ELSEVIER

European Journal of Pharmacology 378 (1999) 63–68

ejp

www.elsevier.nl/locate/ejphar

The effect of *cis*-Diamminedichloroplatinum II on Na^+ and K^+ transport in the rabbit cortical collecting duct

Ookawara Susumu *, Tabei Kaoru, Furuya Hiroaki, Asano Yasushi

Division of Nephrology, Department of Internal Medicine, Jichi Medical School, 3311-1 Minamikawachi-machi, Kawachi-gun, Tochigi, Japan

Received 28 January 1999; received in revised form 7 June 1999; accepted 11 June 1999

Abstract

cis-Diamminedichloroplatinum II (CDDP) is an antineoplastic drug against solid malignant tumors. However, its clinical use is limited by nephrotoxicity. CDDP also causes hypokalemia and in vivo microperfusion method have demonstrated that luminal CDDP increases K^+ secretion by hyperpolarization of the transepithelial voltage difference through stimulating Na^+ transport in the distal segments. However, there is no direct evidence for this mechanism. We therefore examined the effect of luminal CDDP on Na^+ and K^+ transport in the rabbit cortical collecting duct (CCD) using in vitro isolated tubular microperfusion. Luminal CDDP hyperpolarized the transepithelial voltage difference (V_T) in a dose-dependent manner at concentrations from 10^{-5} M to 10^{-3} M and at 10^{-3} M CDDP, V_T was hyperpolarized from -11.6 ± 2.3 mV to -16.6 ± 3.3 mV ($P < 0.001$). A concentration of 10^{-5} M ouabain, 10^{-4} M amiloride and 2 mM $BaCl_2$ all completely abolished CDDP-induced hyperpolarization. To confirm the mechanism, Na^+ and K^+ flux were measured in the presence of 10^{-3} M CDDP. CDDP decreased net K^+ secretion from -22.2 ± 5.7 to -15.2 ± 2.9 pmol mm^{-1} min^{-1} ($P < 0.01$) without any effect on the lumen-to-bath isotope flux of Na^+ (52.6 ± 10.6 to 52.1 ± 10.7 pmol mm^{-1} min^{-1}). These data suggest that luminal CDDP hyperpolarizes V_T primarily by inhibiting K^+ conductance but did not influence Na^+ transport of the luminal membrane. We conclude that the CCD does not play a role in CDDP-induced hypokalemia when CDDP is applied from the luminal side.

Keywords: Microperfusion; *cis*-Diamminedichloroplatinum II; K^+; Collecting duct

1. Introduction

cis-Diamminedichloroplatinum II (CDDP) is a potent chemotherapeutic drug used in a variety of human malignancies, including ovarian, testicular, bladder, head and neck, esophageal and small-cell lung cancers (Borch, 1987). However, its clinical use is limited by its pronounced nephrotoxicity. CDDP-induced kidney damage is characterized by a decrease in glomerular filtration rate (Daugaard et al., 1987; Safirstein et al., 1987), structural damage, including necrosis of the proximal tubule and collecting duct (Madias and Harrington, 1978; Dobyan et al., 1980; Goldstein and Mayor, 1983; Jones et al., 1985; Safirstein et al., 1985; Ward and Fauvie, 1976), mitochondrial injury in proximal tubules (Gordon and Gattone, 1986; Brady et al., 1990), and electrolyte disturbances, including hypokalemia and hypomagnesemia (Shilsky and Anderson, 1979; Blachley and Hill, 1981; Gordon et al., 1982; Jones and Chesney, 1995).

Regarding the electrolyte disturbances, experiments in isolated frog skin have demonstrated that CDDP increases the short-circuit current, transepithelial conductance of Na^+ and Cl^-, and transepithelial potential difference on the outer surface (Van der Berg et al., 1981).

On the basis of this study, Allen and Barratt, 1985 examined the effect of CDDP on the transepithelial voltage difference (V_T) of the distal segments of the rat kidney using in vivo microperfusion. CDDP hyperpolarized V_T in the late distal segments of the rat kidney tubule while amiloride prevented this change. The authors speculated that the increase in V_T across the late distal segments (cortical collecting duct (CCD)) during luminal perfusion with artificial plasma ultrafiltrate containing CDDP was consistent with stimulation of active Na^+ transport and secondarily induced renal K^+ wasting. However, there is no direct evidence for these changes in Na^+ and K^+ transport in distal segments of the rat kidney.

