High-dose aprotinin with gentamicin–vancomycin antibiotic prophylaxis increases blood concentrations of creatinine and cystatin C in patients undergoing coronary artery bypass grafting

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Both aprotinin and gentamicin–vancomycin antibiotic prophylaxis have been used widely in cardiac surgery to prevent bleeding and infections, respectively. As the drugs are excreted almost entirely by glomerular filtration, we investigated their action on renal function when administered either separately or together. To increase consistency, we measured serum concentrations of creatinine and cystatin C, a new marker of glomerular filtration rate, that many recent studies have shown to be more sensitive than serum creatinine. One hundred patients undergoing coronary artery bypass surgery were allocated randomly to one of four groups: group A received antibiotic prophylaxis with cefamandole and no aprotinin; group B received cefamandole and high-dose aprotinin; group C received antibiotic prophylaxis with gentamicin and vancomycin, but no aprotinin; and group D received both high-dose aprotinin and gentamicin–vancomycin antibiotic prophylaxis. Data from 84 patients, for whom data collection was complete, were analysed. In the first week after operative, mean serum concentrations of cystatin C and creatinine either remained constant or decreased slowly in all groups, except for group D. In group D, both markers increased gradually from postoperative day 2 onwards. The increase in cystatin C was significant on postoperative day 5 (from mean 1.02 (SD 0.11) mg litre⁻¹ before operation to 1.35 (0.32) mg litre⁻¹; P<0.05), reaching a peak on postoperative day 7 (1.45 (0.35) mg litre⁻¹; P<0.05), while the increase in creatinine concentration was significant on postoperative day 6 (from 1.05 (0.16) mg dl⁻¹ before operation to 1.29 (0.34) mg dl⁻¹; P<0.05). We conclude that simultaneous administration of high-dose aprotinin and prophylactic use of gentamicin with vancomycin increased serum concentrations of cystatin C and creatinine in the first postoperative week in patients undergoing cardiac surgery.

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artery surgery, in a prospective, randomized study. Preoperative renal function was normal in all patients (defined as plasma creatinine <1.5 mg dL\(^{-1}\), creatinine clearance >70 ml min\(^{-1}\) and plasma urea <60 mg dL\(^{-1}\)). Exclusion criteria were: severe hypertension, diabetes, angiotensin converting enzyme inhibitor therapy, surgical emergency, ejection fraction <40\%, previous administration of aprotinin, allergy to aminoglycosides or vancomycin, and administration of radiological contrast medium in the previous 72 h.

All patients received premedication with diazepam 0.1 mg kg\(^{-1}\) i.m. and morphine sulphate 0.1 mg kg\(^{-1}\) i.m., 1 h before induction of anaesthesia. After insertion of radial arterial, peripheral and central venous catheters, anaesthesia was induced with fentanyl 20–30 µg kg\(^{-1}\), diazepam 0.08 mg kg\(^{-1}\) and pancuronium 0.1 mg kg\(^{-1}\). Anaesthesia was maintained by infusion of fentanyl 0.2 µg kg\(^{-1}\) min\(^{-1}\) and additional doses of diazepam.

Patients underwent standard non-pulsatile hypothermic (28–32°C) cardiopulmonary bypass (CPB) using a Sarns roller pump, with Dideco D-703 compact flow membrane oxygenator, with a continuous flow of 2–2.5 litre min\(^{-1}\) and a perfusion pressure of 50–80 mm Hg.

Systemic heparinization was achieved before CPB with heparin 300 u. kg\(^{-1}\) and supplemented as necessary to maintain a kaolin-activated coagulation time >500 s during CPB. Antegrade crystalloid cold (4°C) intermittent cardioplegia with topical hypothermia was used as a method of myocardial protection. Systemic temperature was measured using rectal and oropharyngeal probes. The perfusate was cooled actively through a heat exchanger (Sarns) until an oropharyngeal temperature of approximately 29°C was reached. Heparin was neutralized with protamine 1 mg/100 u. of heparin.

