CISPLATIN NEPHROTOXICITY AGGRAVATED BY CARDIOVASCULAR DISEASE AND DIABETES IN LUNG CANCER PATIENTS

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**Abbreviations:**

- **Cp:** Cisplatin
- **[creat]:** serum creatinine concentration
- **GFR:** measured glomerular filtration rate
- **$^{99m}$Tc-DPTA:** technetium-labeled diethylene-thiamine pentaacetate
- **Ccreat:** calculated creatinine clearance
- **eGFR:** estimated glomerular filtration rate (by Ccreat)
- **NC:** no major comorbidity
- **CD:** cardiovascular disease (hypertension and ischemic heart disease)
- **DMIH:** diabetes mellitus and ischemic heart disease
ABSTRACT

Background: Aging lung cancer patients may be at increased risk of Cisplatin (Cp) nephrotoxicity, because of comorbidities leading to accelerated aging of the kidneys. Therefore the Cp-induced impairment of renal function was compared between no comorbidity (NC) and hypertension+ ischemic heart disease (CD) patients or others having diabetes mellitus+ ischemic heart disease (DMIH).

Methods: In a preliminary study GFR was measured by clearance of $^{99m}$Tc-DTPA in 38 lung cancer patients with normal serum creatinine concentration ([creat]). Then, the incidence of nephrotoxicity was analysed retrospectively over 1st-4th cycles of Cp treatment among 242 lung cancer patients with initially normal [creat]. GFR was repeatedly estimated by calculated creatinine clearance (eGFR).

Results: Pre-treatment GFR was $57\pm3$ ml/min/m$^2$ in those with normal (n=15) and $42\pm2$ (p<0.05) in others with pathologically increased (n=23) [creat] any time following their 2nd-4th Cp cycle. The retrospective analysis revealed that Cp-induced nephrotoxicity developed in 7.5% of NC (n=80), in 20.9% of CD (n=110) and in 30.8% of DMIH (n=52) subgroups. Within overall fall-out from further Cp chemotherapy, nephrotoxicity was responsible in 14% of NC, 38% in CD and 75% in DMIH patients.

Conclusion: A major portion of our aging lung cancer patients suffered from comorbidities leading to reduced renal resistance to Cp nephrotoxicity.

KEYWORDS

aging kidney, cardiovascular disease, cisplatin nephrotoxicity, diabetes mellitus, lung cancer,
INTRODUCTION

High-dose Cisplatin- (Cp-) based combination chemotherapy regimens are used as front-line treatments of non-small cell and small cell lung cancer [1]. The therapeutic effects of Cp are significantly improved by dose escalation. However, high dose therapy with Cp is limited by its cumulative nephrotoxicity [2]. Cp is toxic to the renal proximal, as well as distal tubules [3]. Different hydration (saline infusion) protocols were developed that reduced nephrotoxicity and allowed dose escalation to therapeutic levels [4]. However, even with vigilant hydration, approximately one-third of patients treated with Cp have transient elevation of blood urea nitrogen levels or other evidence of kidney damage in the days following Cp treatment [5]. According to Berns and Ford [6] about 20% of acute renal failure cases among hospitalized patients are due to Cp nephrotoxicity. De Jongh et al [7] analysed prognostic factors for nephrotoxicity of high-dose Cp in 400 patients with a median age of 54 years and suffering from different solid tumors. Nephrotoxicity was defined as a ≥25% decline of estimated Ccreat at any time during the evaluation period. Twenty nine percent of patients developed nephrotoxicity, but temporary elevation of [creat] above the upper normal limit was observed in 41% of patients. Their multivariate analysis selected age, female gender, smoking, paclitaxel coadministration and hypoalbuminaemia as independent risk factors of Cp-induced decrease of renal function.