* Corresponding author. Tel.: +81-285-58-7346; fax: 81-285-44-4869

0014-2999/99/$ - see front matter
PII: S0014-2999(99)00434-3

Therefore, we examined the effect of luminal application of CDDP on Na^+ and K^+ transport in the rabbit CCD using in vitro isolated tubular microperfusion. Humoral and hemodynamic effects of CDDP can be completely disregarded when using this technique. Our results suggest that luminal CDDP hyperpolarized V_T by primarily inhibiting K^+ conductance but did not influence Na^+ transport of the luminal membrane. We conclude that the CCD does not play a role in CDDP-induced hypokalemia when CDDP is applied from the luminal side.

2. Materials and methods

2.1. Isolation and perfusion of tubules

CCDs were dissected from New Zealand white rabbits anesthetized with pentobarbital (50 mg kg^{-1} body weight). The CCD were placed in a dish containing cold intracellular fluid-like solution of the following composition: $NaHCO_3$, 10 mM; K_2HPO_4, 42.5 mM; KH_2PO_4, 15.0 mM; KCl, 15.0 mM; glucose 3.5 g dl^{-1}; Osm 375 mOsm/kg.H_2O at 4°C. The composition of this fluid is identical to that of Euro-Collins solutions, widely used for perfusion of living donor kidneys in renal transplantation. The medium was chosen because an intracellular fluid-like solutions is the preferred medium for preserving kidney tissue function (Pirie and Potts, 1986) and metabolism (Nagineri et al., 1987). Isolated tubular microperfusion was performed by the method previously described with minor modifications (Tabei et al., 1983; Ando et al., 1989). An artificial solution used both as perfusate and bathing medium had the following composition: NaCl, 105 mM; $NaHCO_3$, 25 mM; KCl, 5 mM; NaH_2PO_4, 1.6 mM; Na_2HPO_4, 0.4 mM; $CaCl_2$, 1.8 mM; Na acetate 10 mM; alanine 5 mM; pH was adjusted to 7.4 after bubbling with a 95% O_2/5% CO_2 gas mixture; osmolarity was adjusted to 298 mOsm/kg.H_2O. The pH of the bath medium and perfusate was checked with a pH meter (Model M-220, Corning, NY). V_T (mV) was measured using a standard technique (Tabei and Imai, 1986), with an electrometer (Model 610C, Keithley Instruments, Cleveland, OH) and a recorder (model R-202, Rikadenki, Tokyo, Japan).

After dissection, the CCD was transferred to a Lucite bath chamber and perfused in vitro. The bathing solution was exchanged constantly at 0.5 ml min^{-1} with an infusion pump (Terumo syringe pump STC-523, Terumo, Tokyo, Japan) throughout the experiment. To facilitate luminal perfusate exchange during each experiment, a polyethylene tube (PE-10; Clay-Adams, Parsippany, NJ) was placed inside *pipette B*, which was connected to the tubular lumen for perfusion. The luminal perfusate flow rate was adjusted by hydrostatic pressure. Luminal perfusate exchange was performed manually by injecting perfusate into the polyethylene tube, washing out the preexisting medium in *pipette B*.

Each tubule was allowed to equilibrate for 60–90 min before experiments, to exclude the effect of any endogenous vasopressin (Ando et al., 1991).

2.2. Net K^+ flux

K^+ transport was quantified as net flux by measuring the K^+ concentration of the perfusate and collected fluid. K^+ concentration was measured with a microanalysis flame photometer (AFA-707-D, APEL, Saitama, Japan). Net K^+ flux (JK) was calculated using the following equation (Tabei et al., 1995);

$$J_K = V_o(K_i - K_o)/L$$

where V_o denotes the collection rate obtained by dividing the constant-volume pipette volume by the collection time (nl $min-1$: all experiments were performed using the same volume pipette [75 nl]), K_i and K_o denote the K^+ concentrations of the perfusate and collection fluids, respectively, and L denotes tubule length (mm). In a preliminary study, we examined the effect of luminal CDDP at a concentration of 10^{-3} M on water flux in the same protocol; water reabsorption rate in the control period was 0.09 ± 0.07 nl min^{-1} mm^{-1} and in the experimental period it was 0.08 ± 0.06 nl min^{-1} mm^{-1} when the perfusion rate was 8.4 ± 1.1 and 9.0 ± 0.9 nl min^{-1}, respectively ($n = 6$, no significant difference between the two). Therefore, we did not measure water flux thereafter, and water flux was not included in this equation.