Patients were allocated randomly to one of four groups using a table of random numbers. Group A received cefamandole 2 g i.v., diluted in 250 ml of 0.9% NaCl saline solution, for antibiotic prophylaxis, 30 min before surgery and every 6 h after operation for 48 h, and no aprotinin. Group B received cefamandole prophylaxis at the same dose as in group A in addition to high-dose aprotinin (Antagosan). Aprotinin was administered as a bolus of 2\(\times10^6\) kallikrein inhibitor units (kiu) over 15 min, starting shortly before induction of anaesthesia, and after a test dose; this was followed by continuous infusion at a rate of 5\(\times10^3\) kiu h\(^{-1}\). An additional dose of 2\(\times10^6\) kiu was added to the CPB prime solution. The high-dose regimen was similar to those used in other studies.\(^3\)\(^4\)\(^10\)\(^12\) Group C received antibiotic prophylaxis with vancomycin and gentamicin, but no aprotinin. Vancomycin was given as 15 mg kg\(^{-1}\), 3 h before surgery, 10 mg kg\(^{-1}\) after CPB interruption and 15 mg kg\(^{-1}\) every 12 h for 48 h after operation. Gentamicin was administered as 1.5 mg kg\(^{-1}\) just after the first dose of vancomycin and 1.5 mg kg\(^{-1}\) every 8 h for 48 h after operation. Each dose was diluted in 250 ml of 0.9% NaCl saline solution and administered over a period not less than 30 min. Group D received both aprotinin and vancomycin–gentamicin antibiotic prophylaxis at the same doses as in the previous groups.

Serum concentrations of cystatin C and creatinine were measured in venous blood samples obtained at the following times: before operation on the day before surgery, before induction of anaesthesia, 10 min after induction, 10 min after CPB weaning, and on days 1–7 after operation. Preoperative measurements were taken as control values. All samples were collected in tubes at room temperature, centrifuged without delay and either analysed immediately or stored at –20°C until analysed. Cystatin C was measured with an N latex cystatin C kit for the Behring nephelometer 100 system analyser in batch mode (BNA–Behring, Marburg, Germany).

Urine output was recorded at three times during surgery: 10 min before starting CPB, 10 min after CPB interruption, and at the end of intervention, and on postoperative days 1 and 2.

**Statistical analysis**

Only those patients whose data throughout the study were complete were analysed. All data are expressed as mean (SD). Categorical values were analysed using the chi-square test. Exact \(P\) values of continuous variables were determined using the Student’s paired and unpaired \(t\) tests and three-way ANOVA using repeated measures applied to each of the outcome variables. The cut-off for statistical significance was set at \(P<0.05\).

**Results**

Data collection was complete for 84 patients and their results were analysed (Table 1). There were no significant differences between groups in age, weight, height, ejection fraction, duration of operation, bypass time or number of aortocoronary grafts. There were no significant differences between groups for dose of vasodilator or vasoconstrictor agents administered. Haemodynamic changes were similar between groups, as was duration of hospital stay.

**Urine output**

Urine output during surgery was mean 240 (124) ml h\(^{-1}\) in patients who received aprotinin and 205 (115) ml h\(^{-1}\) in those who did not (ns). As shown in Figure 1, there was an increase in urine output during CPB in the aprotinin-treated groups (groups B–D, 522 (223) ml h\(^{-1}\) vs groups A–C, 388 (210) ml h\(^{-1}\); \(P=0.047\), but at the end of surgery, mean urine output did not differ significantly between groups.

There were no significant differences between groups in urine output during the first 2 days after operation, although groups treated with aprotinin showed a trend towards increased urine production (groups B–D, 2891 (1215) ml day\(^{-1}\), groups A–C, 2220 (952) ml day\(^{-1}\);
Aprotinin with gentamicin increases serum creatinine and cystatin C

Table 1 Patient data (mean (SD or range))

