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A new toxic biomarker of anticancer agent cisplatin

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Cisplatin is one of the most active drugs for the treatment of cancers. However, cisplatin-induced nephrotoxicity is the most common problem. Although the counteracting apoptotic and survival pathways are balanced as the injured cells either undergo apoptosis or proliferate, the switch molecule of this balance is unclear. This study investigated the expression of Phlda3 as a potential biomarker of renal tubular injury induced by cisplatin in vivo and in vitro, and its functional role in cisplatin-induced toxicity. Phlda3 transcript and protein levels were highly up-regulated in the kidney of mice after cisplatin treatment, which preceded increases in BUN or serum creatinine. Particularly, the level of Phlda3 transcript was substantially increased at an early time, and remained elevated. Treatment of NRK52E with cisplatin caused increases in Phlda3 mRNA and protein. Knockdown of Phlda3 reversed the decrease in cell viability by cisplatin. Cisplatin treatment elicited p53 accumulation via Akt/Mdm2 repression. Phlda3 deficiency attenuated a decrease in Akt phosphorylation by cisplatin, and prevented p53 accumulation. Hence, Phlda3, which leads to p53-mediated tubular cell death, may be an early and sensitive biomarker of cisplatin-induced kidney injury. To assess the effect of Phlda3 inhibition on the anti-cancer effect of cisplatin, we determined whether Phlda3 knockdown diminishes its anti-cancer effect. Cisplatin elicited SKOV-3 cell death, which was not reversed by Phlda3 knockdown, implying that the method to inhibit Phlda3 may be of use in decreasing kidney toxicity without losing the anti-cancer effect of cisplatin.

AS9-2

Epigenetic alterations in human urothelial cells under sustained arsenic exposure in culture

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We have previously demonstrated that long-term low-dose sodium arsenite exposure induced cellular transformation and aberrant gene expression in human urothelial cell lines. Since epigenetic alterations play critical roles in the regulation of gene expression, we performed systematic analysis to compare the DNA methylation profiles between arsenic-exposed human urothelial cells and untreated cells. By aid of the Infinium Assay, we selected 61 genes with promoter methylation differences between parental and arsenic-exposed cells over 75%. Among them, 40 genes showed hypomethylated in arsenic-exposed cells, whereas 21 genes hypermethylated. To identify genes whose expression associated with DNA methylation, we performed the quantitative real-time PCR analysis after treatment of cells with 5-aza-2'-deoxycytidine. The results showed that 76% of hypomethylated gene expression levels in arsenic-exposed cells are higher than parental cells. However, only 42% hypermethylated genes consistently expressed lower levels of transcripts in arsenic-exposed cells than control cells. Bisulfite sequencing analysis was performed to confirm the methylation status in arsenic-exposed cells. In all of the genes analyzed in this study, lipocalin-2 (LCN2), also known as oncogene 24p3 or NGAL, is one of the most highly expressed in long-term arsenic-exposed cells as compared to untreated cells. By siRNA technique, we found that silencing of LCN2 in arsenic-exposed cells exhibited reduced oncogenic potential and decreased production of pro-inflammatory cytokines. Taken together, our present results showed that long-term low dose arsenic exposure could be through epigenetic mechanism to alter the gene expression profile and hence induce cell transformation.