2.3. Unidirectional isotope flux

In addition to measuring the net K^+ flux under various conditions, the lumen-to-bath unidirectional ^{22}Na flux was also measured. $^{22}NaCl$ (New England Nuclear, Boston, MA) was added to the perfusate for measurement of the unidirectional efflux coefficient. The isotope count was obtained using a gamma scintillation counter (Packard auto-gamma 5650, Packard Japan, Tokyo, Japan). The unidirectional efflux coefficient was calculated using the following equation (Furuya et al., 1992);

$$K_e = V_o(1 - \log(C_i/C_o))/A$$

where K_e denotes the efflux coefficients of Na^+ and C_i and C_o denote the isotope count of the perfusate and collected fluid, respectively (cpm nl^{-1}), and A denotes the surface area of the luminal membrane, using a nominal mean internal diameter of 25 mm.

2.4. Experimental protocol

In all experiments, tubule length was more than 1 mm, and the perfusion rate was kept at 4–8 nl min^{-1} by changing the height of the reservoir. To confirm the effect

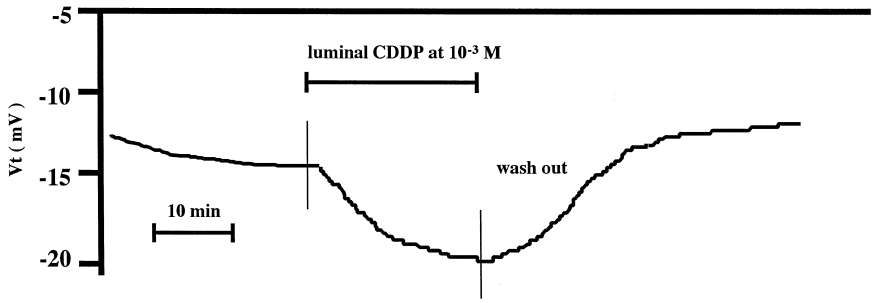

Fig. 1. Effect of 10^{-3} M luminal CDDP on changes of V_T in the rabbit CCD. Original trace is shown.

of luminal CDDP, after an initial 60–90 min equilibration period, 10^{-3} M CDDP was added to the perfusate. V_T was monitored for 20 min and the recovery phase was monitored thereafter. Comparing the half-time for onset of hyperpolarization with the half-time for recovery, they appear to be equal. To examine the dose-dependence of the effect of luminal CDDP, CDDP in concentrations ranging from 10^{-5} M to 10^{-3} M was subsequently added to the perfusate for 20 min each and V_T change was monitored continuously.

To clarify the mechanism of the effect of luminal CDDP on K^+ transport, luminal 2 mM $BaCl_2$ (K^+ channel inhibitor), luminal 10^{-4} M amiloride (Na^+ channel inhibitor and Na^+–H^+ exchanger inhibitor) and basolateral 10^{-5} M ouabain (Na^+–K^+ ATPase inhibitor) were added after 60–90 min of equilibration. CDDP at a concentration of 10^{-3} M was then added to the perfusate with each agent.

For the measurement of net K^+ flux and unidirectional ^{22}Na flux, after four control collections, 10 min after the addition of CDDP at a concentration of 10^{-3} M in the perfusate, three or four experimental samplings were performed.

2.5. Drugs

CDDP and other chemicals were obtained from Sigma (St. Louis, MO). CDDP was prepared daily in dimethyl sulfoxide. Experimental additions represented an increase in solution volume $< 0.1\%$.

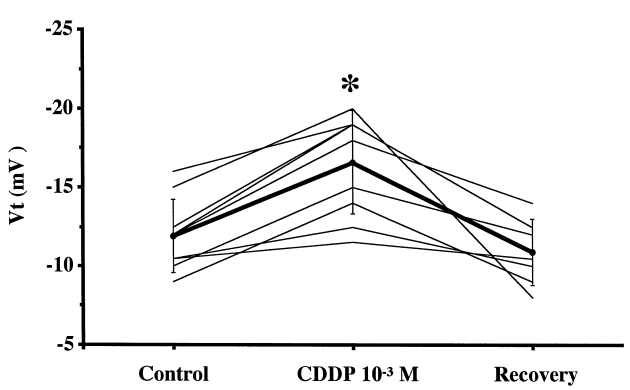

Fig. 2. Effect of 10^{-3} M luminal CDDP on changes of V_T in the rabbit CCD. Luminal CDDP hyperpolarized V_T from -11.9 ± 2.3 to -16.6 ± 3.3 mV ($n = 9$, $^*P < 0.001$) with significant recovery to -11.0 ± 2.1 mV ($n = 7$, $P < 0.001$).