<table>
<thead>
<tr>
<th></th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Group D</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>18</td>
<td>22</td>
<td>21</td>
<td>23</td>
</tr>
<tr>
<td>Age (yr)</td>
<td>61 (50–75)</td>
<td>61 (53–71)</td>
<td>75 (12)</td>
<td>63 (10)</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>73 (6)</td>
<td>66 (13)</td>
<td>75 (12)</td>
<td>63 (10)</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>168 (10)</td>
<td>165 (12)</td>
<td>170 (8)</td>
<td>172 (5)</td>
</tr>
<tr>
<td>Sex (M/F)</td>
<td>17/1</td>
<td>20/2</td>
<td>19/2</td>
<td>19/4</td>
</tr>
<tr>
<td>Ejection fraction</td>
<td>0.62 (0.07)</td>
<td>0.65 (0.04)</td>
<td>0.6 (0.05)</td>
<td>0.64 (0.06)</td>
</tr>
<tr>
<td>Mean arterial pressure (mm Hg)</td>
<td>108 (13)</td>
<td>98 (12)</td>
<td>105 (10)</td>
<td>104 (11)</td>
</tr>
<tr>
<td>Serum creatinine (mg dl–1)</td>
<td>1.08 (0.18)</td>
<td>0.99 (0.17)</td>
<td>1.08 (0.19)</td>
<td>1.06 (0.19)</td>
</tr>
<tr>
<td>Serum cystatin C (mg litre –1)</td>
<td>1.06 (0.15)</td>
<td>1.02 (0.3)</td>
<td>1.04 (0.09)</td>
<td>0.92 (0.2)</td>
</tr>
<tr>
<td>CBP time (min)</td>
<td>250 (55)</td>
<td>261 (53)</td>
<td>247 (56)</td>
<td>265 (62)</td>
</tr>
<tr>
<td>Operation time (min)</td>
<td>98 (35)</td>
<td>105 (33)</td>
<td>101 (42)</td>
<td>103 (31)</td>
</tr>
<tr>
<td>Mean No. of bypass grafts</td>
<td>2.7 (0.5)</td>
<td>2.8 (0.4)</td>
<td>2.7 (0.4)</td>
<td>2.8 (0.3)</td>
</tr>
</tbody>
</table>

Table 2 Serum concentrations of cystatin C (mg litre –1) decreased shortly after induction of anaesthesia and during CPB (mean (SD)). *P<0.05, **P<0.01, ***P<0.001

<table>
<thead>
<tr>
<th>Group</th>
<th>Before induction</th>
<th>After induction</th>
<th>After CPB</th>
<th>24 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>1.03 (0.15)</td>
<td>0.8 (0.22)</td>
<td>0.87 (0.22)*</td>
<td>0.82 (0.19)</td>
</tr>
<tr>
<td>Group B</td>
<td>1.05 (0.22)</td>
<td>0.71 (0.23)**</td>
<td>0.74 (0.26)**</td>
<td>0.95 (0.40)</td>
</tr>
<tr>
<td>Group C</td>
<td>1.09 (0.12)</td>
<td>0.44 (0.07)**</td>
<td>0.89 (0.14)**</td>
<td>0.92 (0.28)</td>
</tr>
<tr>
<td>Group D</td>
<td>1.02 (0.20)</td>
<td>0.87 (0.25)*</td>
<td>0.83 (0.10)*</td>
<td>0.94 (0.09)</td>
</tr>
</tbody>
</table>

Table 3 Although less markedly than cystatin C, creatinine concentrations (mg dl–1) decreased after induction of anaesthesia and during CPB (mean (SD)). *P<0.05, **P<0.01, ***P<0.001

<table>
<thead>
<tr>
<th>Group</th>
<th>Before induction</th>
<th>After induction</th>
<th>After CPB</th>
<th>24 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>1.02 (0.11)</td>
<td>0.96 (0.15)</td>
<td>0.95 (0.09)*</td>
<td>1.07 (0.04)</td>
</tr>
<tr>
<td>Group B</td>
<td>0.99 (0.17)</td>
<td>0.92 (0.20)**</td>
<td>0.91 (0.14)**</td>
<td>0.97 (0.17)</td>
</tr>
<tr>
<td>Group C</td>
<td>1.08 (0.19)</td>
<td>0.85 (0.14)**</td>
<td>0.85 (0.20)**</td>
<td>1.07 (0.28)</td>
</tr>
<tr>
<td>Group D</td>
<td>1.06 (0.19)</td>
<td>0.99 (0.25)</td>
<td>0.92 (0.13)**</td>
<td>1.01 (0.15)</td>
</tr>
</tbody>
</table>

P=0.078). There were no differences in urine output according to type of antibiotic prophylaxis.

Serum cystatin C and creatinine concentrations

There was a significant decrease in both markers in all groups shortly after induction of anaesthesia (P<0.001) (Tables 2, 3) compared with pre-induction concentrations, with a simultaneous high flow urine output during the first hour (group A, 143 (65) ml; group B, 101 (53) ml; group C, 127 (62) ml; group D, 99 (43) ml; ns). During CPB, haemoglobin concentration decreased in all groups from a mean baseline value of 12.1 (3.0) to 7.8 (2.2) g dl–1.

