Drug-induced nephrotoxicity is a major concern, since many pharmacological compounds are filtered through the kidneys for excretion into urine. To discover biochemical biomarkers useful for early identification of nephrotoxicity, metabolic experiments were performed on Sprague-Dawley CD rats treated with the nephrotoxicant gentamicin, captopril or tobramycin. Using a combination of gas chromatography/mass spectrometry (GC/MS) and liquid chromatography/mass spectrometry (LC/MS), a global metabolite identification analysis was performed on urine and kidney samples collected after 1, 5 and 28 days dosing. Increases in polyamines and arabinose were observed in urine from drug-treated rats after a single dose, and prior to observable histological kidney damage and conventional clinical chemistry indications of nephrotoxicity. Thus, these metabolites are potential biomarkers for the early detection of drug-induced nephrotoxicity. Upon prolonged dosing, drug-induced-nephrotoxic changes included a progressive loss of arabinose in urine, concurrent with a decrease in arabinose and glucosamine in kidney tissue. A nephrotoxicity prediction model, based on the levels of branched-chain amino acids in urine, distinguished nephrotoxicant-treated samples from vehicle-control samples with 100% accuracy at day 28 and 90% accuracy at day 1, respectively. Thus, this panel of biomarkers may provide a noninvasive method to detect kidney injury long before the onset of histopathological kidney damage.

Abstraction

The kidney is one of the primary sites of drug biotransformation and this development of many drugs is due to unknown kidney toxicity in clinical studies. Routinely used measures of renal function, such as blood urea nitrogen (BUN) and serum creatinine, have assess limitations. They are non-regional specific and they increase significantly only after substantial kidney injury occurs. Therefore, ladder and earlier biomarkers for kidney injury are highly needed for pre-clinical drug development and to use in humans. Gentamicin, captopril and tobramycin treat three well-characterized nephrotoxins. Gentamicin, the prototype aminoglycoside antibiotic, induces proximal tubular necrosis and serum creatinine, have severe limitations. They are not region-specific and they increase significantly only after substantial kidney injury occurs. Therefore, ladder and earlier biomarkers for kidney injury are highly needed for pre-clinical drug development and to use in humans. Gentamicin, captopril and tobramycin treat three well-characterized nephrotoxins. Gentamicin, the prototype aminoglycoside antibiotic, induces proximal tubular necrosis and serum creatinine, have severe limitations. They are not region-specific and they increase significantly only after substantial kidney injury occurs. Therefore, ladder and earlier biomarkers for kidney injury are highly needed for pre-clinical drug development and to use in humans. Gentamicin, captopril and tobramycin treat three well-characterized nephrotoxins.

Materials and Methods

Groups of Sprague-Dawley CD rats were treated with a single dose of one of 3 nephrotoxins, or vehicle control (Figure 1). Urine and kidney tissue samples were collected after 1, 5 and 28 days after drug dosing. The metabolite extracts were then ionized in positive or negative modes to achieve full abundant separation of metabolites (Figure 2). The metabolite extracts were then ionized in positive or negative modes to achieve full abundant separation of metabolites. Metabolites were identified by comparison to library entries in a metabolomics standard library. Statistical analysis of metabolite data was performed with QIAP (SKA, http://www.skacq.com) and R (http://cran.r-project.org). With a p value of 0.05 we considered a metabolite significantly changed.

Results and Discussion

Histopathological and Clinical Chemistry

Histopathology and clinical chemistry data were collected, so that metabolic changes can be correlated with kidney tissue damage. Histopathological kidney damage was observed for all three nephrotoxins after 28 days of dosing, but not at day 1. Higher BUN and creatinine values were observed in females and in main treatment with captopril for 28 days. No creatinine or BUN changes were observed for gentamicin and tobramycin.

Conclusions

Purine and Pyrimidine Nucleosides Decreased in Kidney Tissue

Treatment with the 3 nephrotoxins caused a significant decrease in purine and pyrimidine nucleosides in kidney tissue, including inosine, x-ammonium, adenosine, adenosine, guanosine and uridine. These decreases may reflect altered activity of renal transporters, altered synthesis/breakdown rates of nucleotides, or altered filtering by the kidneys resulting from the nephrotoxic drug action.

Early Nephrotoxicity Prediction Modeling

Classification models were built for the 38 urine metabolites from nephrotoxicant samples. Top 5 classifiers were leucine, histidine, isoleucine, valine and valine. A nephrotoxicity prediction model, based on the levels of branched-chain amino acids in urine, distinguished nephrotoxicant-treated samples from vehicle-control samples with 100%, 90% and 10% accuracy at day 28, day 5 and 1 day, respectively (Table 1).

Table 1. Nephrotoxic-treated samples can be distinguished from vehicle-treated samples prior to day 28.

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In summary, we identified early biomarkers of nephrotoxicity that can detect tubular kidney damage much earlier than histopathology and BUN or creatinine.

REFERENCES