Supporting Information

Isoform Selective Inactivation of Human Arylamine N-Acetyltransferases by Reactive Metabolites of Carcinogenic Arylamines

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Table of Contents……. **Page S1**

Table S1. Effect of AcCoA on N-OH-AAF and N-OH-4-AABP Inactivation of NAT1 and NAT2 in HeLa Cell Cytosol [a]

NAT1	nmol/mg/min	% Control Activity
Control	4.34 ± 0.13	100 ± 3
50 μM N-OH-AAF	1.77 ± 0.28	41 ± 7
AcCoA [b] + 50 μM N-OH-AAF	3.67 ± 0.21	85 ± 5
50 μM N-OH-4-AABP	1.68 ± 0.35	39 ± 8
AcCoA [b] + 50 μM N-OH-4-AABP	3.58 ± 0.70	83 ± 16
NAT2	nmol/mg/min	% Control Activity
Control	0.15 ± 0.006	100 ± 4
10 μM N-OH-AAF	0.05 ± 0.01	38 ± 9
AcCoA [c] + 10 μM N-OH-AAF	0.13 ± 0.02	94 ± 11
20 μM N-OH-4-AABP	0.06 ± 0.02	41 ± 11
AcCoA [c] + 20 μM N-OH-4-AABP	0.13 ± 0.01	90 ± 9

[a] Results are expressed as the means (± SD) of three experiments.

[b] The AcCoA concentration was 400 μM.

[c] The AcCoA concentration was 800 μM.

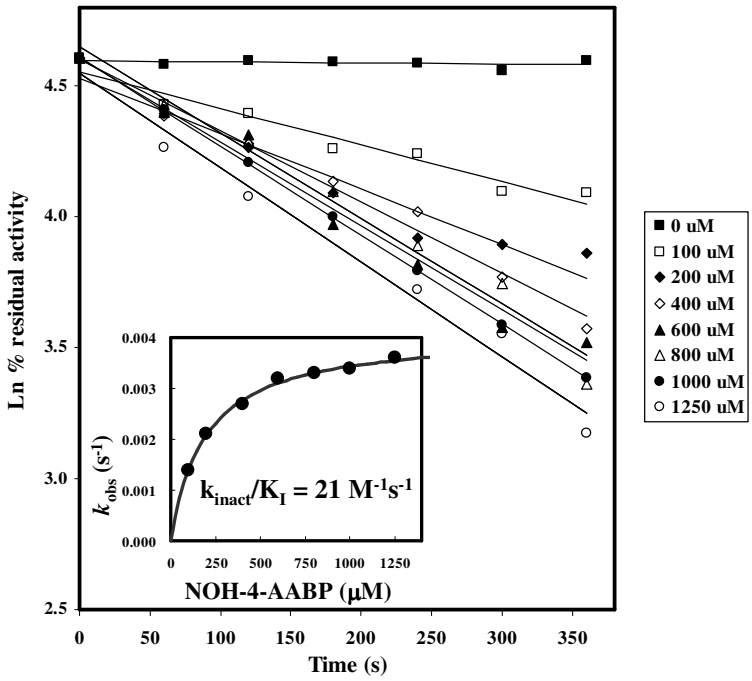


Figure S1. Time- and concentration-dependent inactivation of recombinant human NAT2 by N-hydroxy-4-acetylaminobiphenyl (N-OH-4-AABP). The results represent the mean of two experiments.

