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Pentaerythrityl tetranitrate attenuates structural changes in conduit arteries evoked by long-term NO-synthase inhibition

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1 The aim of the study was to determine whether the pentaerythrityl tetranitrate (PETN), a tolerance devoid exogenous NO donor could prevent morphological changes in the cardiovascular system evoked by long-term NO-synthase inhibition.

2 Three groups of 10-week-old Wistar rats were used: (1) controls, (2) treated by L-N^G-nitroarginine methyl ester (L-NAME) in water (50 mg kg^{-1}), and (3) treated by L-NAME (50 mg kg^{-1} in water) + PETN (2×50 mg kg^{-1}, using gavage). Blood pressure (BP) was measured by the tail plethysmographic method.

3 After sacrificing the animals were perfused (120 mmHg) by glutaraldehyde fixative and processed according to standard electron microscopy procedure. Wall thickness (WT), cross sectional area (CSA), inner diameter (ID) of thoracic aorta (TA), carotid (CA) and septal branch of the left descending coronary artery (RS) were measured in light microscopy.

4 After 6 weeks, the BP was increased to 172 ± 1.7 mmHg ($P<0.01$) in the L-NAME group, compared to 127 ± 1.4 mmHg in controls. In L-NAME + PETN-treated rats, BP was 163 ± 0.9 mmHg ($P<0.01$), and significantly lower ($P<0.01$) in comparison to L-NAME-treated rats. Heart weight and heart/body weight ratio was not significantly changed.

5 In L-NAME-treated rats, both WT and CSA were increased in all three arteries ($P<0.01$). ID was increased only in TA ($P<0.01$). Wall/diameter ratio (WD) was increased in TA ($P<0.01$) and CA ($P<0.01$). In L-NAME + PETN treated rats, WT was found to be increased only in TA ($P<0.01$). In comparison to the L-NAME treated group, WT was decreased in TA ($P<0.01$), in CA ($P<0.01$), in RS ($P<0.05$). CSA was increased only in TA ($P<0.01$), yet in comparison to the L-NAME group it was decreased in CA ($P<0.01$). ID was increased in comparison to both control and L-NAME treated rats only in TA ($P<0.01$). WD did not differ from the control value. In comparison to L-NAME-treated rats, it was decreased in both TA and CA ($P<0.01$).

6 These data suggest that the changes in the cardiovascular system evoked by long-term NO-synthase inhibition were attenuated by simultaneous administration of the exogenous donor of nitric oxide – PETN

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Keywords: Pentaerythrityl tetranitrate; nitric oxide; L-NAME; arterial wall; morphometry; hypertension

Abbreviations: BP, blood pressure; CA, carotid artery; cyclic GMP, cyclic guanosine monophosphate; CSA, cross sectional area; ID, inner diameter; L-NAME, L-N^G-nitroarginine methyl ester; NO, nitric oxide; OsO_4, osmium tetroxide; PETN, pentaerythrityl tetranitrate; RS, septal branch (ramus septalis) of the left descending coronary artery; TA, thoracic aorta; WD, wall/diameter ratio; WT, wall thickness

Introduction

Administration of exogenous donors of nitric oxide as a remedy for angina pectoris is more than a hundred years old (Murrel, 1879), though without understanding the chain of events leading to improvement. The NO-donating properties of organic nitrates were discovered only recently (Ahlner *et al*., 1991). Nitrates are metabolically converted to NO inside the smooth muscle cell, thus producing endothelium independent vasodilatation. The limiting factor of nitrates is the development of tolerance to their haemodynamic and anti-ischaemic effects (Zanzinger *et al*., 1998). Unfortunately, nitrate tolerance is a complex phenomenon composed of many factors, which in some nitrovasodilators, are induced as early as within 2 to 3 days of non-intermittent therapy. Moreover, long-term administration of glyceryl trinitrate is associated with enhanced superoxide production in the blood vessel wall (Münzel *et al*., 1995). In spite of these problems, the significance of NO in the cardiovascular system accelerated research in new therapeutic possibilities of NO supplementation. In recent years, clinical interest has been focused on tolerance devoid organic nitrates.