2.6. Statistical analysis

All values are presented as means $\pm$ S.D. Differences between two groups were evaluated by paired *t*-test and those between three groups by factorial Analysis of Variance (ANOVA) and Scheffe's test at a significance level of 5%.

3. Results

As shown Fig. 1, luminal CDDP at 10^{-3} M always induced hyperpolarization of V_T. This change of V_T was from -11.9 ± 2.3 to -16.6 ± 3.3 mV ($n = 9$, $P < 0.001$) with complete recovery to -11.0 ± 2.1 mV ($n = 7$, $P < 0.001$, Fig. 2). To examine the dose dependence of the effect of luminal CDDP, it was subsequently added to the perfusate in a range of concentrations from 10^{-5} M to 10^{-3} M. Under control conditions, V_T was -10.5 ± 3.5 mV ($n = 6$) and not affected by luminal CDDP at a

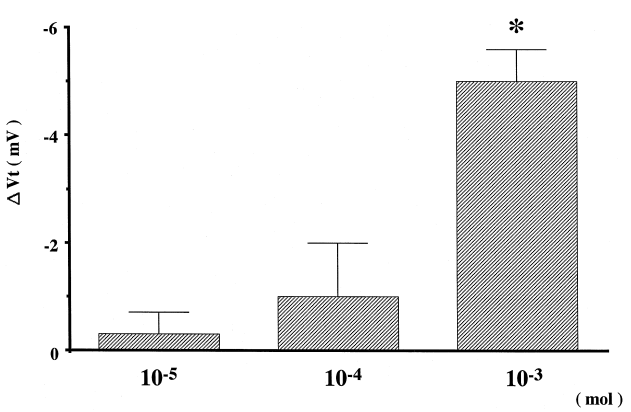

Fig. 3. The dose-dependent effect of luminal CDDP on changes in V_T in the rabbit CCD. Luminal CDDP at a concentration of 10^{-5} M did not change V_T (ΔV_T; -0.3 ± 0.4 mV, $n = 6$) and at 10^{-4} M it hyperpolarized V_T slightly but not significantly (ΔV_T; -1.0 ± 1.0 mV, $n = 6$). However, 10^{-3} M CDDP significantly hyperpolarized V_T (ΔV_T; -5.0 ± 0.6 mV, $n = 6$, $^*P < 0.0001$ vs. control).

concentration of 10^{-5} M (ΔV_T: -0.3 ± 0.4 mV, Fig. 3). However, at 10^{-4} M CDDP, V_T was slightly but not significantly hyperpolarized (ΔV_T: -1.0 ± 1.0 mV) and at 10^{-3} M CDDP, V_T was significantly hyperpolarized (ΔV_T: -5.0 ± 0.6 mV, $P < 0.0001$ vs. control). In further experiments, we therefore used 10^{-3} M CDDP.

In order to clarify the mechanisms by which luminal CDDP hyperpolarized V_T, the effects of various blockers on the action of CDDP were examined (Fig. 4). After a control period, 10^{-5} M ouabain was added to the bathing medium, and then 10^{-3} M CDDP was added to the perfusate. Ouabain significantly depolarized V_T, but addition of CDDP with ouabain did not further change V_T (control: -16.4 ± 8.2 mV, ouabain: 2.9 ± 2.5 mV, CDDP with ouabain: 1.8 ± 2.3 mV, ouabain vs. CDDP with ouabain: NS, $n = 5$). In the same way, luminal 10^{-4} M amiloride significantly depolarized V_T, but addition of CDDP with amiloride did not further change V_T (Control: -9.9 ± 9.0 mV, amiloride: 4.3 ± 0.6 mV, CDDP with amiloride: 3.2 ± 0.7 mV, amiloride vs. CDDP with amiloride: NS, $n = 5$). Luminal 2 mM $BaCl_2$ significantly hyperpolarized V_T and addition of CDDP with $BaCl_2$ did not further change V_T (control: -11.6 ± 2.8 mV, $BaCl_2$: -20.9 ± 6.0 mV, CDDP with $BaCl_2$: -21.1 ± 6.3 mV, $BaCl_2$ vs. CDDP with $BaCl_2$: NS, $n = 6$). These findings suggest that CDDP induces hyperpolarization by stimulating ouabain- and/or amiloride-sensitive Na^+ pathways or by inhibition of barium-sensitive K^+ transport.

