High-calcium vs high-phosphate intake and small artery tone in advanced experimental renal insufficiency

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Abstract

Background. Disturbed calcium–phosphorus balance significantly contributes to uraemic changes in large arteries. We examined the influences of high-calcium and high-phosphate intake on small artery tone in experimental renal insufficiency.

Methods. Sixty-five rats were assigned to 5/6 nephrectomy (NTX) or sham operation. After 15 week disease progression, NTX rats were given high-calcium (3%), high-phosphate (1.5%) or control diet (0.3% calcium, 0.5% phosphate) for 12 weeks. Then isolated segments of small mesenteric arteries were studied using wire and pressure myographs.

Results. Subtotal nephrectomy reduced creatinine clearance by 60% and increased parathyroid hormone (PTH) and phosphate 12-fold and 2.7-fold, respectively. High-phosphate intake further elevated PTH and phosphate (33-fold and 5.5-fold, respectively), while the calcium diet suppressed them (to 3.5 and 62% vs sham, respectively). Ventricular B-type natriuretic peptide synthesis was increased, and blood pressure was 27 and 18 mmHg higher in NTX rats on control and phosphate diet, respectively, than in calcium-fed rats. Vasorelaxation to acetylcholine was impaired by \( \frac{1}{2} \) in uraemic rats, and was further deteriorated by high-phosphate intake, whereas the calcium diet improved endothelium-mediated relaxation via nitric oxide and potassium channels. Small arteries of all NTX groups featured eutrophic inward remodelling: wall-to-lumen ratio was increased 1.3-fold without change in cross-sectional area.

Conclusion. High-phosphate intake had a detrimental influence on secondary hyperparathyroidism and vasodilatation, whereas high-calcium intake reduced blood pressure and PTH, alleviated volume overload and improved vasorelaxation in experimental renal insufficiency. Therefore, alterations in the calcium–phosphorus balance can significantly modulate small artery tone during impaired kidney function.

Keywords: arterial function; calcium; endothelium; kidney failure chronic; phosphate; PTH

Introduction

Cardiovascular complications are a major clinical problem in chronic renal insufficiency (CRI) [1]. Independently of blood pressure (BP) and other classical cardiovascular risk factors, reduced renal function is associated with increased large-artery stiffness [2]. The associated morphological alterations in arteries include smooth muscle hyperplasia, medial calcification and increased intima–media thickness [1,2]. Changes in the function and structure of resistance arteries have also been reported in clinical [1,3] and experimental uraemia [4]. Deficient vasorelaxation of small arteries may even contribute to the elevation of BP during impaired kidney function [3].

Vascular changes in CRI could result from hypertension, volume overload, acidosis, anaemia, uraemic toxins and disturbed calcium–phosphorus balance [1,5]. Elevated levels of phosphate and parathyroid hormone (PTH) are associated with increased risk of cardiovascular complications in renal patients [6], while secondary hyperparathyroidism can influence the remodelling of the vascular wall in CRI [7]. Furthermore, endothelial dysfunction in patients with primary hyperparathyroidism may be restored to normal after successful parathyroidectomy [8].
The use of oral calcium salts as phosphate binders is one approach to reduce the high PTH levels in CRI. However, in haemodialysis patients, the use of calcium-based phosphate binders is associated with progressive coronary artery and aortic calcification, especially if hyperphosphataemia is not well controlled [9]. This is because calcium salts may directly, or indirectly via suppression of PTH, adversely influence the balance of skeletal and extraskeletal calcification [9]. In contrast, in experimental CRI in rats, the use of a high-calcium diet has reduced calcifications in the kidney and thoracic aorta [10] and improved vasorelaxation in the mesenteric artery [4]. The beneficial effects of increased calcium ingestion in 5/6 nephrectomized (NTX) rats could be explained by the effective treatment of hyperphosphataemia and secondary hyperparathyroidism, and the absence of the harmful extraskeletal effects of high-calcium intake can be explained by the fact that this is a model of CRI with significant residual renal function rather than a model of end-stage renal disease [4].