Pentaerythrityl tetranitrate (PETN) represents an effective tolerance devoid NO donor with a pharmacodynamically beneficial long-term effect without inducing oxidative stress (Dück & Richard, 1990; Fink & Bassenge, 1997; Hinz *et al*., 1998) and moreover, it may have a protecting effect against atherosclerosis and endothelial dysfunction (Kojda *et al*., 1995). PETN was found to be the most potent activator of cyclic GMP synthesis compared to other clinically used organic nitrates (Hinz *et al*., 1998). The pharmacologically active phase I is predominantly responsible for the long-lasting vasodilatory and anti-ischaemic effect of PETN (Hinz *et al*., 1998). In spite of these findings the complex pharmacodynamics of PETN is unknown. Moreover, data concerning the long-term effect of PETN on structural alterations of the arterial wall induced by hypertension are completely missing.

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In the present study, we tested the long-term effect of PETN on the arterial wall in the animal model of NO deficient hypertension. The pathophysiological background of this type of hypertension is inhibition of NO synthase, an enzyme metabolizing L-arginine to L-citruline with coproduction of NO. As inhibitor of NO-synthesis N^G-nitro-L-arginine methyl ester (L-NAME) is often used (Rees *et al*., 1990; Delacretaz *et al*., 1994). Besides the remarkable increase in blood pressure, the subsequent decreased basal production of NO resulted also in a pronounced increase in wall thickness of conduit arteries (Delacretaz *et al*., 1994; Kristek & Gerová, 1996). Having in mind Laplace's law it is evident that changes in arterial geometry result in negative consequences on the cardiovascular system. Moreover, increased blood pressure along with increased contractility of smooth muscle cells (Ribeiro *et al*., 1995; Holécyová *et al*., 1996) induced by NO-synthase blockade could exaggerate this effect. Since it is generally accepted that NO inhibits cell proliferation (Garg & Hassid, 1989; Nakaki *et al*., 1990; Arnal *et al*., 1994; Cornwell *et al*., 1994), we supposed that NO liberated from PETN could have an antiproliferative effect on smooth muscle cells of the arterial wall.

The aim of this study was to determine (i) morphological changes in the cardiovascular system in NO-deficient hypertension and (ii) whether simultaneous long-term administration of the NO-synthase blocker L-NAME along with the exogenous donor NO–PETN can prevent the development of structural changes in the arterial wall of the thoracic aorta, carotid artery and coronary artery.

Methods

The procedures followed the guidelines presented in the Guide for the Use of Laboratory Animals (Ethics Committee for Experimental Work, Slovak Academy of Sciences, 1995). The animals were housed at a temperature of 22–24°C, in individual cages under a 12 h light:dark cycle and fed a regular pellet diet.

Male Wistar strain rats weighing 310–320 g were taken for the study. The animals were divided into three groups, of 10 animals each. At the beginning of the experiment the animals were 10 weeks old. Control rats were given tap drinking water. One group of experimental animals were given L-NAME into drinking water at a concentration of 50 mg kg^{-1} b.w. day^{-1} for 6 weeks. The second group of experimental animals were given L-NAME (50 mg kg^{-1} b.w. day^{-1}) into drinking water and they received PETN in tap water, given p.o. by gavage at a concentration of 50 mg kg^{-1} twice daily over the same 6 week period (in the morning and in the afternoon), in the total daily concentration of 100 mg kg^{-1}. In all groups systolic blood pressure and heart rate were measured indirectly in pre-warmed rats by the tail plethysmographic method each week.

At the end of the experiment (after 6 weeks), the animals were killed by an overdose of pentobarbitone (100 mg kg^{-1} b.w., i.p.), the chest was opened and the cardiovascular system perfused at a constant pressure of 120 mmHg for 10 min *via* a cannula placed in the left ventricle. As a fixative, 3% glutaraldehyde in 0.1 M phosphate buffer was used. The middle part of the thoracic aorta, the middle part of the carotid artery and the upper part of the septal branch of the left descending coronary artery were excised. The arteries were cleaned and divided into four segments about 1 mm long and post-fixed with 2% OsO_4 in 0.1 M phosphate buffer. After fixation, the specimens were stained *en bloc* with 2% uranyl acetate, dehydrated through ascending concentration of alcohol and embedded in Durcupan ACM. Three randomly selected blocks of each artery were cut perpendicularly to the long axis. Both inner circumference and arterial wall thickness (tunica intima and tunica media) were measured in light microscopy. The arterial wall thickness was measured at about 45° intervals around the vessel circumference. The inner diameter and the cross section area (tunica intima and tunica media) were calculated. Values are given as mean ± s.e.mean. Anova and Bonferroni test for unpaired variables were used for statistical evaluation. Results were considered significantly different when $P<0.05$.