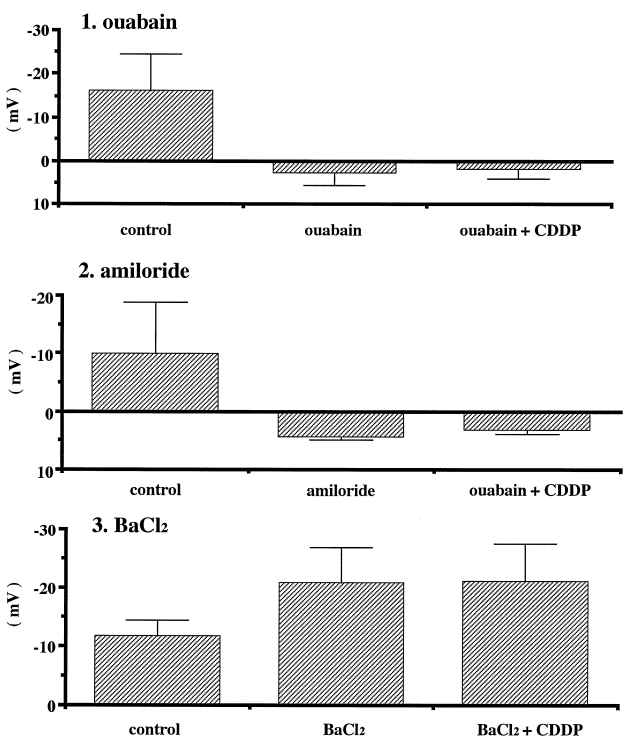

Fig. 4. Effect of 10^{-3} M luminal CDDP with various channel blockers on changes of V_T in the rabbit CCD. Although basolateral ouabain significantly depolarized V_T from -16.4 ± 8.2 to 2.9 ± 2.5 mV ($n = 5$, $P < 0.001$), subsequent exposure of CDDP to ouabain did not further change V_T (1.8 ± 2.3 mV, $n = 5$). Luminal amiloride significantly depolarized V_T from -9.9 ± 9.0 to 4.3 ± 0.6 mV ($n = 5$, $P < 0.001$), but did not further change after subsequent exposure to CDDP (3.2 ± 0.7 mV, $n = 5$). Luminal $BaCl_2$ significantly hyperpolarized V_T from -11.6 ± 2.8 to -20.9 ± 6.0 mV ($n = 5$, $P < 0.01$) with no effect of subsequent exposure to CDDP (-21.1 ± 6.3 mV, $n = 6$).

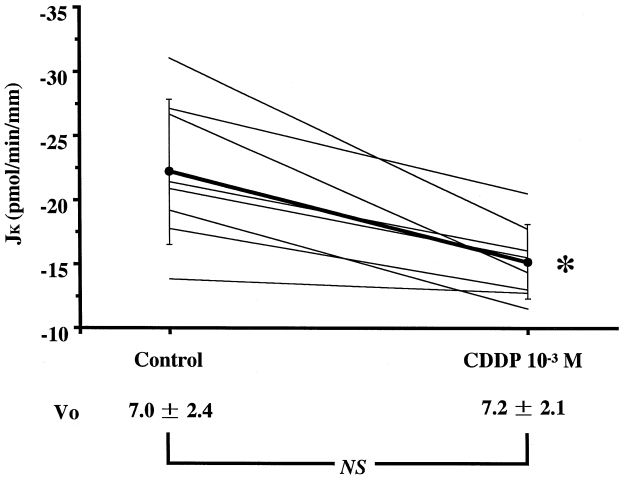

Fig. 5. Effect of 10^{-3} M luminal CDDP on net potassium secretion rate in the rabbit CCD. Luminal CDDP significantly decreased the net K^+ secretion rate from -22.2 ± 5.7 to -15.2 ± 2.9 pmol min^{-1} mm^{-1} ($n = 8$, $^*P < 0.01$).