In the first week after operation, mean serum concentrations of both markers remained constant or decreased slowly in all groups, except for group D. In group D, cystatin C or creatinine increased gradually, starting on postoperative day 2. The increase in cystatin C was significant on postoperative day 5 (P<0.05), reaching a peak (Table 4) on day 7 (P<0.05), while the increase in creatinine was significant on day 6 (P<0.05) (Table 5). A retrospective analysis of available data for group D showed that five patients had preoperative values of creatinine restored within the second postoperative week. One patient in group D developed acute renal failure, as did one patient in group A; neither required dialysis. A peak increase of at least 50% of basal values of both markers occurred in nine (39%) patients in group D. One patient in group C and one in group A died, but they had normal renal function until death.

Serum electrolytes and blood urea nitrogen

There were no significant differences between groups for sodium, potassium, chloride, bicarbonate or blood urea nitrogen concentrations at each time.

Discussion

Cystatin C and creatinine

Cystatin C is a non-glycosylated 13-kDa basic protein with 120 amino acids produced at a constant rate by all body tissues, and is eliminated from blood almost exclusively by glomerular filtration. Several recent studies suggested that serum cystatin C concentrations correlate with GFR. Newman and colleagues showed that cystatin C increased significantly earlier and to a greater extent than serum creatinine as GFR decreased. Some authors have indicated a value of 0.86 (range 0.6–1.2) mg litre–1 as the normal mean cystatin C serum concentration. Newman and colleagues showed that cystatin C increased significantly earlier and to a greater extent than serum creatinine as GFR decreased. Some authors have indicated a value of 0.86 (range 0.6–1.2) mg litre–1 as the normal mean cystatin C serum concentration.

An increase in cystatin C concentration with age has been reported in patients aged more than 50 yr, corresponding to the known age-related decrease in GFR. All patients in our study were older than 50 yr (Table 1).

Serum creatinine is less sensitive than serum cystatin C as a marker of GFR, in particular at a low filtration rate.
As creatinine is determined mainly by muscular mass and food intake, the rate is variable. Creatinine clearance is of little value in clinical practice. Creatinine does not meet the criteria for an ideal GFR marker; difficulties in collecting quantitative urine portions frequently lead to grossly erroneous results and, in some immunological disorders, it can lead to overestimation of GFR. Feindt and colleagues showed that patients who received high-dose aprotinin had considerable tubular proteinuria with respect to the control group, although no differences were found in creatinine concentrations in blood or urine.

In contrast, cystatin C and creatinine are specific in their ability to exclude individuals with normal GFR, but for sensitivity (i.e. identification of individuals with GFR impairment) serum cystatin C is better.

Aprotinin and the kidney

Aprotinin is a serine protease inhibitor derived from bovine lung tissue. It is used to minimize blood loss and to prevent complications associated with transfusion of blood products in patients undergoing cardiac surgery. The precise mechanism by which aprotinin reduces bleeding is unknown: 90% of a given dose of aprotinin is absorbed and collected by the convoluted tubules within a few hours after i.v. administration. It accumulates and is metabolized and eliminated within 5–6 days.

Aprotinin has greater affinity for renal than plasma kallikrein, promotes renal vasodilatation and increases sodium excretion. During aprotinin use, it is possible to observe increased urine \(\alpha_2\)-microglobulin concentration, which has been interpreted as increased tubular activity. For these reasons, some authors believe that its use can cause renal impairment. In contrast, several studies have not identified a significant adverse effect of aprotinin on postoperative renal function. Dietrich and colleagues studied 1784 patients over 3 yr and could not identify a detrimental effect of aprotinin on renal function, even in patients with preoperative serum creatinine >2 mg dl\(^{-1}\). Sundt and colleagues, in a study carried out in 40 patients who underwent heart surgery with hypothermic circulatory arrest, found that renal dysfunction occurred in 65% of patients who received aprotinin (P<0.0001). Prendergast and colleagues studied the use of aprotinin in 70 patients receiving heart transplantation and stressed that there was no evidence of additional renal dysfunction in patients who received aprotinin.

Vancomycin, gentamicin and the kidney

Cephalosporin antibiotics, such as cefamandole, are used routinely in perioperative prophylactic regimens. However, in our hospital, this antibiotic is associated with an incidence of postoperative infections higher than international standards. In particular, a study conducted in cardiac surgery patients at our institute showed a 75% rate of oxacillin-resistant *Staphylococcus aureus* wound infections. In this case, the use of vancomycin prophylaxis is recommended, possibly associated with aminoglycosides, gentamicin or tobramycin, to ensure protection against gram-negative bacteria.