Results

The mean systolic blood pressure of control rats was 127 ± 1.4 mmHg at the end of experiments (16-week-old animals). In age-matched L-NAME-treated rats the blood pressure gradually increased to 172 ± 1.7 mmHg ($P<0.01$). In rats concomitantly administered L-NAME and PETN, the onset of blood pressure elevation was shifted to the right (Figure 1) and at the end of the experiment represented 163 ± 0.9 mmHg. It was significantly lower than in L-NAME-administered rats ($P<0.01$) and significantly higher than in control rats ($P<0.01$).

At the end of the experiment, the heart rate was 374 ± 11.6 beats min^{-1}, in the control group, 280 ± 12.9 beats min^{-1} ($P<0.01$) in L-NAME-treated rats, and 403 ± 15.7 beats min^{-1} in L-NAME plus PETN-treated rats, which was significantly higher ($P<0.01$) than in the L-NAME-treated group. There was no significant difference between the control group and L-NAME plus PETN-treated rats (Figure 2).

There were no significant differences in heart weight in the groups studied. In the control group heart weight was 1.35 ± 0.03 g, in L-NAME-treated animals 1.35 ± 0.04 g, and in L-NAME plus PETN-treated rats 1.36 ± 0.08 g (Figure 3).

The heart/body weight ratio was $3.19 \pm 0.03 \times 10^{-3}$ in the control group, $2.88 \pm 0.13 \times 10^{-3}$ in L-NAME-treated rats, and $3.18 \pm 0.23 \times 10^{-3}$ in L-NAME plus PETN-treated rats. No significant difference was observed among the groups (Figure 3).

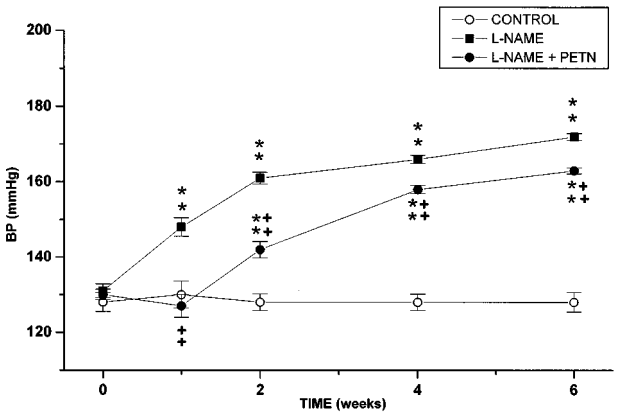

Figure 1 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME), and L-NAME along with pentaerythrityl tetranitrate administration on blood pressure in rats. $**P<0.01$ with respect to the value of the control group, $++P<0.01$ with respect to the value of the L-NAME-administered group.

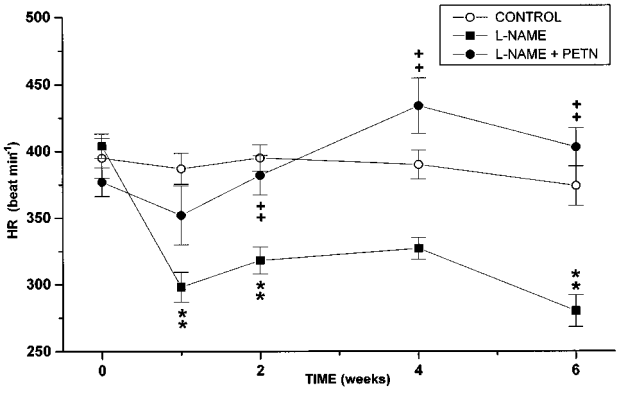

Figure 2 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME), and L-NAME along with pentaerythrityl tetranitrate administration on the heart rate of rats. $^{**}P<0.01$ with respect to the value of the control group, $++P<0.01$ with respect to the value of the L-NAME administered group.