In developed countries lung cancer patients have a median age of 70 years [8]. Of note, the aging kidney may be more susceptible to nephrotoxins [9]. Aging people are affected by comorbidities as, for example, CD and/or DMIH. Underlying and undiagnosed, early stage nephrosclerosis induced by age, CD or DM alone or in combination may be present among older patients who receive antihypertensive, vasodilator or antidiabetic treatment but have no elevation of [creat]. However, in aged cancer patients [creat] and the concentration of urea may be only „pseudonormal” because of decreased muscle mass and protein intake, respectively. In conditions with reduced production of creatinine and low [creat], abnormally low GFR may occur for extended periods before [creat] will reach the upper limit of the reference range [10]. Increased [creat] is to be considered the least sensitive indicator of reduced GFR [10]. Still, based on „normal” serum indices of renal function aged CD or DMIH patients may receive full dose Cp treatment for lung cancer. Launay-Vacher et al [11] have reported that before chemotherapy around 60% of 445 lung cancer patients had
unrecognized, stage 2 kidney disease (GFR=60-89 ml/min) as estimated by creatinine clearance (Ccreat).

Therefore, in order to determine the occurrence of high-dose Cp nephrotoxicity of lung cancer patients [creat], clearance of $^{99m}$Tc-DPTA (GFR) and Ccreat (eGFR) were compared before and after the administration of Cp in patients suffering also from CD, DMIH or being free from these underlying comorbidities (no comorbidity, NC).

**MATERIAL AND METHODS**

*Study subjects and design*

The Pulmo-Oncology Unit of the Department of Pulmonology at Semmelweis University treats 250-300 non-small and small cell lung cancer patients annually. Since Cp-induced reversible or persistent azotaemia was estimated to occur in ~30% of our patients, in order to investigate whether pre-Cp GFR was already reduced when [creat] was still normal, in a preliminary prospective study we measured glomerular filtration rate (GFR) by clearance of $^{99m}$Tc-DPTA in 38, stage IIIB-IV lung cancer patients with normal [creat] and scheduled for Cp-based chemotherapy. $^{99m}$Tc-DPTA clearance was measured at the Department of Radiology and Oncotherapy of Semmelweis University by the same investigator (dr. LD). $^{99m}$Tc-DPTA (Izotóp Intézet Kft., Budapest, Hungary) was administered iv in a dose of 40 MBq. Patients were grouped later according to their highest post-Cp [creat] either remaining within (n=15) or increasing over (n=23) the upper limit of the reference range (>106 μmol/l) any time during their chemotherapy (2-4 cycles). Cp was administered in 75 mg/m² iv in each cycle. Cp infusions followed each other not earlier than 21 days.

Next, we have retrospectively analysed records of patients (n=242) suffering from stage IIIA-IV non-small or small cell lung cancer and receiving chemotherapy between January and December 2006. High-dose Cp therapy was indicated by our Oncoteam and in addition to fulfillment of many other criteria, Cp was recommended only for patients with normal [creat] and urea concentration and without any other apparent symptoms or signs of altered renal function. Based on initial evaluation of the 242 patients, 3 major subgroups were formed according the absence or presence of comorbidities as cardiovascular disease and/or diabetes mellitus. The subgroup NC had no hypertension, ischemic heart disease or diabetes mellitus. The second subgroup was formed based on the presence of long-term, medically controlled hypertension and ischemic heart disease (together cardiovascular disease-CD, n=110), and the third one based on the combined presence of diabetes mellitus and ischemic heart disease...
without hypertension (DMIH, n=52). The diagnosis of chronic arterial hypertension was based on history and the use of antihypertensive medications. Ischemic heart disease was diagnosed based on history, ECG abnormalities and previous treatment by coronary vasodilators, platelet aggregation inhibitors, or percutaneous transluminal coronary angioplasty. None of the CD patients suffered from uncontrolled hypertension, angina pectoris, acute myocardial infarction or cardiac decompensation or from any other acute or severe cardiovascular comorbidity which could have contraindicated chemotherapy with high-dose Cp. DM was diagnosed based on history, receiving insulin (n=5) or oral antidiabetic treatment (n=47) and higher than normal fasting serum glucose concentration. None of the DM patients suffered from uncontrolled hyperglycemia or had symptoms of major complication of diabetes. Urinary protein test showed opalescence (≥1 g/day) in 2 and slight opalescence (0.5-1.0 g/day) in 2 other patients, the majority had negative (<0.5 g/day) results.