Vancomycin does not appear to have adverse effects on the kidney. Maki and colleagues compared cefazolin, cefamandole and vancomycin for surgical prophylaxis in cardiac patients and could not identify any detrimental effect on renal function caused by these drugs. However, Fillastre and Godin showed that vancomycin increased the toxicity associated with aminoglycoside treatment.

In contrast, it is well known that aminoglycosides, in particular gentamicin, are nephrotoxic agents. They accumulate in the cortex of the kidney where they reach concentrations 10-fold greater than those in plasma. Accumulation in the cytoplasm produces membrane alteration, with consequent release of lysosomal enzymes, and mitochondrial dysfunction. Moreover, they inhibit the enzyme phospholipase A2, reducing arachidonic acid, the substrate for prostaglandins. As a result, renal blood flow is reduced, renal perfusion is decreased, and the filtration fraction is increased, leading to a decrease in creatinine clearance.125
Nevertheless, only one patient developed renal failure, but for prostaglandin synthesis. Of note, nephrotoxicity caused by gentamicin does not become immediately manifest, but only after 7–10 days of treatment, when glomerular filtration is reduced and blood creatinine and urea are consequently increased. Nevertheless, these increases were reversible and without sequelae.

Bertram and Summers reported that the use of gentamicin did not measurably affect the kidney over the short period of treatment in patients undergoing cardiac surgery. Morin and colleagues, in a study conducted in the rat, showed that gentamicin nephrotoxicity was potentiated by aprotinin. Serum creatinine was 72% higher in rats in which aprotinin was administered together with gentamicin than if gentamicin was administered alone.

In our study, all groups showed a significant decrease (Tables 2, 3) in cystatin C serum concentrations shortly after induction of general anaesthesia compared with preanaesthesia concentrations. Creatinine behaved similarly, but to a lesser degree. Mean urine output in all patients, measured from induction to CPB, was 118 (56) ml h⁻¹, with no significant difference between groups. The reason for this improvement in renal function remains unclear, but is of little clinical significance. It was probably caused by reduction of sympathetic tone after induction of anaesthesia. At the same time, the decrease in arterial pressure stimulated the renin–angiotensin axis, causing preferential constriction of efferent arterioles thus improving GFR and urine output.

During CPB, we observed a significant difference in urine output between patients in whom aprotinin was administered and those who did not receive it (P = 0.047) (Fig. 1), which agrees with the results of Dietrich and colleagues. It is likely that aprotinin, after the onset of CPB when high plasma concentrations are present, overloads the tubular reabsorption mechanisms, increasing osmolar clearance.

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Our results, in common with others, confirmed that high-dose aprotinin did not affect postoperative renal function in patients undergoing aortocoronary bypass. All patients had constant concentrations of cystatin C, creatinine and urine output during the first postoperative week. Gentamicin–vancomycin prophylaxis also appeared to be relatively safe for the kidney. However, the group treated with both aprotinin and vancomycin–gentamicin prophylaxis showed a statistically significant increase in serum cystatin C on day 5 after operation (P<0.05) (Table 4) and in serum creatinine on day 6 (P<0.05) (Table 5). The delay in the increase in serum markers is compatible with accumulation and a slow clearance rate of aprotinin, but not immediately manifest, but only after 7–10 days of treatment, when glomerular filtration is reduced and blood creatinine and urea are consequently increased. Nevertheless, these increases were reversible and without sequelae.

As mentioned previously, there are several reasons why aprotinin or gentamicin may lead to reduction in GFR. It is likely that the drugs act synergistically against tubular and glomerular functions. They overload the proximal tubule, as confirmed by increased urinary α₁-microglobulin concentrations and osmolar clearance. These events can produce activation of tubulo-glomerular feedback leading to constriction of afferent arterioles, thus reducing glomerular blood flow. Whether reduction of prostaglandin synthesis, inhibition of the kallikrein–kinin system or permeability of the lysosomal membranes also play a role in this series of events is still to be established.

In summary, our results suggest that high-dose aprotinin or prophylactic doses of gentamicin with vancomycin did not seem to impair renal function if not administered in the same patient during coronary artery bypass grafting using moderate hypothermia. However, when the drugs were administered simultaneously, they appeared to produce a modest and transient increase in serum concentrations of cystatin C and creatinine in patients with normal preoperative renal function within the first postoperative week. This does not necessarily imply the emergence of clinically significant adverse events; however, caution is needed when administering these drugs together in patients with preoperative signs of renal dysfunction.

References

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