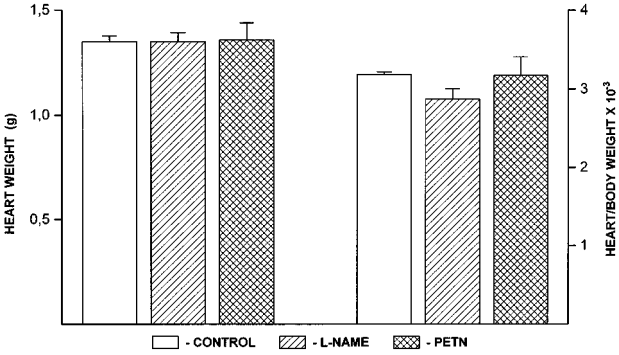

Figure 3 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on heart weight and heart/body weight ratio in rats.

Arterial parameters

Morphometric analysis of the arterial wall (tunica intima + tunica media) of the thoracic aorta, carotid artery, and septal branch of the left descending coronary artery yielded the following data.

Thoracic aorta Arterial wall thickness of 60.41 ± 2.37 μm in control rats was significantly increased in L-NAME-treated rats (80.42 ± 1.30 μm, $P<0.01$), while simultaneous administration of L-NAME and PETN to rats resulted in significantly lower arterial wall thickness ($70{,}73 \pm 1.60$ μm, $P<0.01$), while it was however still significantly higher than in control rats ($P<0.01$) (Figure 4).

Since the wall thickness could be influenced by the perfusion pressure during fixation, we evaluated also the cross section area of the arterial wall. The calculated wall area of $331{,}600 \pm 11{,}600$ μm^2 in control rats was significantly increased both in L-NAME ($479{,}600 \pm 9760$ μm^2, $P<0.01$) and in L-NAME + PETN-($453{,}700 \pm 11{,}900$ μm^2, $P<0.01$) treated rats. No significant difference between the L-NAME and L-NAME + PETN groups was found (Figure 4).

The inner diameter of the aorta was significantly changed ($P<0.01$) among the groups. In control rats it was 1683 ± 39.66 μm, in L-NAME rats 1816 ± 25.53 μm, and in L-NAME + PETN-treated rats it was 1971 ± 20.77 μm (Figure 5)

Changes in both arterial wall thickness and inner diameter of the arteries resulted in significantly increased wall/diameter ratio ($P<0.01$) between L-NAME-treated rats ($4.43 \pm 0.10 \times 10^{-2}$)

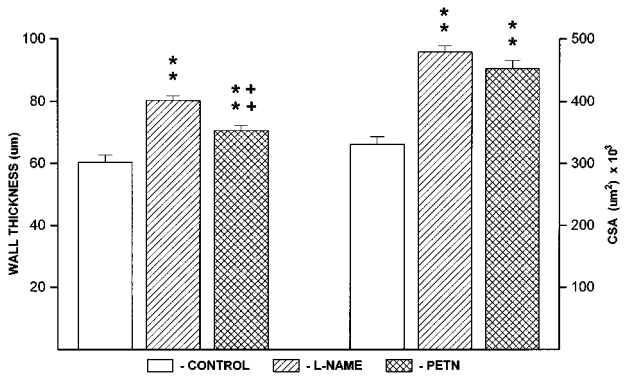

Figure 4 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on the wall thickness and cross section area of the thoracic aorta in rats. $^{**}P<0.01$ with respect to the value of the control group, $++P<0.01$ with respect to the value of the L-NAME-administered group.