Patients received several subsequent combined chemotherapy courses always containing high-dose Cp (75 mg/m² iv) and each pre-Cp and the highest post-Cp [creat] and urea concentration values were recorded. Cp-induced persistent azotaemia (=Cp nephrotoxicity) was a frequent cause of exclusion from further Cp treatment. The number of these patients was compared between the three groups. Clinical data like age, gender, chronic comorbidity, blood pressure and stage of lung cancer were collected. Out of laboratory data serum glucose, [creat] and urea concentrations were analysed. [creat] was determined based on the modified Jaffe two point kinetic reaction using commercially available test of Dialab (Wiener Neudorf, Austria). Ccreat (=estimated GFR, eGFR) was calculated according the Cockcroft-Gault equation [12]. This calculation was selected because the mean age of our patients was below 65 years [13].

**Other treatments and drugs**

Cp was provided by TEVA Hungary and EBEWE Pharma (Unterach, Austria) and administered in a dose of 75 mg/m². One of three additional chemotherapeutic agent were given in combination with Cp: gemcitabine (1250 mg/m², Eli Lilly, the Netherlands), etoposide (3x120 mg/m²) and paclitaxel (175 mg/m², both from Brystol-Mayers-Squibb, Princeton, NJ). Neutropenia was treated with granulocyte colony stimulating factor (filgrasin, 48 MilliU, AMGEN, Breda, the Netherlands), severe thrombocytopenia with platelet transfusion, anaemia with erythropoietin (Epoetin alfa, 40 000 IU/week, Jansen-Cilag, Centocor, Leiden, Netherlands) and/or transfusion as indicated. Patients received
antinociceptive, antiemetic drugs, bisphosphonate, methylprednisolon and other symptom relievers, as needed.

**Hydration protocol**

The intravenous infusion of 500 ml 0.9% NaCl followed by either gemcitabine, taxol, or etoposide in another 500 ml of saline. After a third 500 ml of saline infusion Cp was infused again in 500 ml. At our Department infusion of 500 ml volume lasts usually 20-30 minutes. Following Cp the 5th 500 ml saline was infused (total volume of saline 2500 ml within ~2.5 hours) and the infusion treatment was ended with 100 ml 20% mannitol (Baxter) iv. methylprednisolon (40 mg), and various antiemetic drugs were also given in many patients.

**Statistical analysis**

Data are reported as means±SEM. Statistical analysis was performed with GraphPad software (Graph Pad Prism 5.0 by Graph Pad Software Inc., San Diego, USA) using Fischer’s exact test, Chi square test, t-test (paired and unpaired) as appropriate. One or two-way analysis of variance (ANOVA) and Kruskal-Wallis test was used to compared more than two groups. Normally distributed data were analysed by ANOVA and non-Gaussian distributed or nonparametric values were analysed by Kruskal-Wallis test. After one-way ANOVA, if significant difference (p<0.05) was found, the Newman-Kuels multiple comparison post-hoc test was used for further analysis. After two-way ANOVA, Bonferroni post-test was used. After Kruskal-Wallis test Dunn’s multiple comparison post-hoc test was performed. The applied tests are mentioned under the figures and the tables.

**RESULTS**

Out of the 38 lung cancer patients 23 responded with pathologically increased [creat] to Cp (Table 1), although pre-treatment [creat]-s were normal in both groups. Pre-treatment GFR, as measured by clearance of $^{99m}$Tc-DPTA, was significantly, by about 25% reduced in those lung cancer patients, who responded with azotaemia to 2-4 cycles of Cp treatment.

To confirm this preliminary observation a restrospective analysis of Cp nephrotoxicity in 242 lung cancer patients treated by Cp was performed. About two-third of them suffered from major comorbidities like CD or DMIH (Table 2). No differences were noted in the distribution of gender, dose of Cp, or in the ratio of patients receiving any of the other
chemotherapeutics. Blood pressure and heart rate were also similar in the three subgroups indicating good blood pressure control in the CD subgroup.

The number of patients made possible the analysis of 4 subsequent Cp cycles. After the first Cp treatment [creat] increased in all subgroups, but the change was significantly greater in subgroups CD and DMIH than NC (both p<0.05, Table 3). The second Cp dose could be given to less patients in all three subgroups, in many of them because of Cp nephrotoxicity (see later). Although pre-Cp [creat]-s were again in the reference range, the second Cp infusion induced a larger increase of [creat] than the first one. This time, concentrations entered the pathological range in the DMIH and were close to this also in the CD, but not in the NC subgroup. The 3rd Cp cycle was administered to only ~50% of patients initially treated. Pre-Cp [creat] was again normal, but post-Cp values increased into the pathological range in the CD and DMIH, but not in NC patients. Most patients who developed azotaemia after the 3rd Cp administration demonstrated normalized [creat] after a few weeks, only few of them dropped-out at this phase. The 4th high-dose Cp was given to normal creatinine patients, but again, those in subgroups CD and DMIH developed azotaemia, which was not observed in the NC subgroup (Table 3). Serum urea concentrations followed a similar pattern to [creat] (not shown).