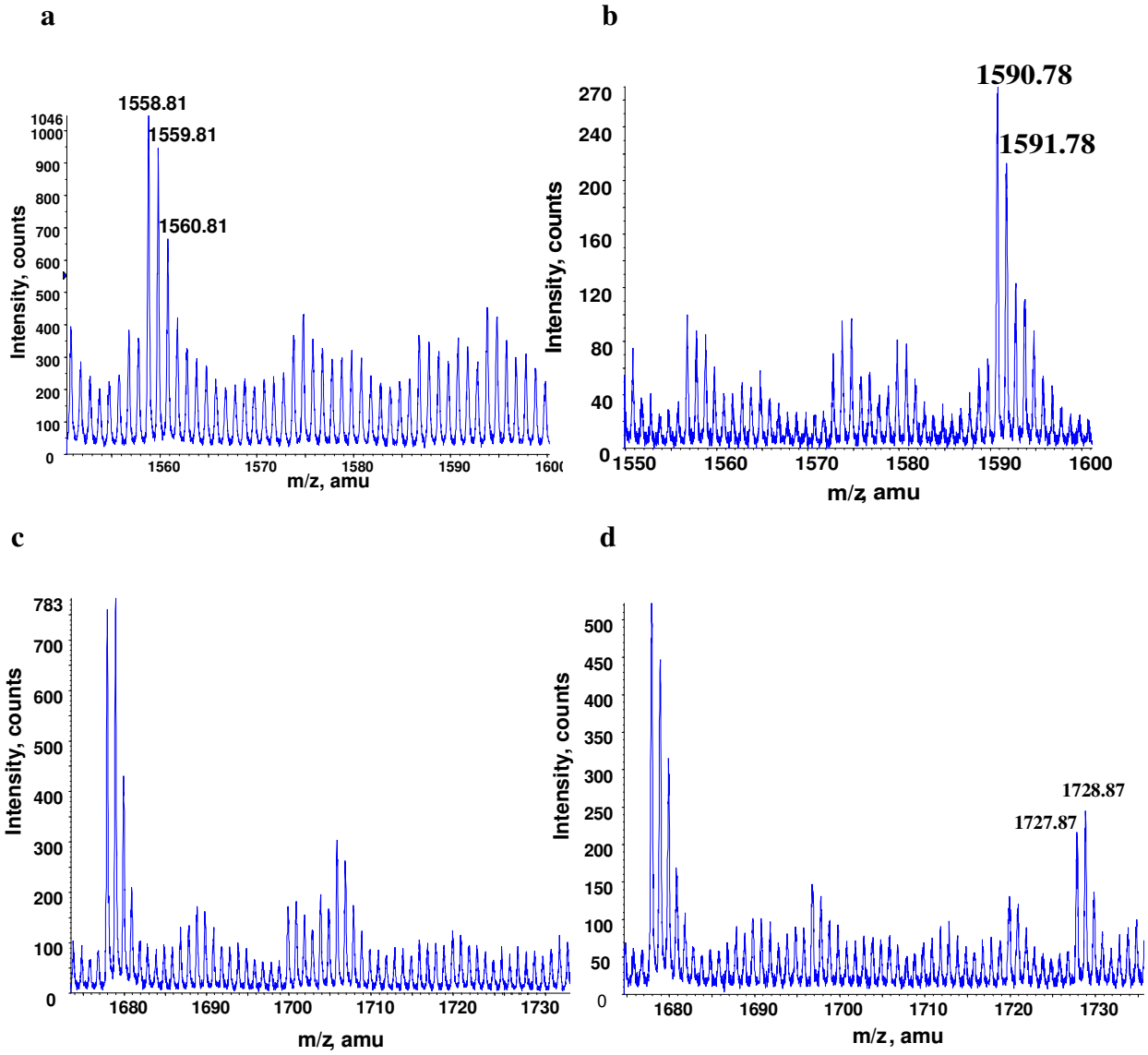
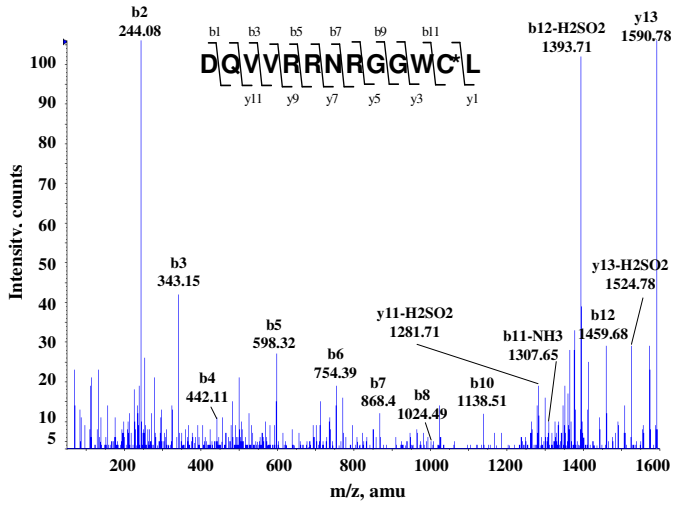


Figure S2. Segments of the MALDI Q-TOF mass spectra of pepsin digests of (a, and c) native human NAT1; (b, and d) N-hydroxy-2-acetylaminofluorene (N-OH-AAF)-inactivated NAT1.