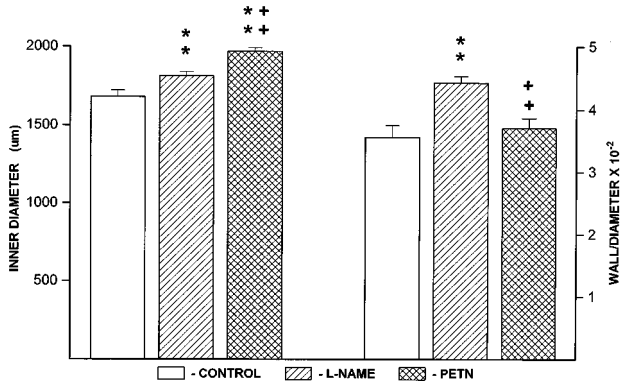

Figure 5 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on the wall thickness and cross section area of the carotid artery in rats. $^{**}P<0.01$ with respect to the value of the control group, $++P<0.01$ with respect to the value of the L-NAME-administered group.

on the one side and the control group ($3.56 \pm 0.19 \times 10^{-2}$) and L-NAME + PETN-treated rats ($3.71 \pm 0.15 \times 10^{-2}$) on the other side. There was no significant difference between control rats and L-NAME plus PETN-administered rats (Figure 5).

Carotid artery The thickness of the arterial wall was 23.95 ± 1.76 μm, in control rats, in L-NAME-administered rats it was increased (36.42 ± 1.33 μm, $P<0.01$), and in L-NAME + PETN-treated animals it was significantly lower than in the L-NAME group (22.69 ± 1.47 μm, $P<0.01$). No difference was observed between L-NAME + PETN-treated rats and control rats (Figure 6).

Similarly as in wall thickness, differences were observed among the groups in cross section areas. The area of arterial wall in control rats was $59{,}500 \pm 3060$ μm^2, in L-NAME rats it was increased to $94{,}400 \pm 4040$ μm^2 ($P<0.01$), and in L-NAME + PETN-($63{,}000 \pm 2650$ μm^2) treated rats it was significantly lower than in the L-NAME group ($P<0.01$), while no difference was observed between L-NAME + PETN and control rats (Figure 6).

The inner diameter did not differ among the groups. It was 778 ± 33.9 μm in control rats, 818 ± 20.27 μm in the L-NAME group, and 876 ± 26.69 μm in L-NAME + PETN rats (Figure 7).

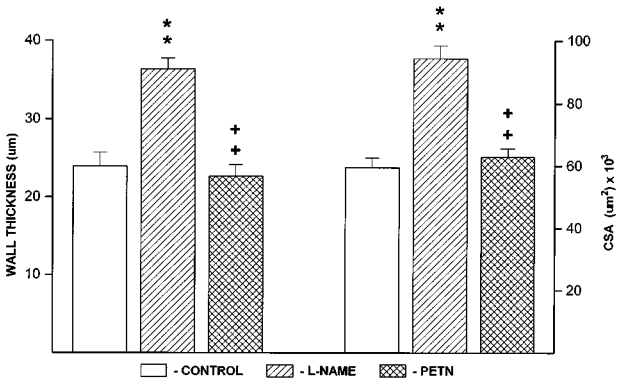

Figure 6 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on the wall thickness and cross section area of the septal branch in left descending coronary artery in rats. $^{**}P<0.01$ with respect to the value of the control group, $++P<0.01$ with respect to the value of the L-NAME administered group.

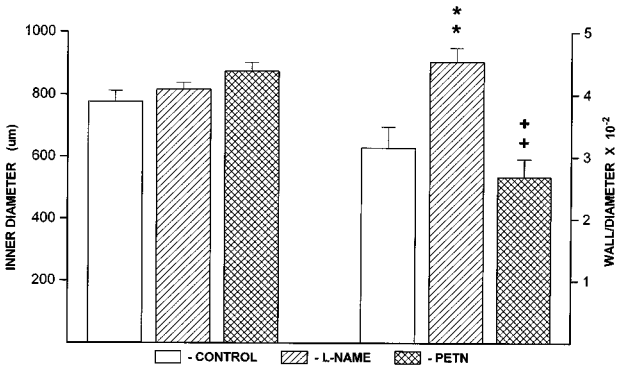

Figure 7 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on the inner diameter and wall/diameter ratio of the thoracic aorta in rats. $^{**}P<0.01$ with respect to the value of the control group, $++P<0.01$ with respect to the value of the L-NAME-administered group.