Figure 1 depicts that Cp-induced persistent azotaemia (Cp nephrotoxicity) developed in 7.5% of NC, in 20.9% of CD (p<0.05 vs NC) and in 30.8% of DMIH patients (p<0.01 vs NC). When the contribution of nephrotoxicity to overall fall-out from further Cp chemotherapy was examined, the ratio was 14% in NC, 38% in CD (p<0.01 vs NC) and 75% in DMIH patients (p<0.0001 vs NC and p<0.01 vs CD).

Figure 2 demonstrates eGFR-s of those patients who remained treatable by Cp before and after the 1st-4th cycles. Pre-Cp values moderately and successively decreased after Cp cycles but remained comparable in the three groups throughout the 1st-4th cycles. It seemed that eGFR as calculated by the Cockcroft-Gault equation was not sensitive enough in predicting higher vulnerability of the kidney in CD or DMIH patients. On the other hand, relative to the Cp-naive values eGFR before the 4th Cp cycle was diminished only by about 4% in NC, but 16 and 18% in CD and DMIH groups, respectively. Post-Cp eGFR-s, overall, followed an inverse pattern relative to [creat]-s. NC patients responded by significantly reduced eGFR only after the 1st, while the two comorbid groups on all cycles of Cp. Statistical analysis indicated that eGFR was more reduced (p<0.05) in CD and DMIH versus NC patients after the 2nd cycle.
DISCUSSION

The present study indicates that: 1) CD and/or DMIH are frequent among lung cancer patients; 2) these comorbidities predispose patients to Cp nephrotoxicity, while those elderly persons who are free from CD and DMIH are also quite resistant to the renal side effects of Cp; and 3) development of post-Cp azotaemia seems to be predictable by a moderate, but significant reduction of GFR as estimated by calculated Ccreat or measurement of 99mTc-DPTA clearance. Although it has been known that serum markers of renal function remain in the reference range when there has already been a decline of GFR [10], the present observations indicate that especially in patients suffering from CD or DMIH, normal [creat] misguide about the state of renal function or predictable renal tolerance towards the toxic effects of high-dose Cp. Estimation of Ccreat might provide more valuable information and predict more reliably the risk of nephrotoxicity.

Advanced age, CD or DM and especially their combination may increase the sensitivity of the kidney to the above toxic effects of Cp. Aging is associated with glomerulosclerosis and arteriosclerosis of intrarenal vessels leading to loss of functional nephrons [9]. Renal blood flow and GFR are reduced and many of the normal renal tubular absorptionSECRETION abilities are blunted. Silva et al has suggested [9] that aging should be seen as the erosion of generous spare capacity or loss of renal „safety margins”. The tubular systems becomes less capable of conserving/excreting NaCl [14]. Among the elderly exsiccation and/or hyponatraemia becomes prevalent [15]. In the aging kidney the number as well as the volume of tubules are reduced, the volume of the renal interstitium increases due to fibrosis which may be associated with inflammation [16]. Intrarenal arterial sclerosis is noted with increased frequency in patients aged 10-19, 20-39, 40-64 and above 65 years, respectively [17]. Important details have become uncovered about the physiologic mechanisms of kidney aging, as glomerular capillary hypertension, glomerulosclerosis, reduced number of nephrons and NO deficiency [18, 19]. Aging induces increased renal synthesis of ROS [20] and advanced glycation end products [21]. Biomarkers of senesence, like acute phase proteins accumulate in the aged kidney [22].

