

and the cortical collection duct (cCD). Both segments were relatively resistant to the toxic effects of luminal inorganic mercury ( $^{203}\text{Hg}$ , 9.3 $\mu\text{M}$  for the cCD and 20 $\mu\text{M}$  for cTAL) when compared to its acute toxic effects on the proximal tubule, which are prominent cellular uptake of vital dye, cellular swelling, marked luminal membrane blebbing and a great increase in the leakiness of the tight junctions. The cCD showed no acute toxic effects from luminal mercury while the cTAL was sensitive to the presences of the luminal inorganic mercury with some slight blebbing of the luminal membrane and an increase in the leakiness of the tight junctions, but there was no cellular swelling nor cellular uptake of the vital dye. The transport of luminal inorganic mercury was measured by its disappearance rate from the lumen (JD,  $\text{fmol min}^{-1}$  ( $\text{mm tubular length}^{-1}$ )), its appearance rate in the bath (JA,  $\text{fmol min}^{-1}$  ( $\text{mm tubular length}^{-1}$ )) and by cellular accumulation ( $\text{fmol}$  ( $\text{mm tubular length}^{-1}$ )). The JD was 33.5  $\pm$  2.8 and 81.6  $\pm$  9.5 for the cCD and cTAL respectively, while the JA was 9.3  $\pm$  2.1 and 70.0  $\pm$  17.4 for the cCD and cTAL respectively. The asymmetry of the JD and JA within each segment may be due to cellular uptake of the mercury, which was 1, 342  $\pm$  347 and 1, 526  $\pm$  164 in the cCD and cTAL respectively. For both the cCD and cTAL, approximately 80% of the cellular accumulated mercury was associated with the tubular structure and 20% with the cytoplasm. We conclude that the cCD and the cTAL can transport inorganic mercury from the luminal fluid. A significant fraction of the accumulated mercury in the cell may be due to non-specific binding. Also, these segments are relatively resistant to the acute toxic effects of inorganic mercury.

**977** MERCURIC ION ( $\text{Hg}^{2+}$ ) INCREASES THE SENSITIVITY OF KIDNEY CELLS TO APOPTOSIS BY INHIBITING NUCLEAR FACTOR- $\kappa\text{B}$  (NF- $\kappa\text{B}$ ) ACTIVATION.

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NF- $\kappa\text{B}$  is a thiol-dependent transcriptional factor that promotes cell survival and protects from apoptotic stimuli. Recently, we reported (TAP 173:176-187, 2001) that  $\text{Hg}^{2+}$ , one of the strongest thiol-binding agents known, impairs NF- $\kappa\text{B}$  activation and transcriptional activity in normal rat kidney epithelial (NRK52E) cells at low concentrations (< 20  $\mu\text{M}$ ) by binding to specific reduced thiol moieties in the NF- $\kappa\text{B}$  activation pathway. Since NF- $\kappa\text{B}$  prevents apoptosis, we hypothesized that attenuation of NF- $\kappa\text{B}$  activation by  $\text{Hg}^{2+}$  may increase the sensitivity of kidney cells to apoptotic agents to which kidney cells are otherwise resistant because of their NF- $\kappa\text{B}$  activating capacity. In untreated cells, fewer than 5% were apoptotic when evaluated by DNA fragmentation (TUNEL) or flow cytometric DNA profile analyses.  $\text{Hg}^{2+}$  (5  $\mu\text{M}$ ) treatment for 24 hrs increased this proportion by 1.5- to 2-fold. Neither lipopolysaccharide (LPS) (1  $\mu\text{g}/\text{ml}$ ) nor tumor necrosis factor- $\alpha$  (TNF) (100 U/ml), potent inducers of NF- $\kappa\text{B}$ , altered the proportion of apoptotic cells, compared with untreated controls. However, when LPS or TNF were given following  $\text{Hg}^{2+}$  treatment (5  $\mu\text{M}$  for 30 min.), the proportion of cells undergoing apoptosis at 22 hrs increased by 4- to 6-fold over that seen following LPS or TNF alone. In contrast,  $\text{Hg}^{2+}$  pretreatment did not increase the amount of apoptosis caused by apoptosis-inducers that do not activate NF- $\kappa\text{B}$  (staurosporine, Fas ligand). These findings support the view that  $\text{Hg}^{2+}$  enhances the sensitivity of kidney cells to apoptotic stimuli as a consequence of inhibition of NF- $\kappa\text{B}$  activity. Since apoptosis is known to play a key role in the pathogenesis of renal failure resulting from toxicant injury to proximal tubular cells, promotion of apoptosis *via* inhibition of NF- $\kappa\text{B}$  activity may define a novel molecular mechanism by which  $\text{Hg}^{2+}$  toxicity is initiated in kidney cells. Supported by ES04696 and ES07033 from the NIH.

**978** DIFFERENTIAL SENSITIVITY TO CHEMICALLY INDUCED INJURY IN PRIMARY CULTURES OF RENAL EPITHELIAL CELLS FROM CONTROL AND UNINEPHRECTOMIZED RATS.