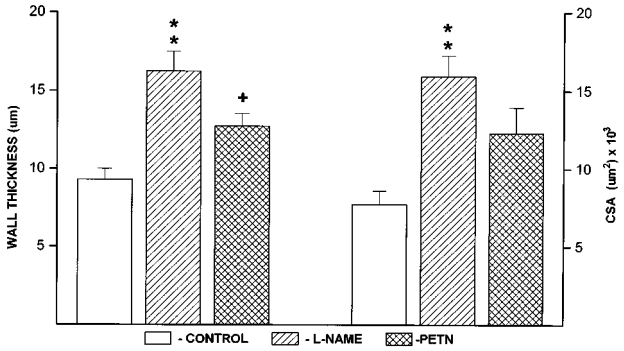

Figure 8 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on the inner diameter and wall/diameter ratio of the carotid artery in rats. $^{**}P<0.01$ with respect to the value of the control group, $+P<0.05$ with respect to the value of the L-NAME-administered group.

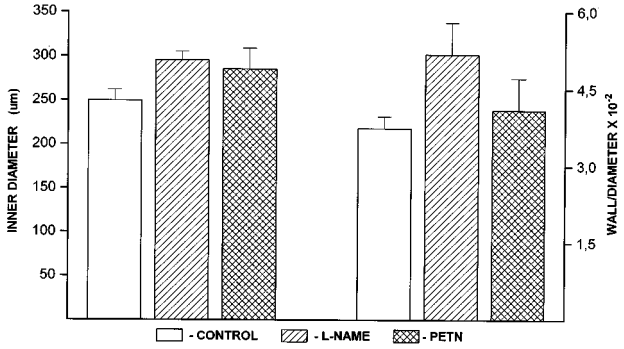

Figure 9 Long-term effect of L-N^G-nitroarginine methyl ester (L-NAME) treatment and L-NAME along with PETN administration on the inner diameter and wall/diameter ratio of the septal branch of the left descending coronary artery in rats.

The wall/diameter ratio was $3.16\pm0.33\times10^{-2}$ in control rats. In L-NAME rats it was $4.53\pm0.22\times10^{-2}$ significantly higher than in control group ($P<0.01$), in L-NAME+PETN rats it was significantly decreased to $2.68\pm0.28\times10^{-2}$ ($P<0.01$). There were no significant differences between the latter group and control rats (Figure 7).

Septal branch of the left descending coronary artery The wall thickness 9.33 ± 0.67 μm of the control artery increased significantly to 16.28 ± 1.24 μm ($P<0.01$) in L-NAME-treated rats. In L-NAME plus PETN-treated rats the wall thickness was 12.76 ± 0.79 μm, which was significantly lower ($P<0.05$) than in L-NAME-treated rats. No differences were observed in comparison to control rats (Figure 8).

The calculated arterial wall area of 7750 ± 840 μm^2 increased to $16{,}000\pm1315$ μm^2 in L-NAME-administered rats ($P<0.01$). In the group of rats treated with both L-NAME and PETN, the area was $12{,}300\pm1650$ μm^2. The difference in arterial wall area between the L-NAME plus PETN group and control rats was not significant (Figure 8).

The inner diameter of the arteries did not significantly change among the groups. In the control artery the inner diameter was 250 ± 11.88 μm, in the L-NAME-treated artery it was 295 ± 9.12 μm, and in the L-NAME plus PETN group the inner diameter was 286 ± 23.16 μm (Figure 9).

Furthermore, the wall/diameter ratio of the arteries did not differ significantly among the groups. In the control artery it represented $3.70\pm0.23\times10^{-2}$, in L-NAME-treated rats it was $5.19\pm0.410\times10^{-2}$, and in rats administered L-NAME together with PETN it was $4.10\pm0.62\times10^{-2}$ (Figure 9).