Chronic systemic hypertension [23] and diabetes mellitus [24, 25] accelerate aging of the kidney. Ohta et al [24] demonstrated higher pulse wave velocity, a sign of atherosclerosis, in main renal arteries and the renal interlobar arteries of middle-aged patients suffering from hypertension and diabetes mellitus. Hypertensive nephrosclerosis is associated with chronic ischaemic damage to the tubulointerstitium [27], the major target of Cp nephrotoxicity. In
addition to physiologic aging, intrarenal arterial sclerosis is accelerated by long-term hypertension and diabetes mellitus [18]. DM is complicated with early renal function decline [24, 25]. As a result of all these, the course of other primary renal diseases become accelerated (glomerulonephritis, acute renal failure, endotoxin-induced thrombosis [28, 29]. Accordingly, the development of azotaemia following high-dose Cp administration may betray underlying renal disease induced by CD or DM.

Based on the above data it is not surprizing that nephrotoxicity of high-dose Cp was exaggerated in aged lung cancer patients suffering from CD or DMIH. Underlying DMIH was associated with the greatest frequency of azotaemia following high-dose Cp. Since none of our patients developed acute or chronic renal failure or needed renal replacement therapy one could ask: is this moderately severe renal side effect of Cp so important clinically, once life expectancy of lung cancer patients is unfavourable anyway? The issue of Cp-induced azotaemia is very important, because: 1) during subsequent chemotherapy cycles lower dose or no Cp will be administered, 2) the Cp-induced glomerulotubular imbalance results enhanced urinary loss of Mg$^{2+}$, Ca$^{2+}$, K$^+$, Na$^+$, Cl$^-$ and water. These effects on electrolyte and water homeostasis may induce cardiac arrhythmia, circulatory shock and death.

There is a long list of agents which may ameliorate the nephrotoxicity of Cp in experimental animals [30]. The United State FDA has approved the organic thiophosphate amifostine (Ethyol) for use in patients receiving Cp [30, 31]. Reported efficacy of this tissue cytoprotector in cancer patients is not unequivocal [32] and the drug is expensive. The only available preventive measure of universally proven efficacy is hyperhydration by isotonic saline before, during and after the infusion of Cp [4]. To our knowledge, there has been no hydration protocol accepted and followed universally in lung cancer patients. The critical events seem to occur almost immediately - within short hours - after Cp administration [33]. Therefore, protective measures should be applied before, during and immediately after Cp infusion [33]. There have been no prospective, randomized studies for finding out what is appropriate hydration around Cp infusion in elderly people with underlying „subclinical” renal disease [34]. Companies manufacturing Cp recommend pre-Cp hydration enough to induce 100-200 ml/hour (2400-4800 ml/day) saline diuresis by the time of Cp infusion, but the recommended volume, velocity and timing of intravenous saline infusion before, during and after Cp seem not to be optimal for reaching the above goal. Although salt loading is more important than hydration [35], the exact definition what „saline diuresis” means has not been detailed either. We define it as voiding urine of ~1% NaCl concentration. Saline infusion, in contrast to water loading is only slowly followed by saline diuresis. The elapse of
1 hour after the start of 0.9% NaCl infusion of 1000 ml is probably insufficient for induction of saline diuresis. Of note, high urine flow (water diuresis) alone is not renoprotective against Cp [36], even more, the antitumor efficacy of Cp may be reduced by water loading [36]. Healthy adults consuming a Western diet with conventional salt intake (~3 grams per day) void 1000-1500 ml urine during 24 hours and urinary NaCl concentration is less than 0.4% [37]. Based on these estimations saline diuresis of a healthy adult may be not greater than ~50 ml/hour. Aged and anorexic cancer patients probably do have markedly reduced urinary saline excretion, which might have never been systematically tested. Diuretics administered for hypertension or other common reasons in elderly people may make these patients even more exsiccated at the time they arrive to chemotherapy (early morning). Ozols et al [38] and Townsend and Hanigan [39] administer a total volume of 6 liters of saline during the day of Cp administration. De Jong et al [7] infuse first 1 liter of 0.9% NaCl, then Cp in 250 ml volume of hypertonic (3%) NaCl during not less, than 3 hours (250 ml/≥3 hours), followed by 2 liters of 0.9% NaCl (~6 hours total treatment time). In 2006 at our Department high-dose Cp has been infused in 500 ml 0.9% saline (during about 30 minutes) after prior infusion of 1 liter 0.9% NaCl (~1 hour). Patients have received another 1.0-1.5 liter NaCl and 100 ml 20% mannitol after Cp. The total infused volume has been around 3-4 liters and administered within 3-4 hours. Thus it seems, we may have provided weaker pre-Cp hydration, administered Cp less protracted and certainly we have never controlled urinary volume or urinary Na⁺ concentration (saline diuresis) any time before, during or after Cp administration. The administration of mannitol has also been somewhat contradictory, since this agent might also have worsened preexisting renal dysfunction [40].

