Supporting Information

Abundant rodent furan-derived urinary metabolites are associated with tobacco smoke exposure in humans

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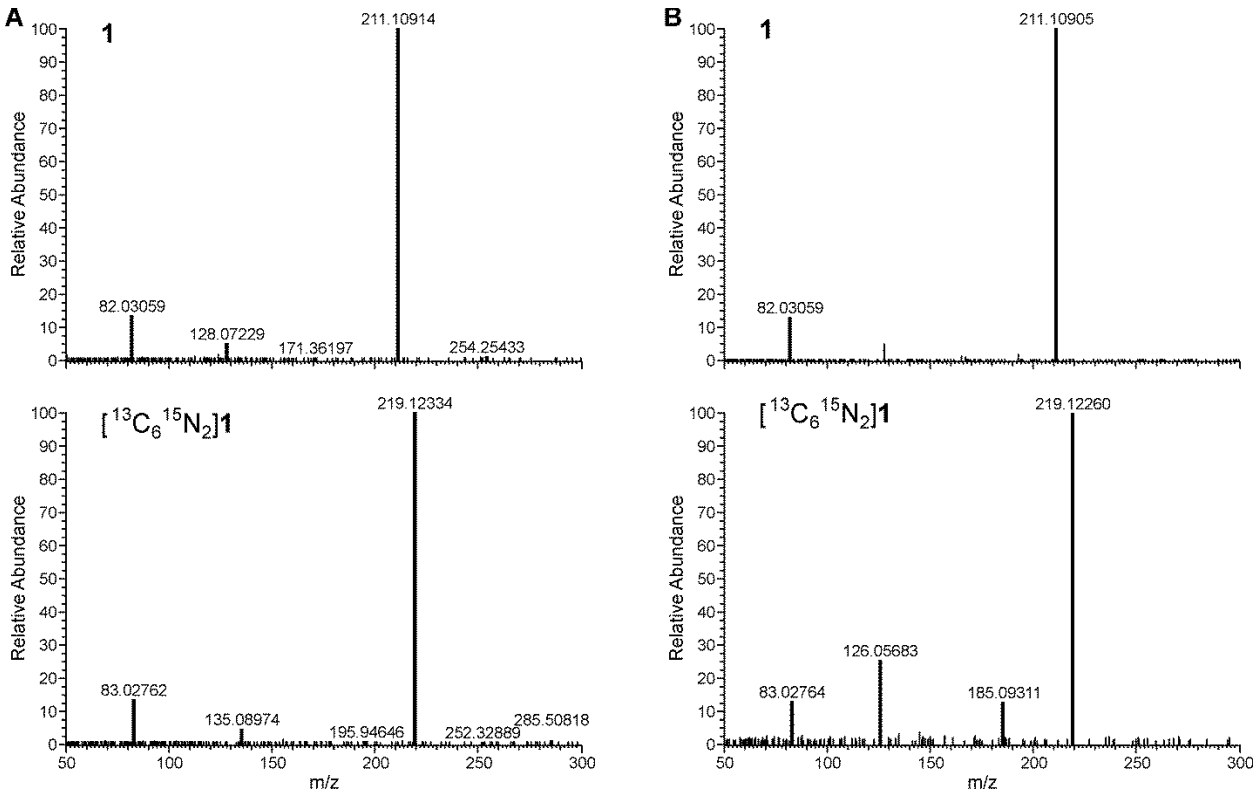

Figure S1. Representative high resolution mass spectra of A) the synthetic standard for furan metabolite **1** and B) metabolite **1** in human urine. [$^{13}C_6{}^{15}N_2$]**1** is the internal standard.