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Primary cultures of renal proximal tubular (PT) and distal tubular (DT) cells from rats that had undergone uninephrectomy and compensatory renal growth (NPX) retain changes in morphology, enzyme and transport activities, and mitochondrial function that occur in kidneys of NPX rats. Acute cytotoxicity of tert-butyl hydroperoxide (tBH; 0.1, 0.5, 1, 5, 10 mM) was determined by measurement of lactate dehydrogenase (LDH) release in both freshly isolated and primary cultures of renal PT and DT cells from control and NPX rats. tBH-induced death of both cell types from NPX rats was greater than that in corresponding cells from control rats (net LDH release = 33.6% vs. 53.7% for PT cells from control and NPX; 10.8% vs. 62.1% for DT cells from control and NPX). tBH-induced malondialdehyde formation was 2- to 4-fold greater in PT and DT cells from NPX rats than in cells from control rats. Similarly, tBH and KCN (0.1-1 mM; a mitochondrial toxicant) both induced significantly greater cell death in confluent primary cultures of PT

and DT cells from NPX rats than in corresponding cells from control rats. For inorganic Hg (5, 10, 50  $\mu\text{M}$ ), however, cytotoxic effects in cells from NPX rats were similar to those in corresponding cells from control rats at the two lower concentrations but were markedly higher at the highest concentration. Uptake of inorganic Hg (0.1, 1, 5  $\mu\text{M}$ ) across the brush-border and basolateral membranes of PT and DT cells grown on filter inserts was actually lower in cells from NPX rats at the two lower doses but was higher at the highest dose as compared to measurements in cells from control rats. These results indicate a general greater sensitivity of cells from NPX rats to chemical toxicants than cells from control rats. For inorganic Hg, however, more complex dose-response relationships exist between transport and toxicity. (Supported by NIEHS Grant ES05157, ES05980, and DK40725.)

**979** IS MERCURY IN URINE INDICATIVE OF EXPOSURE TO LOW LEVELS OF MERCURY VAPOR?

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Exposure to elemental mercury has received increased attention as a result of ubiquitous use, improved analytical detection, and awareness of health effects. Although urinary mercury levels are considered the best predictor of mercury vapor exposures, a lack of relationship has been reported between airborne and urinary mercury levels at low air levels (i.e., <50  $\mu\text{g}/\text{m}^3$ ). We reviewed the literature on the correlation of mercury in air and urine to evaluate whether airborne mercury exposures at health-based action levels (e.g., 1  $\mu\text{g}/\text{m}^3$ ) are measurable in urine above background. A meta-analysis was conducted of ten studies reporting paired air and urine mercury data (149 samples total) and meeting specified criteria for data quality and sufficiency. The log-transformed dataset showed a strong correlation between mercury in air and in urine ( $r=0.77$ ), although the relationship was best fit by a series of parallel lines with different intercepts for each study ( $R^2=0.81$ ). Predicted ratios of air to urine mercury levels at 50  $\mu\text{g}/\text{m}^3$  air concentration ranged from 1:1 to 1:3. Toward the lower end of the dataset (i.e., 10  $\mu\text{g}/\text{m}^3$ ), predicted urinary mercury levels encompassed two distinct ranges: values on the order of 20  $\mu\text{g}/\text{L}$  and 30-60  $\mu\text{g}/\text{L}$ . Extrapolation to 1  $\mu\text{g}/\text{m}^3$  resulted in predicted urinary levels of 4-5 and 6-13  $\mu\text{g}/\text{L}$ . Higher predicted urine mercury levels were associated with use of static area air samplers by some studies, rather than more accurate personal air samplers. Urinary mercury predictions based primarily on personal air samplers at 1 and 10  $\mu\text{g}/\text{m}^3$  are consistent with reported mean (4  $\mu\text{g}/\text{L}$ ) and upper-bound (20  $\mu\text{g}/\text{L}$ ) background levels, respectively. Thus, although mercury levels in air and urine are correlated below 50  $\mu\text{g}/\text{m}^3$ , the impact of mercury in air below 10  $\mu\text{g}/\text{m}^3$  is likely indistinguishable from background urinary mercury levels.

**980** MERCURIC CHLORIDE SUPPRESSES CYCLOOXYGENASE-2 EXPRESSION AND PROSTAGLANDIN PRODUCTION IN A HUMAN MONOCYTIC CELL LINE.

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We examined the effects of a brief treatment of low-dose mercuric chloride on the activation of arachidonic acid metabolism in the human monocyte cell line THP-1. HPLC analyses revealed that LPS treatment stimulated induction of cyclooxygenase-2 expression and production of prostaglandin E2 (a major arachidonate metabolite) in these cells. Unstimulated THP-1 cells express the constitutive cyclooxygenase-1 isoform and a low level of cyclooxygenase-2, with a basal production of prostaglandin E2. THP-1 cells were pre-treated with 1 microM mercuric chloride for one hour, and then were incubated with LPS (1 microg/ml) for 4 hours. Cell viability was not affected by treatment with inorganic mercury, as assessed by trypan blue exclusion. Treatment with mercuric chloride suppressed both the basal level and LPS-stimulated expression of cyclooxygenase-2, as measured by Western-blot analysis. Correspondingly, EIA analysis revealed that both basal and LPS-stimulated prostaglandin E2 synthesis was attenuated by mercuric chloride treatment. We did not observe alteration of cyclooxygenase-1 expression. These results demonstrate an effect of mercury on attenuating the arachidonic acid pathway in activated monocytes/macrophages. Modulation of this pathway by inorganic mercury suggests that part of the adverse effects of exposure to low levels of this metal may be suppression of inflammatory and immune responses.

**981** EFFECTS OF METHYLMERCURY EXPOSURE ON THE IMMUNE AND NERVOUS SYSTEM RESPONSES OF CBA/J MICE TO A CHRONIC TOXOPLASMA GONDII INFECTION.

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Previous studies from our laboratory suggest that exposure to methyl mercury (MeHg) does not increase the susceptibility to acute toxoplasmosis in CBA/J mice. Therefore, we investigated endpoints associated with immunotoxicity and neuro-