a



b

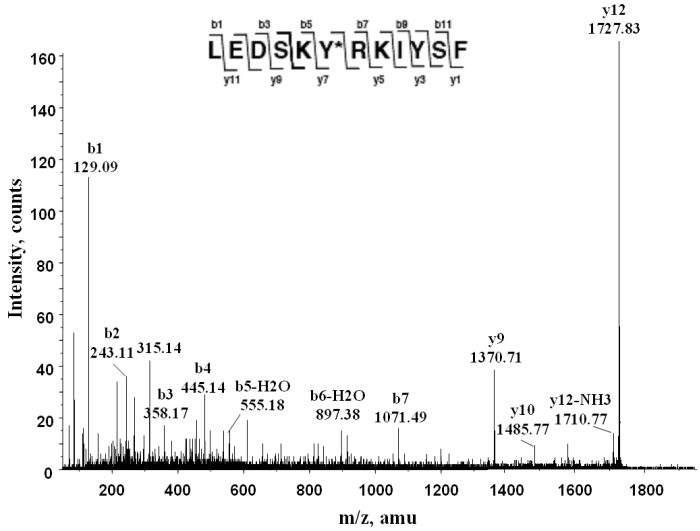


Figure S3. MALDI Q-TOF tandem mass spectra: (a) 1590.78 Da peptide obtained by pepsin digestion of N-OH-AAF-inactivated NAT1, (b) 1727.87 Da peptide obtained by pepsin digestion of N-OH-AAF-inactivated NAT1.