In this study, we examined the functional properties of small mesenteric arteries at three different levels of calcium–phosphorus balance in advanced experimental renal insufficiency. Following surgical NTX, renal impairment was first allowed to develop for a 15-week disease progression period. Then, the NTX animals were allocated to high-calcium, high-phosphate or control diet groups, and followed up for a further 12 weeks. Thereafter, the effects of alterations in calcium–phosphorus balance on small artery tone were investigated. The present results show that high-calcium intake provided benefits to the function of small arteries, whereas high-phosphate intake impaired vasorelaxation in NTX rats.

**Methods**

**Animals and experimental design**

Systolic BPs of male Sprague-Dawley rats were measured using the tail-cuff method (Model 129 BP Meter; IITC Inc., Woodland Hills, CA, USA), and NTX or sham operation were carried out at the age of 8 weeks under ketamine/diazepam anaesthesia (75 and 2.5 mg/kg, respectively). In the NTX groups, surgical resection of the upper and lower poles were performed, comprising about two-thirds of the left kidney, followed by contralateral nephrectomy [4,11]. In the sham group both kidneys were decapsulated. Antibiotics (metronidazole 60 mg/kg, cefuroxim 225 mg/kg) were given post-operatively, and pain was relieved with buprenorphine (0.2 mg/kg, 3 times daily, 3 days). Regular rat chow contained 0.9% calcium, 0.8% phosphate, 0.27% sodium and 1500 IU/kg vitamin D (Lactamin R34, AnalyCen, Lindköping, Sweden). Twelve weeks after the operations, eight NTX and eight sham rats were anaesthetized (urethane 1.3 g/kg, intraperitoneally), small mesenteric arteries were excised for functional and morphological studies [4], and the hearts were removed for B-type natriuretic peptide (BNP) determinations [12]. The flowchart of the study is depicted in Figure 1.

Fifteen weeks after surgery, the NTX rats were divided into three groups (n=13 in each) with equal systolic BPs, body weights, urine outputs and plasma creatinines (determined from the tail vein at week 21). Then, for 12 weeks, sham and NTX groups continued on 0.3% calcium and 0.5% phosphate, the NTX-Ca group on 3.0% calcium and 0.5% phosphate, and the NTX-Pi group on 0.3% calcium and 1.5% phosphate (AnalyCen, Lindköping, Sweden). Owing to mortality in this form of experimental CRI, the final animal numbers at the end of the 27-week experiment in the sham, NTX, NTX-Ca and NTX-Pi groups were 10, 7, 11 and 7, respectively (Figure 1). During week 35 (27 weeks after surgery), 24 h water consumption and urine output were measured. Then, blood and heart samples were taken and stored at −70 °C, and standard sections of small mesenteric arteries were carefully excised [4].

Plasma 1,25-dihydroxyvitamin D₃ [1,25(OH)₂D₃] was determined using a commercial kit (IDS Ltd, Boldon, UK), and other determinations were made as previously reported [4,11]. The Mulvany wire multimyograph Model 610A (J.P. Trading, Aarhus, Denmark) was employed for functional studies of vascular preparations. Second-order branches from the mesenteric arterial bed were suspended as rings (length 1.9 mm) on two 40 μm stainless steel wires, each of which was attached to a myograph jaw.

![Fig. 1. The flowchart of the study: Surgery (5/6 nephrectomy, NTX; sham operation, sham) was performed at entry, after which the 15-week disease progression period began. After 12 weeks of disease progression, the functioning of small arteries in vitro was examined before the diets. Then the NTX animals were further allocated to high-calcium (3%), high-phosphate (1.5%) or control diet (0.3% calcium, 0.5% phosphate) groups for the final 12 weeks of the experiment (treatment period). The values in parentheses denote the number of animals/group at each phase of the experiment.](image-url)
The physiological salt solution (PSS; pH 7.4) contained (mmol/l) the following: NaCl 119.0, NaHCO3 25.0, glucose 11.1, CaCl2 1.6, KCl 4.7, KH2PO4 1.2 and MgSO4 1.2, and was aerated with 95% O2 and 5% CO2. The preparations were normalized so that the vessel internal diameter was set at 90% of that obtained when exposed to an intraluminal pressure of 100 mmHg in the relaxed state [4,11].