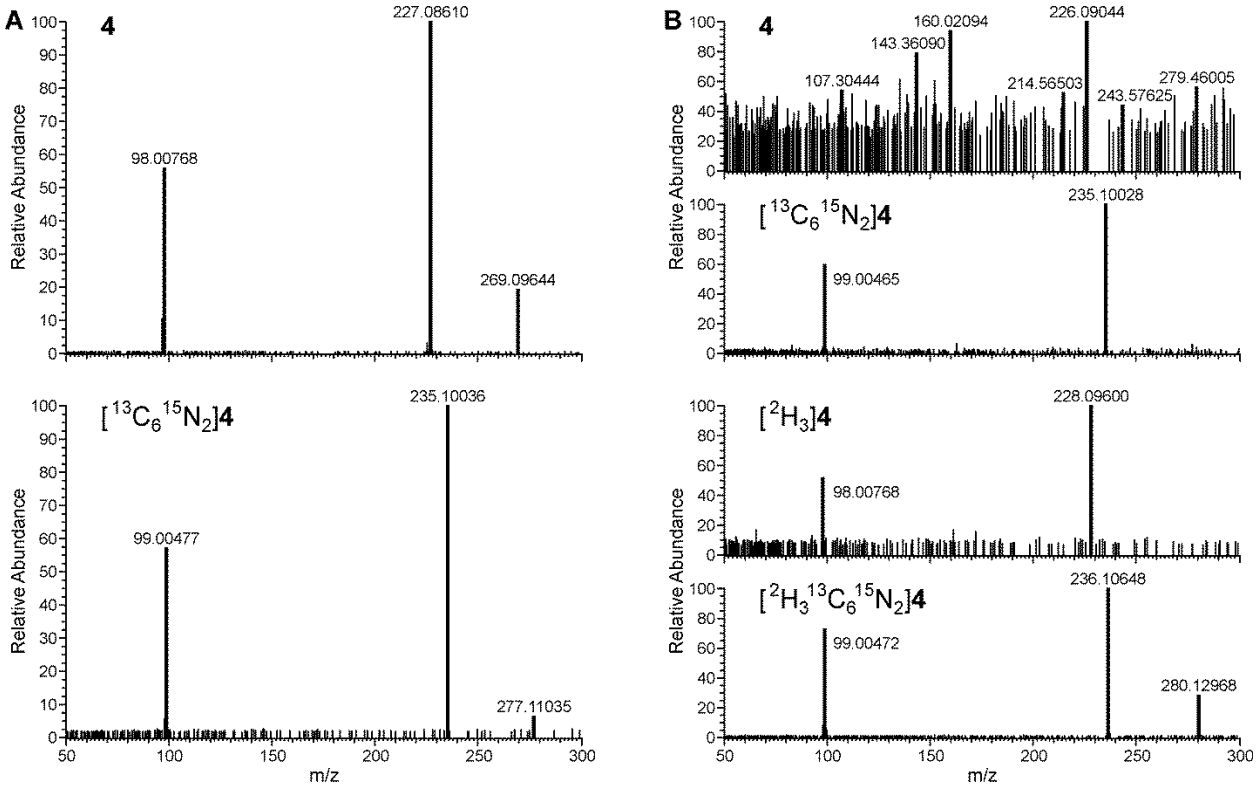

Figure S2. Representative high resolution mass spectra of A) synthetic standard for furan metabolite **4** and B) metabolite **2** as [$^{2}H_{3}$]**4** and metabolite **4** in human urine. Levels of metabolite **4** were not sufficiently high to get a good high resolution mass spectrum. [$^{13}C_{6}{}^{15}N_{2}$]**4** and [$^{2}H_{3}{}^{13}C_{6}{}^{15}N_{2}$]**4** are the internal standards.

