradykinin did not play a role in BP homeostasis in the basal condition, or that the basal concentration of bradykinin in SHR was not high enough to influence the BP unless its breakdown was prevented by captopril. The physiological or pathological situations in which this endogenous bradykinin might play a role, or if there may be physiological conditions in which endogenous CEI(s) may produce a sufficiently elevated

concentration of bradykinin, are not clear. Modulation of a hypertensive response is one possibility; however, hypertension induced by inhibition of NO-synthesis did not appear to be one such condition since pretreatment with the bradykinin antagonist did not alter the hypertensive response to L-NAME. The lack of response to bradykinin antagonist is not in accordance with observations made by Benetos *et al.* (1986b) on nonNx, normotensive rats where a hypertensive response to the Arg-Pro-Hyp-Gly-Thi-Ser-DPhe-Thi-Arg.trifluoroacetic acid bradykinin antagonist was observed. However, Cachofeira *et al.* (1992) failed to observe a hypertensive response to the same antagonist, and we found no effect by increasing the concentration of bradykinin antagonist 10 times, as was done in the study of Benetos *et al.* (1986b) or by using nonNx, conscious SHR rats.

It has been suggested that the hypotensive effect of bradykinin is mediated through the formation of NO. However, this concept has been challenged by our previous study on nonNx normotensive rats (Bjørnstad-Østensen & Berg, 1994): As in other studies (Whittle *et al.*, 1989; Rees *et al.*, 1990), we found that the acute hypotension following an i.v. injection of bradykinin was not attenuated by the NO-synthase inhibitor, L-NAME, whereas the duration of the hypotensive response was abbreviated. However, this abbreviation did not indicate that NO was a mediator for bradykinin but was explained by a compensatory adrenergic response following the acute bradykinin-induced fall in BP, which after L-NAME no longer was counteracted by a continuous NO-release. In the present study L-NAME showed a tendency to reduce the hypotensive effect of captopril. However, this effect was not statistically significant, and the participation of the NO-system as a mediator for bradykinin was therefore highly questionable. The mediator responsible for he hypotensive effect of bradykinin is still not known, however, vasodilator prostanoids and endothelial hyperpolarizing factor (EDHF), as well as NO, have been suggested (for review, see Mombuli & Vanhoutte, 1995).

However, the starting BP in SHR was observed to be in the normotensive range in the present study. This was due to the use of anaesthetized, Nx rats. When BP was measured in conscious SHR rats, BP 2 or 24 h after Nx was almost as high as before Nx. However, on subsequent anaesthesia, BP was reduced to the same levels as observed in the present study, whereas an equally low BP was not observed in anaesthetized, nonNx SHR rats (T. Berg, unpublished observations). In a recent study, we have shown that NO is a partial mediator of bradykinin-induced hypotension in anaesthetized, nonNx SHR but not WKY rats (T. Berg, unpublished observations). Furthermore, these SHR rats were more sensitive to bradykinin than normotensive rats but not when the BP in the SHR rats was lowered by preadministration of phentolamine. Participation of NO in mediating the hypotensive response to bradykinin may therefore be a pressure-induced mechanism, and may explain why under the present conditions with anaesthetized, Nx SHR rats with normotensive BP, L-NAME did not attenuate the hypotensive response to captopril. Moreover, it may also seem that in hypertensive animals, mechanisms to oppose the high blood pressure may be activated such as the generation of endogenous bradykinin, as demonstrated in the present study, as well as additional activation of mediators, such as NO, to amplify its response. Evidently, the kinin-production was not as quickly 'turned off' as the NO-mediator mechanism since the former was observed after the BP was reduced by the anaesthesia, whereas the latter was not.

L-NAME induced first a fall in BP followed by a rise of BP. In SHR, the rise in BP reached hypertensive levels, whereas in WKY, BP stabilized at a level not significantly different from that prior to administration of L-NAME. The same was observed when the observation period for L-NAME was prolonged to 45 min (T. Berg, unpublished observations). The lack of a hypertensive response in WKY rats was due to the use of anaesthetized, Nx rats, since we have previously shown that the same dose of L-NAME raised BP from 119 ± 4 to 159 ± 5 mmHg in nonNx WKY rats (Bjørnstad-Østensen & Berg, 1994), and preliminary results indicate that conscious Nx WKY rats respond to L-NAME with hypertension (T. Berg, unpublished observations). The mechanism for this difference is not clear, since it is not observed in SHR rats.

To ensure that the kinin antagonist was active throughout the experimental period, captopril was administered before the hypertensive response to L-NAME was fully developed. However, to correct for variations in BP due to L-NAME during the observation period for captopril, an L-NAME-only, without captopril, time-control was included. By doing so, it could be concluded that the late hypertensive response observed in rats pretreated with L-NAME could in part be ascribed to a continuous hypertensive response to L-NAME itself which at the time when captopril was given, had not yet reached its maximum. Moreover, in the group pretreated with the bradykinin antagonist as well as L-NAME, the acute hypotensive response to captopril was abolished and the late hypertensive response was similar to that in the L-NAME time control. On the other hand, the hypertensive response in the L-NAME group without bradykinin antagonist was stronger than that seen in both the time-control and the bradykinin antagonist + L-NAME group. This increased hypertensive response appeared to represent an adrenergic activation induced by the captopril-kinin-dependent fall in BP since in rats pretreated with the α_1 and α_2-adrenoceptor blocker, phentolamine, in addition to L-NAME, BP remained low as in the control group. Thus, as in normotensive nonNx rats receiving exogenous bradykinin (Bjørnstad-Østensen & Berg, 1994), the captopril-induced hypotension activated an adrenergic response which without L-NAME was counteracted by a continuous NO-production. And *vice versa*, when the continuous homeostatic balance between the adrenergic and the NO-system was interrupted by L-NAME, the adrenergic component was disclosed.

In conclusion, the present results show that captopril induced a hypotensive response in spontaneously hypertensive rats but not in normotensive rats. Due to the elimination of the renin-angiotensin system by running the experiments in Nx rats, this response was totally dependent on kinins as was also shown by the use of a bradykinin receptor antagonist. The kinin-dependent hypotensive response was not mediated through activation of NO-production, probably due to the low BP in Nx, anaesthetized SHR rats. However, the hypotension activated an adrenergic compensatory response which was visualized when the counter-balancing NO-system was blocked. From the present results it could be deduced that endogenous kinin-production has a bearing on BP-regulation in hypertensive and not in normotensive rats. However, this kinin-production did not, under the present conditions, play a role in the BP-balance in the basal condition or in modulating the hypertensive response induced by inhibition of NO-synthesis.

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