Small-artery morphology was examined using a pressure myograph (Living Systems Instrumentation Inc., Burlington, VT, USA) as previously reported, and the development of myogenic tone was inhibited using Ca2+ 7.3 (blockers of small and large 

conductance Ca2+ channels [KCa], respectively] and relaxations were depicted as a percentage of pre-existing contraction. The area under curve (AUC) of the ACh response was determined to present (i) total relaxations and (ii) contributions of NO, cyclooxygenase-derived compounds and KCa to the responses. Statistical analysis was by one-way analysis of variance (ANOVA) and the least-significant difference test. If the variable distribution was skewed, the Kruskal–Wallis and Mann–Whitney U-tests were applied, and P-values were corrected with the Bonferroni equation. ANOVA for repeated measurements was applied for data consisting of repeated observations. Results were expressed as mean ± SEM, and P < 0.05 denoted significance.

Results

Animal data and calcium excretion

Before the diets, systolic BP was ~18 mmHg higher in NTX than in sham rats. During the diets, the NTX and NTX-Pi groups exhibited an elevation of BP, whereas increased calcium intake lowered BP (Figure 2A, Table 1). Body weights did not differ in the NTX and NTX-Ca groups at week 35, while the NTX-Pi group weighed less than the NTX group. Body weights in the NTX-Ca and NTX-Pi groups were lower than in sham rats, while the difference between the NTX and sham groups was not significant (P = 0.070). Heart weight, fluid intake and urine output were increased in all NTX groups. Urine volumes were highest in the NTX-Pi group. The 24 h urine calcium excretion was increased in all NTX groups, and was approximately 7-fold higher in the NTX-Ca than NTX group (Table 1).

Laboratory findings in plasma

Plasma creatinine was elevated by approximately 1.4-fold in all NTX groups before the diets (Table 2). At week 35, creatinine was further increased in all NTX groups, but was lower in the NTX-Ca than NTX-Pi group. Creatinine clearance at week 35 was reduced in all NTX groups when compared with the sham group, while the differences between the NTX, NTX-Ca and NTX-Pi groups were not significant (Table 2). Plasma phosphate and PTH were increased in the NTX group, further elevated in the NTX-Pi group and reduced in NTX-Ca rats. Plasma ionized calcium was increased in the NTX-Ca and decreased in the NTX-Pi group. Plasma 1.25(OH)2D3 was lower in all NTX groups than in sham rats, and was not influenced by the diets (Table 2).

BNP results

Before the diets, ventricular BNP synthesis was higher in NTX than in sham rats: the BNP mRNA levels (normalized to 18S) were 1.7 ± 0.1 vs 1.0 ± 0.1,
and BNP peptide levels (fmol/mg) were 2.15±0.16 vs 1.40±0.08, respectively (n=8; P<0.05 for both analyses). At the close of the study, ventricular BNP mRNA and BNP peptide levels were higher in NTX and NTX-Pi rats than in sham rats, while a high-calcium diet reduced these variables in NTX rats (Figure 2B and C).

**Mesenteric arterial relaxation and morphology before the diets**

At study week 20, vasorelaxation to ACh was impaired in NTX rats (Figure 3A), while relaxation to nitroprusside did not differ from sham rats (Figure 3B). Wall-to-lumen ratio of small arteries was higher in NTX than in sham rats (8.0±0.4 vs 6.7±0.3%, respectively, n=8; P<0.05), whereas wall thickness, lumen diameter and wall cross-sectional area did not differ (data not shown).

**Mesenteric arterial relaxation at the end of study**

**Endothelium-independent relaxations.** The relaxations induced by nitroprusside were similar in NTX, NTX-Ca and sham groups, while the response was enhanced in NTX-Pi rats when compared with sham and NTX-Ca rats (Figure 4A). The relaxations to the K_Ca opener EET were impaired in the NTX and NTX-Pi groups when compared with sham, whereas the response to EET did not differ from sham in the NTX-Ca group (Figure 4B).