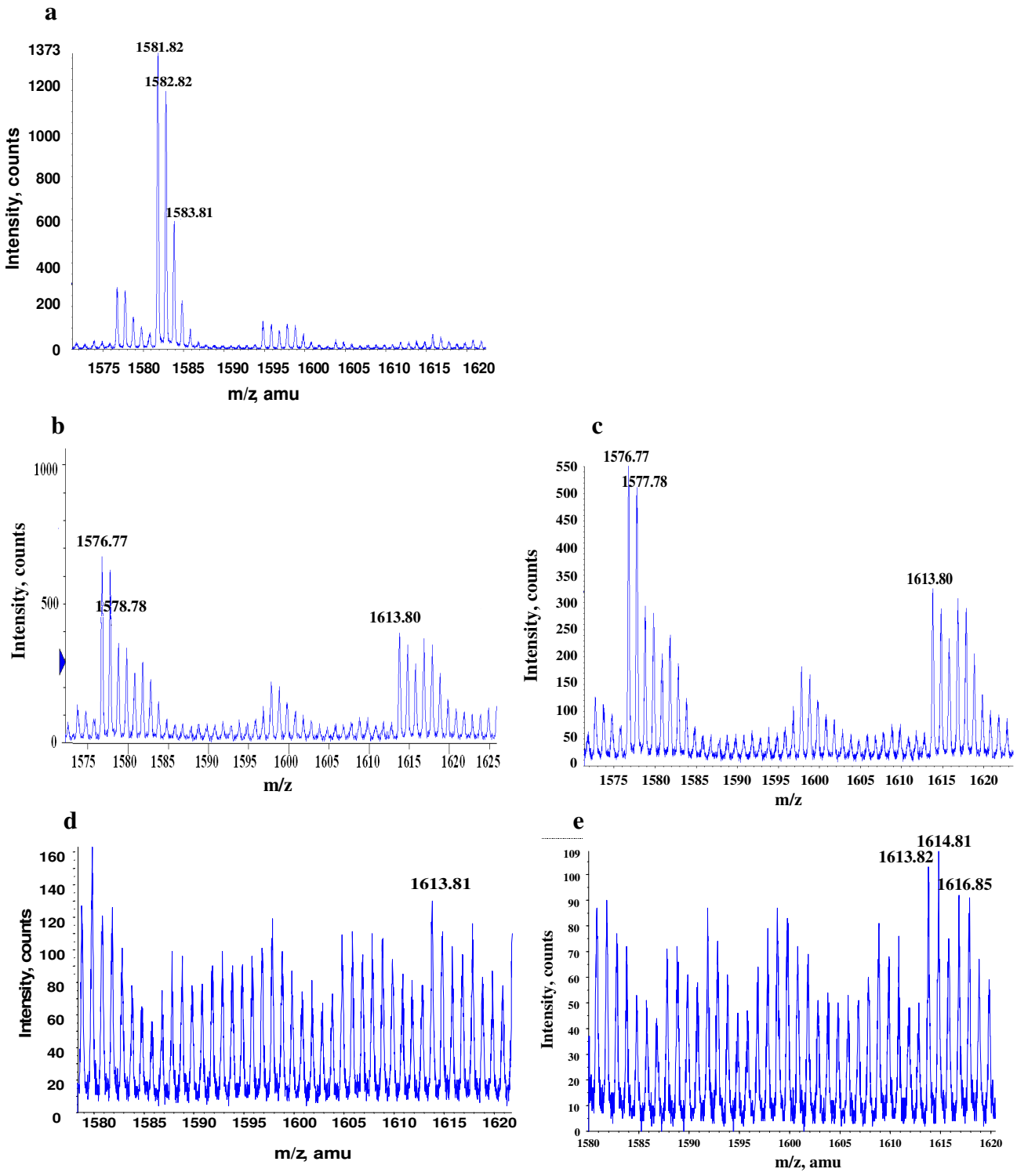


Figure S4. Segments of the MALDI Q-TOF mass spectra of pepsin digests of (a) native human NAT2; (b) N-OH-4-AABP-inactivated NAT2; (c) 4-nitrosobiphenyl (4-NO-BP)-inactivated NAT2; (d) N-OH-AAF-inactivated NAT2; and (e) 2-nitrosofluorene (2-NO-F)-inactivated NAT2.