According to Hanigan et al [41] the infusion of saline protects kidney cells by increasing the osmolality, not the concentration of Na⁺ or Cl⁻ per se. These authors have documented that the in vitro toxicity of Cp in cultured proximal tubular cells could be equally prevented by increasing osmolality of the medium by mannitol, sucrose, sodium gluconate or NaCl. Cp was toxic in hypoosmolar media (~220 mosm/kg), but high-normal osmolality (~280 mosm/kg) provided nearly full protection. It has been hypothesized that change of osmolality induced change of cell volume, nuclear volume and consequent change of chromatin structure with less accessibility of Cp to DNA. It is considered also possible that the renal cellular metabolism of Cp, - which increases it’s toxicity - is altered by increased osmolality.
In 2008 the European Society of Clinical Pharmacy Special Interest Group on Cancer Care has formulated clear recommendations on the prevention of Cp nephrotoxicity [42]. The cornerstones of the recommendations are 1) the estimation of GFR or creatinine clearance using the abbreviated Modified Diet in Renal Disease (aMDRD) or the Cockcroft-Gault equation, 2) maintenance or induction of euvoalaemia before, during and after Cp, 3) the slow administration of Cp, 4) the maintenance of 3-4 /24 h saline diuresis during the preceding day and 2-3 days after Cp, 5) avoidance of diuretics including furosemide and mannitol.

In conclusion, before starting high-dose Cp chemotherapy of lung cancer patients, the risk of nephrotoxicity should always be evaluated by eGFR [42] and also based on coexistent CD or DMIH, because these conditions narrow the tolerance of the kidneys to Cp. The 2008 recommendations of the European Society of Clinical Pharmacy Special Interest Group on Cancer Care on the prevention of Cp nephrotoxicity [42] should be followed also regarding hydration by physiological saline before, during and after Cp administration in aged, multimorbid lung cancer patients. However, vigorous saline infusion of CD and DMIH patients may lead to volume overload and pulmonary edema. Further studies are needed for finding out, how to effectively prevent Cp nephrotoxicity without the harm of cardiac overload.

Acknowledgement
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References


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**Table 1.** Pre-Cisplatin glomerular filtration rate (GFR) of 38, initially non-azotaemic lung cancer patients. Twenty three patients developed Cp nephrotoxicity, 15 patients did not.

<table>
<thead>
<tr>
<th>subgroups</th>
<th>pre-Cp [creat] (μmol/l)</th>
<th>post-Cp [creat] (μmol/l)</th>
<th>pre-Cp GFR (ml/min/m²)</th>
<th>age (years)</th>
<th>man/women (number)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cp nephrotoxicity n=23</td>
<td>79±4*1</td>
<td>167±12*1</td>
<td>42.4±2.3*1</td>
<td>63.6±1.5ns1</td>
<td>13/10ns2</td>
</tr>
<tr>
<td>No Cp nephrotoxicity n=15</td>
<td>68±3</td>
<td>87±4</td>
<td>56.9±2.7</td>
<td>60.5±2.8</td>
<td>8/7</td>
</tr>
</tbody>
</table>

ns: p>0.05, *: p<0.05; 1: unpaired t-test 2: Fischer exact test (both 1 and 2 vs No Cp nephrotoxicity)