**Endothelium-dependent relaxations**

Vasorelaxation to ACh was impaired in the NTX group and further deteriorated in the NTX-Pi group, while the response in the NTX-Ca group did not differ from the sham rats (Figure 4C). The AUC of the ACh response was smaller in the NTX and NTX-Pi, but not in the NTX-Ca group, when compared with the sham group (Figure 5). The addition of the NOS inhibitor L-NAME reduced the AUC of the ACh response, but the change was less marked in NTX and NTX-Pi rats.

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**Table 1.** Experimental group data and urine calcium excretion

<table>
<thead>
<tr>
<th>Variable</th>
<th>Sham</th>
<th>NTX</th>
<th>NTX-Ca</th>
<th>NTX-Pi</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (g)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 23</td>
<td>507±9</td>
<td>479±10</td>
<td>483±10</td>
<td>486±13</td>
</tr>
<tr>
<td>Week 35</td>
<td>565±8</td>
<td>507±41</td>
<td>481±15*</td>
<td>431±38*†</td>
</tr>
<tr>
<td>Final systolic blood pressure (mmHg)</td>
<td>130±2</td>
<td>170±5*</td>
<td>143±4*†</td>
<td>161±4*</td>
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<tr>
<td>Heart weight/body weight (g/kg)</td>
<td>3.25±0.04</td>
<td>4.16±0.50*</td>
<td>4.02±0.15*</td>
<td>4.46±0.43*</td>
</tr>
<tr>
<td>Fluid intake (ml/24 h)</td>
<td>29.7±1.9</td>
<td>56.1±6.0*</td>
<td>57.3±5.4*</td>
<td>72.5±7.8*</td>
</tr>
<tr>
<td>Urine</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Volume (ml/24 h)</td>
<td>21.3±1.4</td>
<td>40.3±4.5*</td>
<td>43.7±5.0*†</td>
<td>63.5±6.1*†</td>
</tr>
<tr>
<td>Calcium (μmol/24 h)</td>
<td>22.5±4.7</td>
<td>76.6±9.7*</td>
<td>560.0±48.5*†</td>
<td>56.1±6.5*</td>
</tr>
</tbody>
</table>

Mean±SEM, n=7–11. NTX, NTX-Ca and NTX-Pi are 5/6 nephrectomized rats, 3% calcium-fed NTX rats and 1.5% phosphorus-fed NTX rats, respectively.  
*P<0.05 vs sham; †P<0.05 vs NTX, ‡P<0.05 vs NTX-Pi.
than in NTX-Ca and sham rats. The reduction of the ACh response by \( K_{Ca} \) blockade with apamin and iberiotoxin was also lower in the NTX group than the NTX-Ca and sham groups. \( K_{Ca} \) blockade did not have any influence on the ACh relaxation in NTX-Pi rats, while the cyclooxygenase inhibitor, diclofenac, was without effect on the ACh response in all groups (Figure 5).