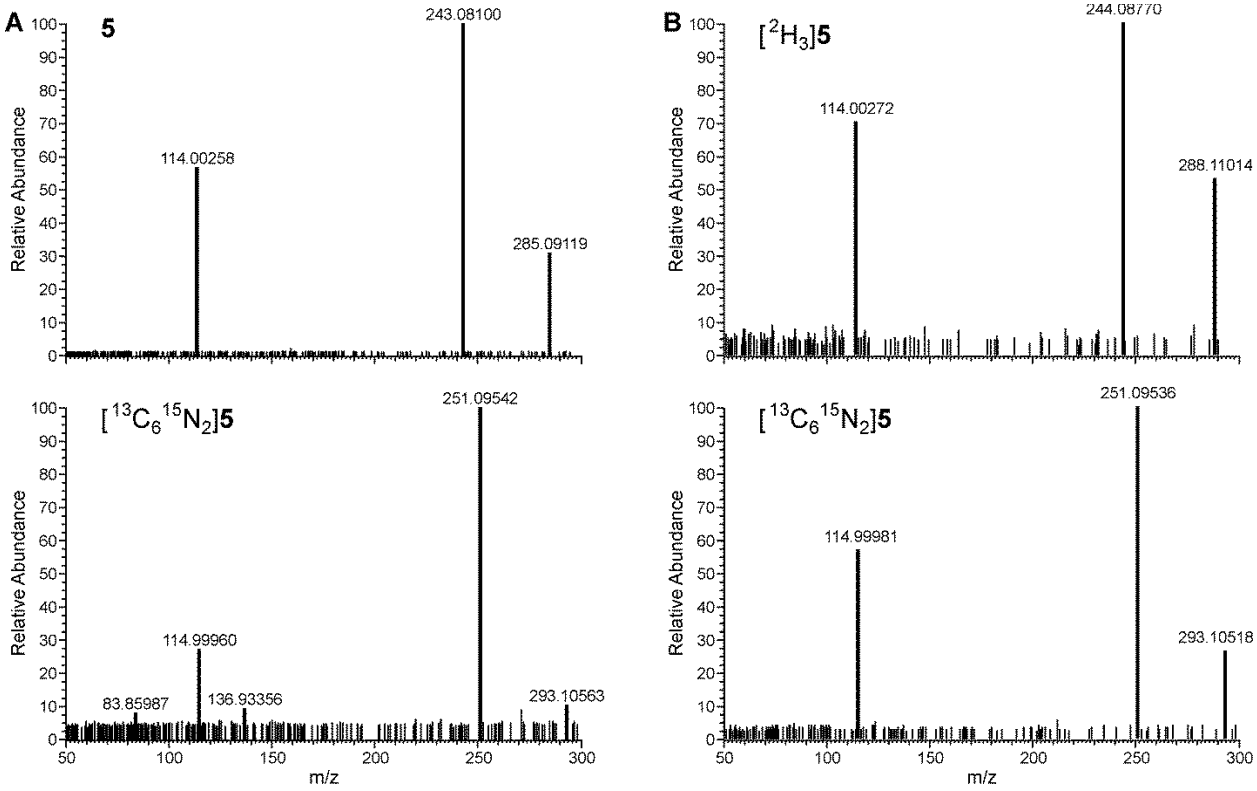

Figure S3. Representative high resolution mass spectra of A) synthetic standard for furan metabolite **5** and B) metabolite **3** as [$^{2}H_{3}$]**5** in human urine. [$^{13}C_{6}{}^{15}N_{2}$]**5** is the internal standard.