**Table 2.** Clinical data of patients receiving high-dose Cp for lung cancer (mean±SEM)

<table>
<thead>
<tr>
<th></th>
<th>NC (n=80)</th>
<th>CD (n=110)</th>
<th>DMIH (n=52)</th>
</tr>
</thead>
<tbody>
<tr>
<td>age (years)</td>
<td>56±1</td>
<td>60±1*1</td>
<td>62±1*1</td>
</tr>
<tr>
<td>male/female</td>
<td>45/35</td>
<td>66/44ns2</td>
<td>33/19ns2</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>24.3±0.4</td>
<td>25.4±0.4ns1</td>
<td>25.4±0.6ns1</td>
</tr>
<tr>
<td>dose of Cp per treatment (mg)</td>
<td>123±2</td>
<td>126±2ns3</td>
<td>124±4ns3</td>
</tr>
<tr>
<td>total dose of Cp</td>
<td>375±22</td>
<td>399±22ns1</td>
<td>392±43ns1</td>
</tr>
<tr>
<td>mean number of cycles</td>
<td>3.1±0.2</td>
<td>3.2±0.2ns3</td>
<td>3.2±0.3ns3</td>
</tr>
<tr>
<td>number of patients receiving gemcitabine/etoposide/taxol</td>
<td>38/41/1</td>
<td>52/55/3ns4</td>
<td>25/24/3ns4</td>
</tr>
<tr>
<td>blood pressure (mmHg)</td>
<td>132±2/81±1</td>
<td>134±2/81±1ns1</td>
<td>137±3/84±2ns1</td>
</tr>
<tr>
<td>heart rate (min⁻¹)</td>
<td>81±1</td>
<td>82±1ns1</td>
<td>86±2ns1</td>
</tr>
</tbody>
</table>

NC: no major comorbidity, CD: hypertension and ischemic heart disease, DMIH: diabetes mellitus and ischemic heart disease, *: p<0.05, ns: p>0.05 vs NC; 1: ANOVA with Newman-Keuls multiple comparison post hoc test, 2: Fischer exact test, 3: Kruskal-Wallis test with Dunn’s multiple comparison post hoc test, 4: Chi square test (all 1, 2, 3, 4 vs NC)
**Table 3.** Serum creatinine concentrations (μmol/l) in lung cancer patients receiving 1-4 cycles of high-dose Cp chemotherapy for lung cancer

<table>
<thead>
<tr>
<th>Cp cycle</th>
<th>NC</th>
<th>CD</th>
<th>DMIH</th>
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<tbody>
<tr>
<td>1st</td>
<td>n</td>
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<td>after</td>
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<td>77±1</td>
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<td></td>
<td>110</td>
<td>78±1ns</td>
<td>95±4*</td>
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<td></td>
<td>52</td>
<td>77±3ns</td>
<td>94±4*</td>
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<tr>
<td>2nd</td>
<td>n</td>
<td>before</td>
<td>after</td>
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<tr>
<td></td>
<td>68</td>
<td>80±2</td>
<td>88±4*</td>
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<tr>
<td></td>
<td>96</td>
<td>86±2ns</td>
<td>105±5*</td>
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<td></td>
<td>49</td>
<td>82±3ns</td>
<td>110±6*</td>
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<td>after</td>
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<td></td>
<td>42</td>
<td>85±3</td>
<td>91±5*</td>
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<td></td>
<td>64</td>
<td>91±3ns</td>
<td>113±5*</td>
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<td>83±4ns</td>
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<td>47</td>
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<td></td>
<td>30</td>
<td>89±3ns</td>
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</tbody>
</table>

NC: no major comorbidity, CD: hypertension and ischemic heart disease, DMIH: diabetes mellitus and ischemic heart disease, *: p<0.05 vs before (paired t-test); #: p<0.05, ns: p>0.05 vs NC (two-way Anova with Bonferroni posttest)
Figure legends

**Figure 1.** Frequency of nephrotoxicity and nephrotoxicity related drop-out from further Cisplatin treatment in lung cancer patients receiving 1-4 cycles of high dose Cisplatin and suffering from hypertension and ischemic heart disease (CD) or diabetes mellitus and ischemic heart disease (DMIH) or being free from these severe comorbidities (NC). *: p<0.05, **: p<0.01 vs NC; #: p<0.01 vs CD (two-tailed difference tests).

**Figure 2.** Pre- and post-Cisplatin eGFR values during the 1st-4th cycles of high dose Cisplatin treatments in lung cancer patients suffering from no major comorbidity (NC), from controlled hypertension and ischemic heart disease (CD) and controlled diabetes mellitus and ischemic heart disease (DMIH). Numbers in parentheses represent the number of patients. *: p<0.05 vs before (paired t-test); #: p<0.05, ns: not significant vs NC (two-way Anova with Bonferroni posttest). Numbers in parentheses represent the number of patients.
Figure 1.