### Table 2. Laboratory findings in plasma

<table>
<thead>
<tr>
<th>Variable</th>
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<th>NTX-Ca</th>
<th>NTX-Pi</th>
</tr>
</thead>
<tbody>
<tr>
<td>Creatinine (µmol/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 21</td>
<td>63.1 ± 1.4</td>
<td>88.4 ± 3.6*</td>
<td>92.8 ± 3.0*</td>
<td>90.4 ± 2.8*</td>
</tr>
<tr>
<td>Week 35</td>
<td>66.6 ± 2.2</td>
<td>170.0 ± 37.7*</td>
<td>116.5 ± 7.3*</td>
<td>178.0 ± 30.9*</td>
</tr>
<tr>
<td>Creatinine clearance (ml/min)</td>
<td>1.84 ± 0.11</td>
<td>0.78 ± 0.16*</td>
<td>0.88 ± 0.06*</td>
<td>0.69 ± 0.18*</td>
</tr>
<tr>
<td>Phosphate (mmol/l)</td>
<td>1.22 ± 0.06</td>
<td>2.69 ± 0.61*</td>
<td>0.76 ± 0.06*</td>
<td>5.47 ± 1.21*</td>
</tr>
<tr>
<td>PTH (pg/ml)</td>
<td>110.1 ± 18.2</td>
<td>1300.5 ± 410.6*</td>
<td>3.8 ± 0.5*</td>
<td>3619.7 ± 255.0*</td>
</tr>
<tr>
<td>Ionized calcium (mmol/l)</td>
<td>1.35 ± 0.01</td>
<td>1.34 ± 0.03</td>
<td>1.53 ± 0.03*</td>
<td>0.93 ± 0.09*</td>
</tr>
<tr>
<td>1,25(OH)₂ D₃ (pmol/l)</td>
<td>273.0 ± 27.8</td>
<td>81.2 ± 23.9*</td>
<td>88.0 ± 14.5*</td>
<td>69.0 ± 29.0*</td>
</tr>
<tr>
<td>pH</td>
<td>7.41 ± 0.02</td>
<td>7.35 ± 0.03*</td>
<td>7.40 ± 0.02</td>
<td>7.38 ± 0.02</td>
</tr>
</tbody>
</table>

Mean ± SEM, \( n = 7–11 \), groups as in Table 1. *\( P < 0.05 \) vs sham, \( ^{1} P < 0.05 \) vs NTX, \( ^{2} P < 0.05 \) vs NTX-Pi.

Mesenteric arterial contractions and morphology at the end of study

All study groups showed comparable vasoconstrictor sensitivity and maximal wall tensions induced by NA (Table 3). Wall-to-lumen ratio of small arteries was increased 1.3-fold in the NTX, NTX-Ca and NTX-Pi groups when compared with sham. Lumen diameter was lower in the NTX than the sham group, while wall thickness and cross-sectional area did not differ between the groups (Table 3). Increased wall-to-lumen ratio and decreased lumen diameter, in the absence of changes in wall thickness and cross-sectional area, indicate that resistance arteries from rats with CRI featured eutrophic inward remodelling [13]. This corresponds to previous results in rats with CRI [4]. Taken together, no changes were detected in vasoconstrictor responses or dimensions of arteries that would explain the above alterations in vasorelaxation following the modifications of the calcium–phosphorus balance.

Discussion

The present results showed that a high-calcium diet suppressed PTH and improved vasorelaxation in experimental CRI, whereas a high-phosphate intake had a detrimental influence on secondary hyperparathyroidism and endothelium-mediated vasodilatation. High-calcium intake also reduced BP, whereby several potential mechanisms can explain the changes in vasorelaxation following alterations in calcium–phosphorus balance. The present diets were without influence on plasma 1,25(OH)₂ D₃, indicating that putative influences of calcium and phosphorus intake on active vitamin D levels did not explain the functional changes in small arteries. We previously found that a high-calcium diet, at an earlier stage.
of experimental CRI, could enhance vasorelaxation in the absence of changes in renal function or BP [4].

In order to mimic clinical CRI, the rats were followed for 15 weeks after NTX, and thereafter the 12-week diets were given. Based on creatinine clearance determinations, ~40% of the glomerular filtration rate remained in the NTX group, while plasma phosphate was increased the 2.2-fold and PTH 11.8-fold when compared with the sham group. A high-phosphate diet reduced body weight and deteriorated the metabolic changes of CRI: hypocalcaemia was observed, plasma phosphate increased 4.5-fold and PTH increased 33-fold when compared with the sham group. Urine output was increased 2-fold in the NTX group since renal mass reduction causes glomerular hyperfiltration and increases functional stress to the remaining nephrons, thus impairing urine-concentrating capacity [10]. The additional increase in urine volume in the NTX-Pi group indicates that urine-concentrating capacity was further impaired following high-phosphate intake.