Discussion

In the present study, we investigated the effect of the exogenous NO donor PETN on morphological changes in the cardiovascular system evoked by NO deficient hypertension. Long-term administration of the NO synthase inhibitor L-NAME resulted in sustained significant elevation of blood pressure. These results are fully consistent with our previous findings (Kristek *et al*., 1995; Kristek & Gerová, 1996) and observations of other authors with a similar length of L-NAME administration (Baylis *et al*., 1992; Ribeiro *et al*., 1992; Jover *et al*., 1993). Simultaneous non-intermittent long-lasting administration of PETN and NO synthase inhibitor prevented partially the increase in blood pressure. In this group the onset of blood pressure elevation was shifted to the right, and although there was a significant increase in blood pressure at the end of the experiment (after 6 weeks), but this increase was significantly lower in comparison to the rats administered NO

synthase inhibitor alone. The less enhanced blood pressure was most probably due to activation of guanylyl cyclase by NO liberated from PETN. Activation of soluble guanylate cyclase results in conversion of guanosine triphosphate to the second messenger cyclic guanosine monophosphate (Mittal & Murad, 1982). As shown by Schultz *et al*. (1997) and Hinz *et al*. (1998), PETN proved to be the most potent compound stimulating cyclic GMP in comparison to the other clinically relevant organic nitrates (glyceryl trinitrate, isosorbide dinitrate, isosorbide-5-mononitrate). Moreover, PETN is an NO donor devoid of tolerance (Kojda *et al*., 1995; Fink & Bassange, 1997; Hinz *et al*., 1998).

Surprisingly, in contrast to what was found in the other experimental models of hypertension, we did not observe changes in either cardiac weight or heart/body weight ratio in either group studied. Cardiac hypertrophy usually accompanies increase of blood pressure. In NO deficient hypertension there are some discrepancies between different reports concerning cardiac hypertrophy. Some authors, including our previous findings, observed cardiac hypertrophy (Bernátová *et al*., 1996; Kristek & Gerová, 1996), others found that long-lasting NO synthase inhibition was not accompanied by cardiac hypertrophy (Arnal *et al*., 1993; Deng *et al*., 1993) or even that L-NAME may exert some direct antiproliferative actions (Zatz & Baylis, 1998). To date the reason for these conflicting findings is not known. Differences in the breeding of animals (our findings), length of administration or dose of L-NAME may account partially though not completely for the discording data, yet other mechanisms must also be involved.

Long-term administration of the NO synthase inhibitor resulted in a remarkable increase of arterial wall thickness. This observation is in agreement with our previous findings (Kristek & Gerová, 1996) on the coronary and carotid artery, and with findings of Delacretaz *et al*. (1994) on the carotid artery, Morton *et al*. (1993) and Deng *et al*. (1993) on the mesenteric vascular bed. But on the other hand, Dunn & Wilson (1993) did not find any changes in either conduit arteries or resistant mesenteric vessels. An increase in arterial wall thickness in NO deficiency seems to be in good agreement with findings of Garg & Hassid (1989) and Nakaki *et al*. (1990), Arnal *et al*. (1994) and Cornwell *et al*. (1994), who reported that NO inhibited the proliferation and mitogenesis of smooth muscle cells and thus it could be expected that a decrease of NO level due to NO synthase blockade would exert an opposite effect. A significant decrease of NO level after L-NAME administration is generally accepted (Gerova *et al*., 1998; Rees *et al*., 1990). Nevertheless, structural adaptation of the arterial wall in hypertension is a complex process and we suppose that a summation of various impulses is operative synergically in the regulation of arterial wall thickness. It is likely that besides the decrease of NO also blood pressure plays an important role. Since in spontaneously hypertensive rats increase in arterial wall thickness is present before blood pressure elevation (Lee, 1985) the role of blood pressure is not necessarilly dominant in pathological remodelling. NO influences various processes in the cardiovascular system, thus changes in the activity of the renin angiotensin system (Auch-Schwelk *et al*., 1993), bradykinin level (Hecker *et al*., 1994), endothelins (Rizvi & Meyers, 1997), apoptosis (Brüne *et al*., 1998), etc, should also be considered. Besides the cellular component of the arterial wall (endothelial and muscle cells), the extracellular matrix could also play an important role in both arterial wall mass and in the elasticity of the arterial wall (Kristek *et al*., 1996; Rizvi & Myers, 1997). The question which stimulus is predominantly operative remains however open.