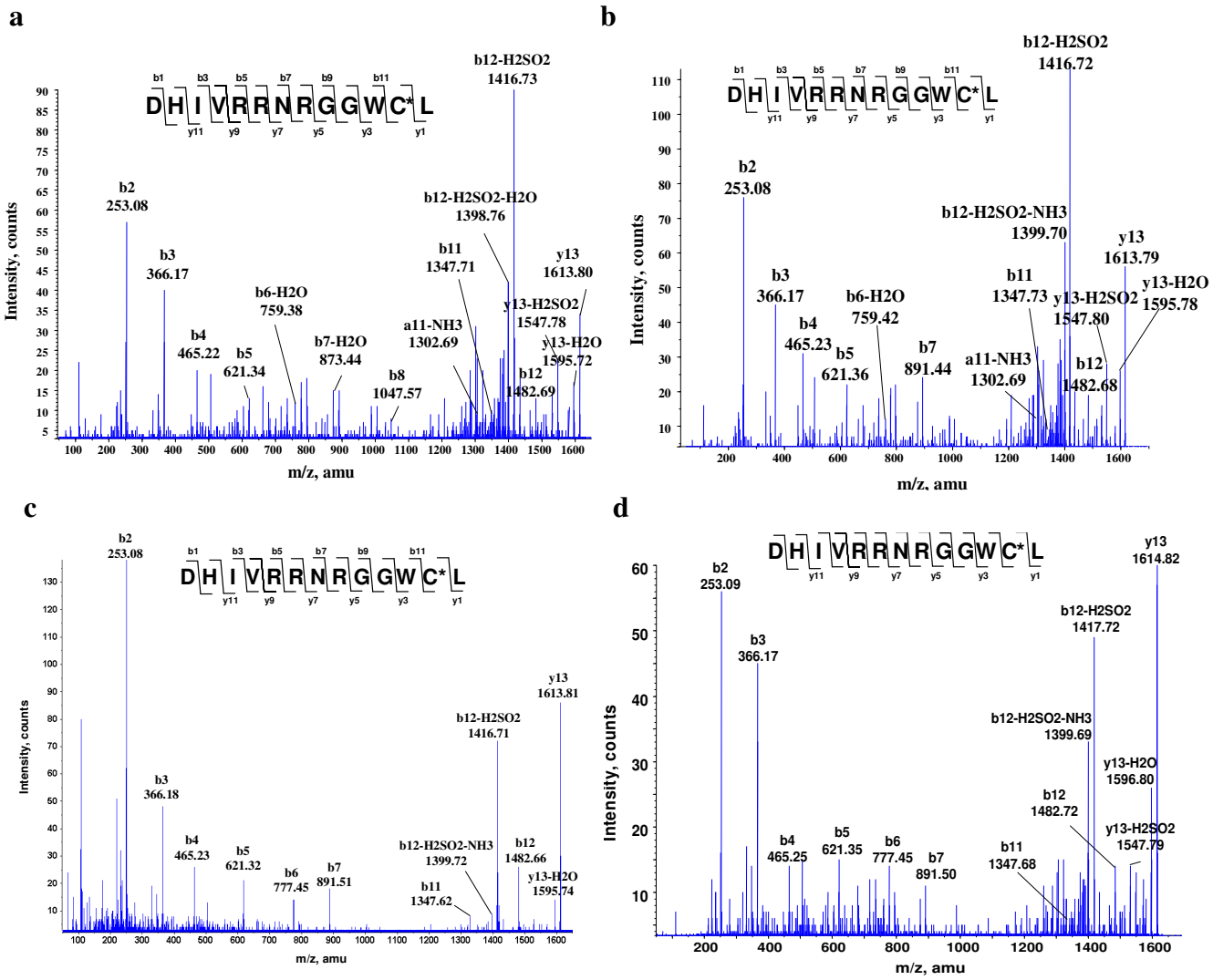


Figure S5. MALDI Q-TOF tandem mass spectra: (a) 1613.80 Da peptide obtained by pepsin digestion of N-OH-4-AABP-inactivated NAT2, (b) 1613.80 Da peptide obtained by pepsin digestion of 4-NO-BP-inactivated NAT2, (c) 1613.81 Da peptide obtained by pepsin digestion of N-OH-AAF-inactivated NAT2, (d) 1613.81 Da peptide obtained by pepsin digestion of 2-NO-F-inactivated NAT2.