Ventricular BNP synthesis was measured as a marker of cardiac load [12], and the results of the BNP measurements showed good parallelism with the BP measurements (Figure 2). It is of note that BNP synthesis in NTX rats was already elevated before the diets. At the close of the study, ventricular levels of BNP mRNA and BNP peptide were increased in NTX and NTX-Pi rats, whereas these variables in the NTX-Ca group did not differ from sham. These findings document the volume and pressure excess in CRI, and indicate that the cardiac load was alleviated by high-calcium intake. Calcium supplementation has been reported to increase renal sodium excretion [14], the mechanism of which may partially explain the present effects on cardiac natriuretic peptides.

Impaired endothelial function has been reported in CRI [3,4,11], and this view is supported by our findings. The ACh-induced vasodilatation is predominantly mediated via NO and endothelium-derived hyperpolarization [15], and we addressed the relative roles of these components by calculating the AUC changes of the ACh relaxation induced by NOS inhibition and K⁺ channel blockade. The contribution of NO to the ACh response was reduced in NTX and NTX-Pi rats, but did not differ from the sham group in NTX-Ca rats. The contribution of KCa to the endothelium-dependent relaxation was also decreased in NTX and NTX-Pi rats, but not in NTX-Ca rats. The NTX-Pi rats featured negligible relaxation via K⁺ channels in response to ACh. These results showed that impaired endothelium-dependent dilatation in experimental CRI involved both NO and K⁺ channel-mediated vasorelaxation, and that these impairments could be amended by a high-calcium diet.

Several mechanisms can explain impaired vasorelaxation via endothelial NO in CRI. Increased concentrations of endogenous NOS inhibitors may reduce NO synthesis [16], while endothelial cells may also feature deficiency of L-arginine [17]. NOS activity could be affected by secondary hyperparathyroidism, since parathyroidectomy has been reported to increase NO production in NTX rats [18]. Such a mechanism would correspond to the present results, since increased calcium intake that suppressed plasma PTH enhanced relaxation via NO, while high-phosphate

Fig. 4. Vasorelaxation at the close of the study: responses to nitroprusside (A) and 11,12-epoxyeicosatrienoic acid (EET; B) in endothelium-denuded and acetylcholine in endothelium-intact (C) small mesenteric arterial rings. Mean ± SEM, n = 7–11.
intake elevated PTH, impaired vasorelaxation via endogenous NO.

The sensitivity of arterial smooth muscle to nitroprusside was similar in other groups, while the NTX-Pi rats featured an enhanced response to nitroprusside. This may reflect a compensatory increase in smooth muscle sensitivity to exogenous NO due to reduced bioavailability of endogenous NO. Smooth muscle relaxation induced by the KCa agonist EET was impaired in NTX and NTX-Pi rats, whereas this response did not differ from the sham group in the NTX-Ca rats. Thus, vasorelaxation via KCa in the smooth muscle was decreased in CRI, and was improved by a calcium diet. Previously, PTH has been found to increase the synthesis of the KCa blocker 20-hydroxyeicosatetraenoic acid in renal tubular cells [15]. Whether calcium–phosphorus balance influences the expression of the components of the renin–angiotensin system in the vascular wall remains to be studied.

The development of secondary hyperparathyroidism is characteristic of CRI, while treatment of hyperphosphataemia can ameliorate hyperparathyroidism and decrease cardiovascular complications in renal patients [5,19]. In the present study, high-calcium diet reduced plasma phosphate and suppressed PTH, but induced hypercalcaemia in NTX rats. Since increased calcium ingestion resulted in functional benefits in small arteries, the present findings suggest that hyperphosphataemia, but not mild hypercalcaemia, is deleterious to the vasculature in this form of experimental CRI. In haemodialysis patients, high plasma levels of phosphorus and calcium and high intake of calcium salts are risk factors for cardiovascular calcification [9]. The beneficial effects of the high-calcium diet in our study appear to contradict such clinical findings. However, unlike the situation in haemodialysis patients, the NTX rats show significant residual renal function, and can buffer large quantities of calcium to the urine (Table 1). As constantly