Long-term administration of L-NAME along with PETN resulted in decrease of arterial wall thickness in all three arteries studied. The cross section area of both carotid and coronary arteries did not differ from control values. To our knowledge, there are no morphological data available for comparison of our findings. An indirect comparison presents itself with our study (Kristek & Gerova, 1998), based on L-NAME plus molsidomine administration to rats over the same period (6 weeks), which showed a significant decrease of arterial wall thickness in the thoracic aorta, carotid artery, and septal branch of the left descending coronary artery. The main beneficial effect of PETN in NO deficiency could be reasonably explained by NO liberation from the exogenous donors and its inhibitory and antiproliferative effect on smooth muscle cells (Garg & Hassid, 1989; Arnal *et al*., 1994; Cornwell *et al*., 1994). The question why PETN did not affect all arteries to the same extent remains however unanswered. Compared to L-NAME-treated rats, in L-NAME plus PETN-treated rats we observed in the thoracic aorta a significant decrease in arterial wall thickness but not in cross section area. The fact that PETN is metabolized through numerous metabolites with various (largely unknown) effects on the arterial wall could explain the differences in the effect of PETN on various types of arteries.

In L-NAME-treated rats, the inner diameter was significantly increased only in the thoracic aorta. No significant differences were observed in either the carotid or coronary artery. Our findings on the carotid artery are in good agreement with observations of Delacretaz *et al*. (1994) who did not find changes in the inner diameter of the carotid artery after L-NAME administration.

One can speculate that an increase in cyclic GMP synthesis (due to PETN administration) should result in dilation of conduit arteries in comparison to L-NAME-treated rats. In spite of this presumption, a significant increase in the inner diameter was observed only in the thoracic aorta. It was not found either in the carotid artery or in the septal branch of the left descending coronary artery, though in the carotid artery the difference was close to significance. After 6 days of continual PETN infusion, Fink & Bassenge (1997) observed a stable increase in coronary diameter of about 10%. The discrepancy between our and their results could be due to different species (dog), different artery (left circumflex coronary artery), different duration of treatment (6 days). The simultaneous administration of the exogenous donor along with the NO synthase inhibitor could also have come into play. The not well marked changes in the inner diameter support the observation that the vasodilatory activity of nitrovasodilators is exerted preferentially in the venous part of the cardiovascular system (Ahlner *et al*., 1991).

After L-NAME administration, the wall/diameter ratio was significantly increased in both the thoracic aorta and carotid artery. An increasing trend was also observed in the coronary artery, but the difference was not significant. In the group treated by L-NAME along with PETN the wall/diameter ratio was significantly decreased in comparison to L-NAME-treated rats in both the thoracic aorta and carotid artery. It tended to be decreased also in the coronary artery. No significant differences were observed in comparison to control rats in either of the three arteries. We suppose that these not well marked results concerning the septal branch of the left descending coronary artery may be caused by its architecture, which is not uniform in individual rats. The variability in arterial branching may result in an increased requirement on statistical significance.

It should be stressed that proximal conduit arteries and especially the aorta, have a considerable haemodynamic importance as a 'Windkessel' function, which transforms the

rhythmic cardiac output towards the periphery, and also as a site for various sets of arterial baroreceptors (Folkow, 1995). The significance of changes in the geometry of the arterial wall is expressed in Laplace's law. It is thus clear that any changes in the arterial geometry (inner diameter, wall thickness, wall/diameter ratio) exert a negative effect on physiological parameters of the cardiovascular system (wall tension, transmural pressure). Attenuation of structural changes of the arterial wall in NO-deficiency caused by PETN administration results conceivably in functional consequences leading very probably to improved supply of nutritional demand of the respective areas.

In summary, L-NAME administration to rats 6 weeks lasting resulted in sustained increase of blood pressure and pronounced morphological changes in conduit arteries. Simultaneous supplementation of NO in the form of the exogenous NO donor PETN during this period attenuated blood pressure elevation, decreased the heart rate, and improved morphological changes in the thoracic aorta, carotid artery, and coronary artery evoked by NO synthase inhibition.

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