To clarify the detailed mechanism, we next examined the effect of luminal CDDP on K^+ and Na^+ transport. The response of K^+ and Na^+ transport to 10^{-3} M luminal CDDP in CCD is shown in Figs. 5 and 6. CDDP at 10^{-3} M decreased net K^+ secretion rate from -22.2 ± 5.7 to -15.2 ± 2.9 pmol mm^{-1} min^{-1} ($n = 8$, $P < 0.01$). However, CDDP did not significantly change the lumen-to-bath isotope flux of Na^+ (52.6 ± 10.6 to 52.1 ± 10.7 pmol mm^{-1} min^{-1}, $n = 4$, NS).

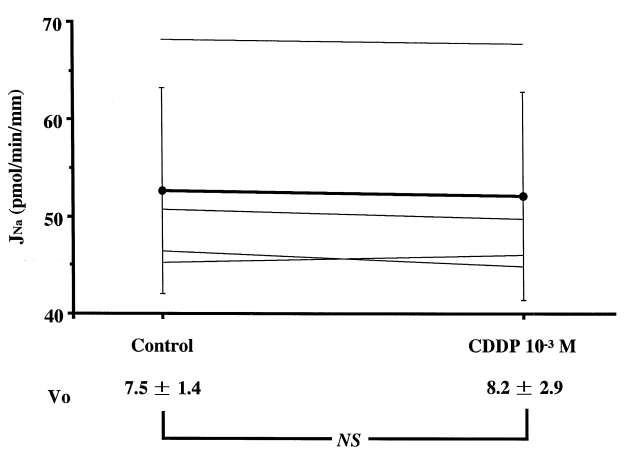

Fig. 6. Effect of 10^{-3} M luminal CDDP on ^{22}Na lumen-to-bath unidirectional flux in the rabbit CCD. Luminal CDDP did not affect ^{22}Na lumen-to-bath unidirectional flux (52.6 ± 10.6 pmol min^{-1} mm^{-1} in controls and 52.1 ± 10.7 pmol min^{-1} mm^{-1} in the experiments, $n = 4$).

4. Discussion

CDDP is a potent anticancer agent that revolutionized the treatment of various solid tumors (Borch, 1987). However, the dose used in cancer chemotherapy is limited by nephrotoxic side effects. CDDP-induced acute renal failure is associated with focal, primarily sublethal, cell injury throughout the proximal tubule. Injured cells, especially those in the S3 segments, undergo a variety of changes including loss of the brush border, alterations in mitochondrial configuration, and development of swollen endoplasmic reticulum together with increased numbers of vacuoles and segregated nuclei (Jones et al., 1985). Mitochondrial injury is thought to be central to CDDP toxicity in the proximal tubule because CDDP has been shown to cause loss of cytosolic K^+ and a fall in oxygen consumption, both of which reflect mitochondrial function including Na^+–K^+ ATPase activity and ATP utilization (Brady et al., 1990). Furthermore, it has been reported that CDDP induces morphological changes (dilatation of basolateral space) in the CCD and functional abnormalities (various electrolyte disturbances including hypokalemia, hypomagnesemia and hypocalcemia) (Shilsky and Anderson, 1979; Blachley and Hill, 1981; Gordon et al., 1982; Goldstein and Mayor, 1983; Jones and Chesney, 1995; Jones et al., 1985).

Van der Berg et al. (1981) reported that CDDP applied to the outer surface of frog skin epithelium increased active Na^+ transport by increasing the permeability of the outer cell membrane to Na^+. At the same time it decreased transepithelial electrical resistance in part by its effects on the outer cell membrane, but it also increased the permeability or conductance of the shunt pathway. Furthermore, in experiments using in vivo microperfusion of the distal segments of the rat kidney, after application of luminal CDDP, V_T was significantly hyperpolarized and amiloride abolished the change of V_T induced by CDDP (Allen and Barratt, 1985). These investigators suggested that luminal CDDP stimulated amiloride-sensitive Na^+ channels and speculated that, by increasing the V_T in the distal segments of the kidney, CDDP would favor voltage-dependent K^+ movement into the tubular lumen and thus contribute to renal K^+ wasting. However, there is no direct evidence for this mechanism.