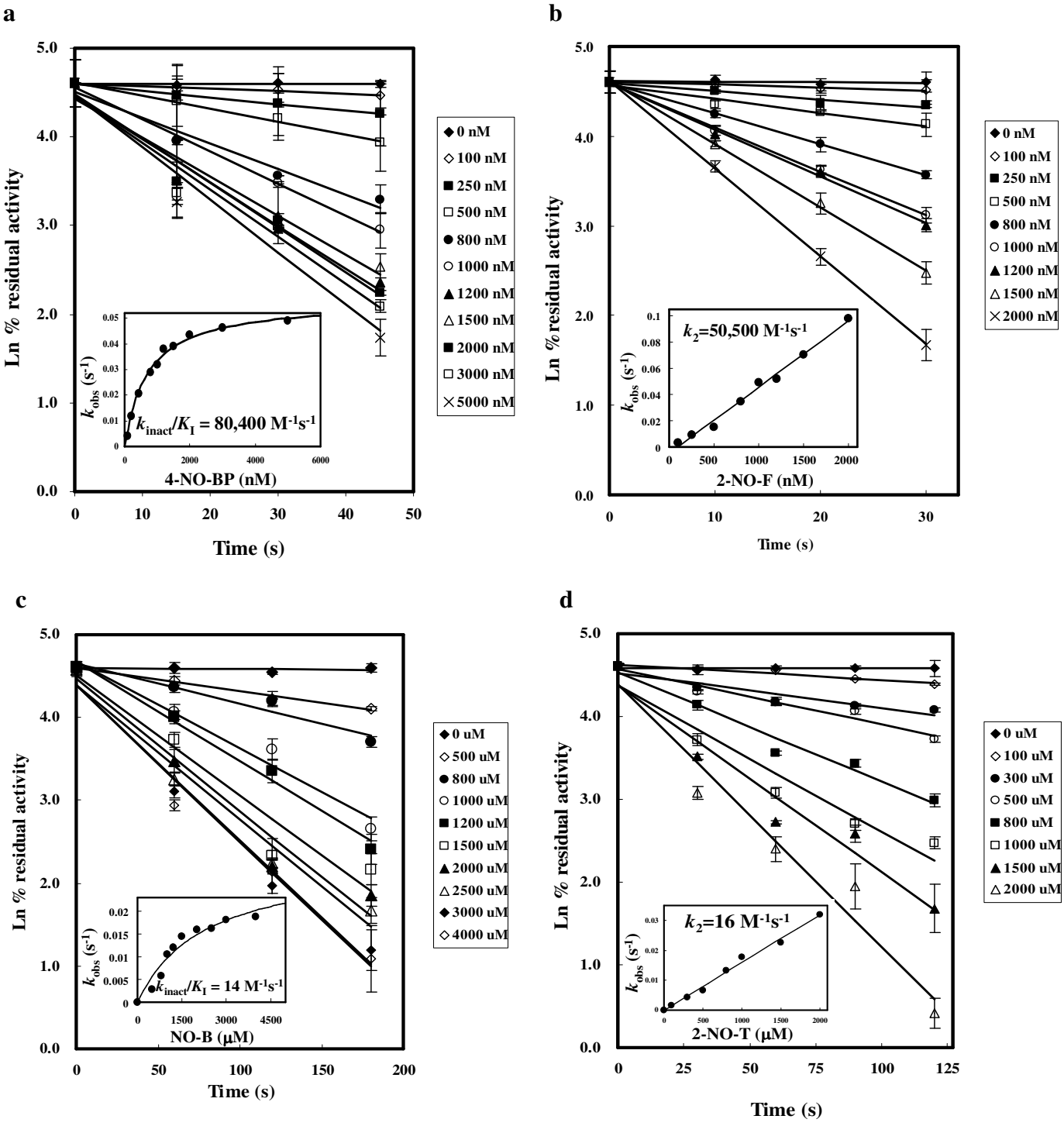


Figure S6. Time- and concentration-dependent inactivation of human NAT2 by (a) 4-NO-BP, (b) 2-NO-F, (c) nitrosobenzene (NO-B), (d) 2-nitrosotoluene (2-NO-T).

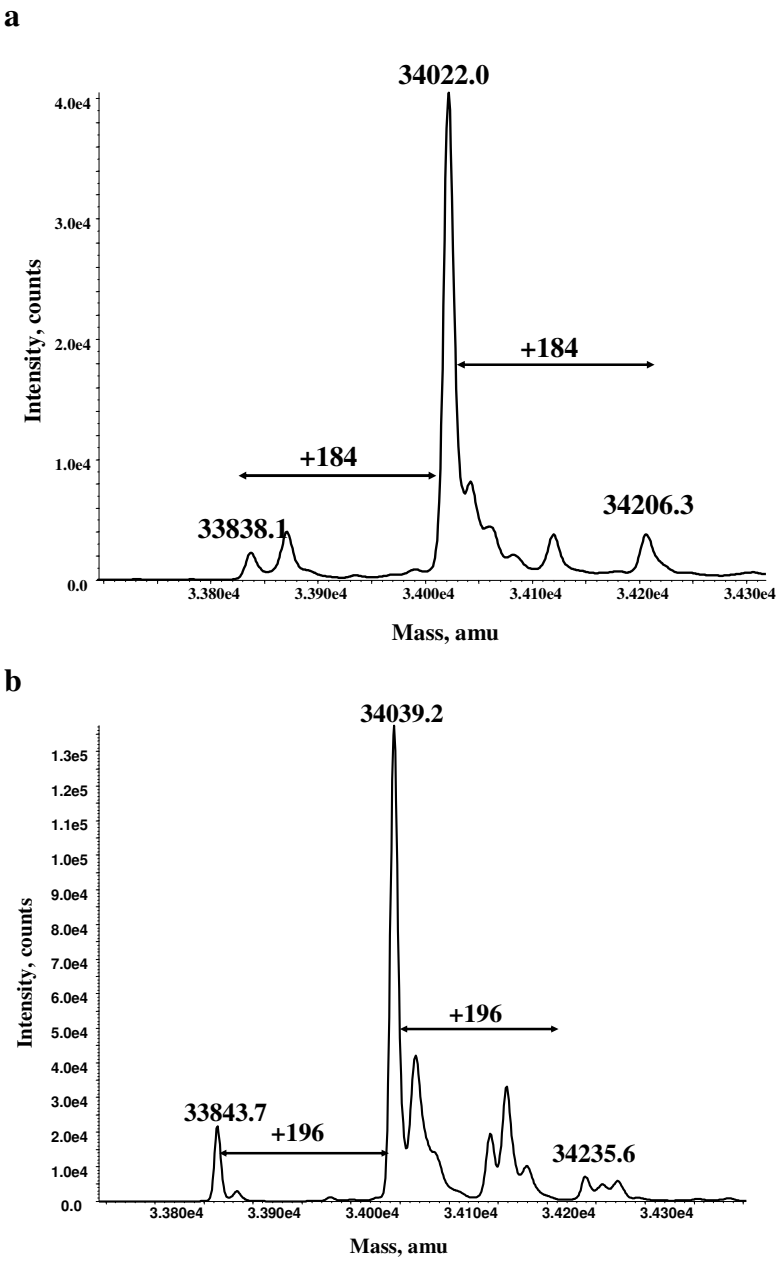


Figure S7. Deconvoluted Nano-ESI Q-TOF mass spectra: (a) 4-NO-BP-inactivated NAT2, (b) 2-NO-F-inactivated NAT2. The theoretical mass of recombinant human NAT2 is 33841.8 Da.