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### Table 3. Contractions and morphology of small mesenteric arteries

<table>
<thead>
<tr>
<th>Variable</th>
<th>Sham</th>
<th>NTX</th>
<th>NTX-Ca</th>
<th>NTX-Pi</th>
</tr>
</thead>
<tbody>
<tr>
<td>Noradrenaline pD2 (−log mol/l)</td>
<td>5.62 ± 0.05</td>
<td>5.52 ± 0.05</td>
<td>5.59 ± 0.05</td>
<td>5.55 ± 0.06</td>
</tr>
<tr>
<td>Maximal wall tension (mN/mm)</td>
<td>12.22 ± 0.92</td>
<td>12.09 ± 1.71</td>
<td>13.91 ± 1.42</td>
<td>8.45 ± 2.29</td>
</tr>
<tr>
<td>Morphology at 100 mm Hg</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wall-to-lumen ratio (%)</td>
<td>6.3 ± 0.2</td>
<td>8.5 ± 0.7*</td>
<td>8.0 ± 0.2*</td>
<td>8.4 ± 0.7*</td>
</tr>
<tr>
<td>Lumen diameter (µm)</td>
<td>402.8 ± 11.5</td>
<td>350.0 ± 20.3*</td>
<td>385.3 ± 10.9</td>
<td>380.7 ± 15.8</td>
</tr>
<tr>
<td>Wall thickness (µm)</td>
<td>25.3 ± 0.7</td>
<td>29.6 ± 2.8</td>
<td>30.6 ± 0.9</td>
<td>31.9 ± 2.9</td>
</tr>
<tr>
<td>Wall cross-section (µm²)</td>
<td>34059 ± 1564</td>
<td>35898 ± 4375</td>
<td>40168 ± 2042</td>
<td>41851 ± 4966</td>
</tr>
</tbody>
</table>

Mean ± SEM, n = 7–11, groups as in Table 1. pD2, negative logarithm of concentration producing 50% of maximum; *P < 0.05 vs sham.
Growing animals, rats can also buffer calcium to their bones. Therefore, the NTX rat model of CRI, which in the present study corresponds to chronic stage 3 kidney disease, appears to be resistant to the unfavourable cardiovascular effects of high-calcium intake. Subsequently, these results cannot be extrapolated to humans with end-stage renal disease (stage 5), and the present experimental findings have limited clinical relevance to adult human dialysis patients. It should also be noted that the present approach cannot distinguish between direct and indirect effects of a high-calcium or high-phosphate diet on vascular tone.

Although a high intake of calcium supplements may pre-dispose the end-stage renal disease patients to increased vascular calcifications [9], the link between calcium intake and soft tissue calcification is not straightforward. Inhibition of calcification in the vasculature is actually an active process mediated by special proteins [20]. Whether high dietary intake of calcium is a risk factor for extraskeletal calcification in chronic stages 2–4 clinical kidney disease remains to be explored in the future. In patients with CRI, a 2–3-fold elevation in plasma PTH has been considered beneficial to the bones, but the cardiovascular actions of chronic increases in PTH are unknown. The present results showed that a high-calcium intake, which suppressed PTH levels, also reduced BP and cardiac load, and improved endothelium-mediated relaxation. In contrast, high-phosphate intake had a detrimental influence on secondary hyperparathyroidism and vasodilatation, the effects of which were independent of the level of BP and cardiac load. These experimental results suggest that alterations in calcium–phosphorus balance can significantly modulate resistance artery tone in CRI.

Acknowledgements. This study was supported by the Medical Research Fund of Tampere University Hospital, Academy of Finland, Finnish Kidney Foundation, Finnish Foundation for Cardiovascular Research, Paavo Nurmi Foundation, Pirkanmaa Regional Fund of Finnish Cultural Foundation and Sigrid Juselius Foundation.

Conflict of interest statement. None declared.

References


Received for publication: 4.3.06
Accepted in revised form: 18.4.06