Our studies were designed to observe the acute effects of CDDP applied from the tubular lumen. It is well known that CDDP exists in two forms depending upon the chloride concentration of medium. When CDDP enters the cell and is exposed to the low cytosolic chloride contents, it loses chloride and becomes charged and trapped in the cell. On the other hand, at the outer membrane surface exposed to a high chloride medium as it was in this study, CDDP is stable and uncharged.

In the present study, although CDDP hyperpolarized V_T in the same way as in the experiments using in vivo microperfusion (Allen and Barratt, 1985), this hyperpolarization was abolished by luminal $BaCl_2$ or amiloride, and by basolateral ouabain. These findings suggest that the hyperpolarization induced by CDDP is caused by inhibition of luminal K^+ transport or stimulation of amiloride-sensitive Na^+ channels or basolateral Na^+–K^+ ATPase. To examine whether K^+ or Na^+ transport is involved in the effect of CDDP, we directly measured net K^+ secretion rate and lumen-to-bath unidirectional isotope Na^+ flux. CDDP significantly decreased net K^+ secretion rate but did not change the lumen-to-bath unidirectional isotope Na^+ flux. These data suggest that CDDP induced the hyperpolarization not through stimulation of amiloride-sensitive Na^+ transport or basolateral Na^+–K^+ ATPase but by inhibition of barium-sensitive K^+ conductance. Scott et al. (1995) reported that acute application of 5×10^{-6} M CDDP attenuated voltage-activated K^+ currents under all conditions from -60 mV to 70 mV by approximately 50% within minutes in cultured dorsal root ganglion neurons. Our results for the inhibition of K^+ conductance are consistent with this report and suggest that CDDP can interact with membrane constituents to modify the underlying K^+ channel activity.

Although we found that 10^{-3} M CDDP directly inhibits barium-sensitive K^+ channels, the question arises as to whether such high concentrations of CDDP occur in vivo in the tubular lumen. When CDDP was given intravenously to dogs at a dose of 3 mg kg^{-1}, urinary platinum concentrations reached more than 10^{-3} M (Cvitkovic et al., 1977), under certain conditions, CDDP concentration in the luminal fluid of the CCD could reach 10^{-3} M or more.

In summary, CDDP applied from the tubular lumen hyperpolarized V_T and this change was abolished by application of luminal $BaCl_2$. At the same time, CDDP decreased net K^+ secretion rate but had no effect on Na^+ transport in the rabbit CCD. The mechanism of CDDP-induced hyperpolarization may be to suppress barium-sensitive K^+ transport on the luminal side. However, the mechanism of CDDP-induced hypokalemia still remains unclear and further investigations are required.

Acknowledgements

We thank Mrs. Hiromi Kasakura and Miss Yukie Mashiyama for their expert technical and secretarial help.

References

Allen, G.G., Barratt, L.J., 1985. Effect of cisplatin on the transepithelial potential difference of rat distal tubule. Kidney Int. 27, 842–847.

Ando, Y., Tabei, K., Furuya, H., Asano, Y., 1989. Glucagon stimulates chloride transport independently of cyclic AMP in the rat medullary TAL. Kidney Int. 31, 760–767.

Ando, Y., Tabei, K., Asano, Y., 1991. Luminal vasopressin modulates transport in the rabbit collecting duct. J. Clin. Invest. 88, 952–959.

Blachley, J.D., Hill, J.B., 1981. Renal and electrolyte disturbances associated with *cis*-platin. Ann. Int. Med. 95, 628–632.

Borch, R.F., 1987. The platinum anticancer drugs. In: Powis, G., Prough, R.A. (Eds.), Metabolism and Action of Anticancer Drugs. Taylor and Francis, London, pp. 163–193.

Brady, H.R., Kone, B.C., Storomski, M.E., Zeidel, M.L., Giebisch, G., Gullans, S.R., 1990. Mitochondrial injury: an early event in cisplatin toxicity to renal proximal tubules. Am. J. Physiol. 258, F1181–1187, Renal Fluid Electrolyte Physiol. 27.

Cvitkovic, E., Spaulding, J., Bethune, V., Martin, J., Whitmore, W.F., 1977. Improvement of *cis*-dichlorodiammineplatinum (NSC 119875): therapeutic index in an animal model. Cancer 39, 1357–1359.

Daugaard, G., Abildgaard, U., Holstein-Rathlou, N.H., Amptorp, O., Oleson, H.P., Ceynac, P.P., 1987. Functional and histopathological changes in dog kidneys after administration of cisplatin. Renal. Physiol. 10, 54–64.