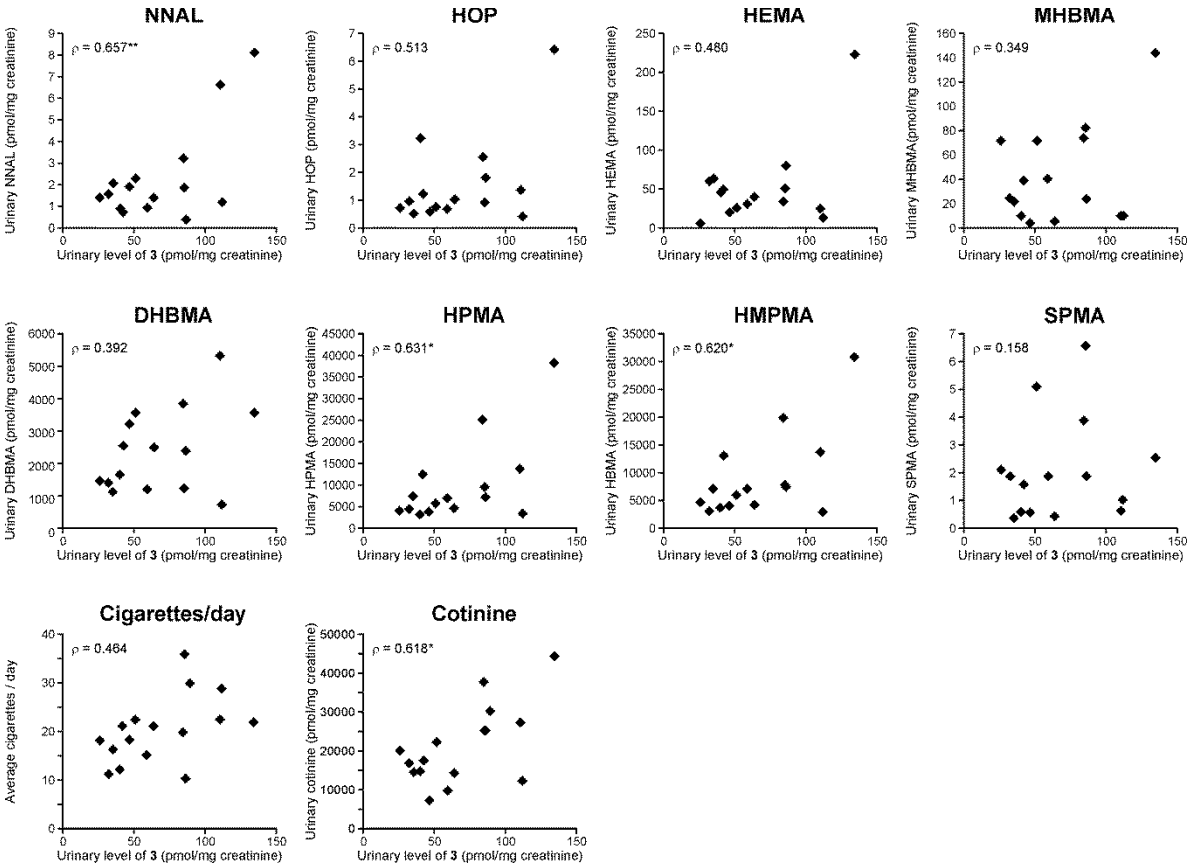

Figure S4. Comparison of metabolite **3** levels with previously measured tobacco smoke exposure biomarkers: 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol (NNAL), 1-hydroxypyrene (HOP), 2-hydroxyethyl mercapturic acid (HEMA), 1-(*N*-acetylcysteinyl)-2-hydroxy-3-butene (MHBMA), 1,2-dihydroxy-4-(*N*-acetylcysteinyl)butane (DHBMA), 1-(*N*-acetylcysteinyl)-propan-3-ol (HPMA), 2-(*N*-acetylcysteinyl)butan-4-ol (HMPMA), and (*N*-acetylcysteinyl)benzene (SPMA). Degree of correlation was assessed using the Pearson's product moment correlation coefficient (ρ). A *p*-value of < 0.05 was considered significant. **p*-value = 0.014, ***p*-value = 0.003

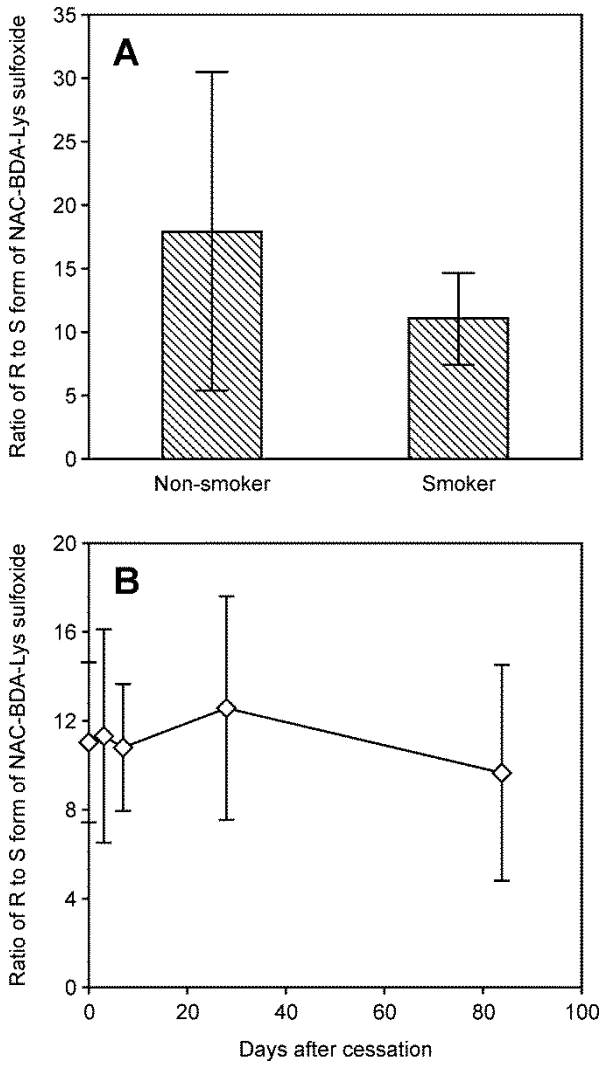

Figure S5. Ratio of the R to S sulfoxide diastereomers of metabolite **3** in human urine. Ratios were determined by analyzing LC-MS/MS chromatograms of human urine from non-smokers and smokers (A) and smokers after cessation (B). Data is plotted as average R to S ratio ± S.D.

Table S1. Analysis characteristics of furan metabolites in human urine			
Metabolite	Limits of detection (fmol)	Average ± S.D. (pmol/mL)	Precision[a] (%CV)
1	4	330 ± 100[b]	31%
2 (as [$^2H_3$]**4**)	4	26 ± 9	35%
3 (as [$^2H_3$]**5**)	4	45 ± 3	6%
4	4	0.73 ± 0.22	30%
5	4	not detected	

[a]Precision was measured by repeatedly processing pooled smoker urine at the same time as other samples and determining the standard deviation of the metabolite levels (n = 12). [b]n = 11