Dobyan, D.C., Levi, J., Jacob, C., Kosek, J., Weiner, N.W., 1980. Mechanism of *cis*-Platinum nephrotoxicity: 2. Morphologic observations. J. Pharmacol. Exp. Ther. 213, 551–556.

Furuya, H., Tabei, K., Muto, S., Asano, Y., 1992. Effect of insulin on potassium secretion in the rabbit cortical collecting duct. Am. J. Physiol. 262, F30–35, Renal Fluid Electrolyte Physiol. 29.

Goldstein, R.S., Mayor, G.H., 1983. The nephrotoxicity of cisplatin. Life. Sci. 32, 685–690.

Gordon, J.A., Gattone, V.H., 1986. Mitochondrial alterations in cisplatin-induced acute renal failure. Am. J. Physiol. 250, F991–998, Renal Fluid Electrolyte Physiol. 19.

Gordon, J.A., Peterson, L.N., Anderson, R.J., 1982. Water metabolism after cisplatin in the rat. Am. J. Physiol. 243, F36–43.

Jones, D.P., Chesney, R.W., 1995. Renal toxicity of cancer chemotherapeutic agents in children: ifosfamide and cisplatin. Curr. Opin. Pediatr. 7, 208–213.

Jones, T.W., Chopra, S., Kaufman, J.S., Flamenbaum, W., Trump, B.F., 1985. *cis*-diamminedichloroplatinum II-induced acute renal failure in the rat. Lab. Invest. 52, 363–374.

Madias, N.E., Harrington, J.Y., 1978. Platinum nephrotoxicity. Am. J. Med. 65, 307–314.

Nagineri, C.N., Leveille, P.J., Lee, D.B.N., Yanagawa, N., 1987. Isolation of cells from rabbit renal proximal tubules by using a hyperosmolar intracellular-like solution. Biochem. J. 223, 353–358.

Pirie, S.C., Potts, D.J., 1986. A comparison of the relative effectiveness of three transplant preservation fluids upon integrity and function of rabbit proximal convoluted tubules perfused in vitro. Clin. Sci. Lond. 70, 443–452.

Safirstein, R., Winston, J., Goldstein, M., Moel, D., Dickman, S., Guttenplan, J., 1985. Cisplatin nephrotoxicity. Am. J. Kidney. Dis. 8, 356–367.

Safirstein, R., Winston, J., Moel, D., Dicman, S., Guttenplan, J., 1987. Cisplatin nephrotoxicity: insights in mechanism. Int. J. Androl. 10, 325–346.

Scott, R.H., Woods, A.J., Lacey, M.J., Fernando, D., Crawford, J.H., Andrews, P.L., 1995. An electrophysiological investigation of the effects of cisplatin and the protective actions of dexamethasone on cultured dorsal root ganglion neurones from neonatal rats. Naunyn-Schmiedeberg's. Arch. Pharmacol. 352, 247–255.

Shilsky, R.L., Anderson, T., 1979. Hypomagnesemia and renal Mg wasting in patients receiving *cis*-platin. Ann. Int. Med. 90, 929–931.

Tabei, K., Imai, M., 1986. K transport in upper portion of descending limb of long-loop nephron from hamsters. Am. J. Physiol. 252, F387–392, Renal Fluid Electrolyte Physiol. 21.

Tabei, K., Levenson, D.J., Brenner, B.M., 1983. Early enhancement of fluid transport in rabbit proximal straight tubules after loss of contralateral renal excretory function. J. Clin. Invest. 72, 871–881.

Tabei, K., Muto, S., Furuya, H., Sakairi, Y., Ando, Y., Asano, Y., 1995. Potassium secretion is inhibited by metabolic acidosis in rabbit cortical collecting ducts in vitro. Am. J. Physiol. 268, F490–495, Renal Fluid Electrolyte Physiol. 37.

Van der Berg, E.K. Jr., Brazy, P., Huang, A.T., Dennis, V.W., 1981. *cis*-platin-induced changes in sodium, chloride, and urea transport by the frog skin. Kidney. Int. 19, 8–14.

Ward, J., Fauvie, K.A., 1976. The nephrotoxic effects of *cis*-Diamminedichloroplatinum II (NSC119875) in male F344 rats. Toxicol. Appl. Pharmacol. 38, 535–547.