## Supplementary Information

Chemical detoxification of small molecules by *C. elegans*

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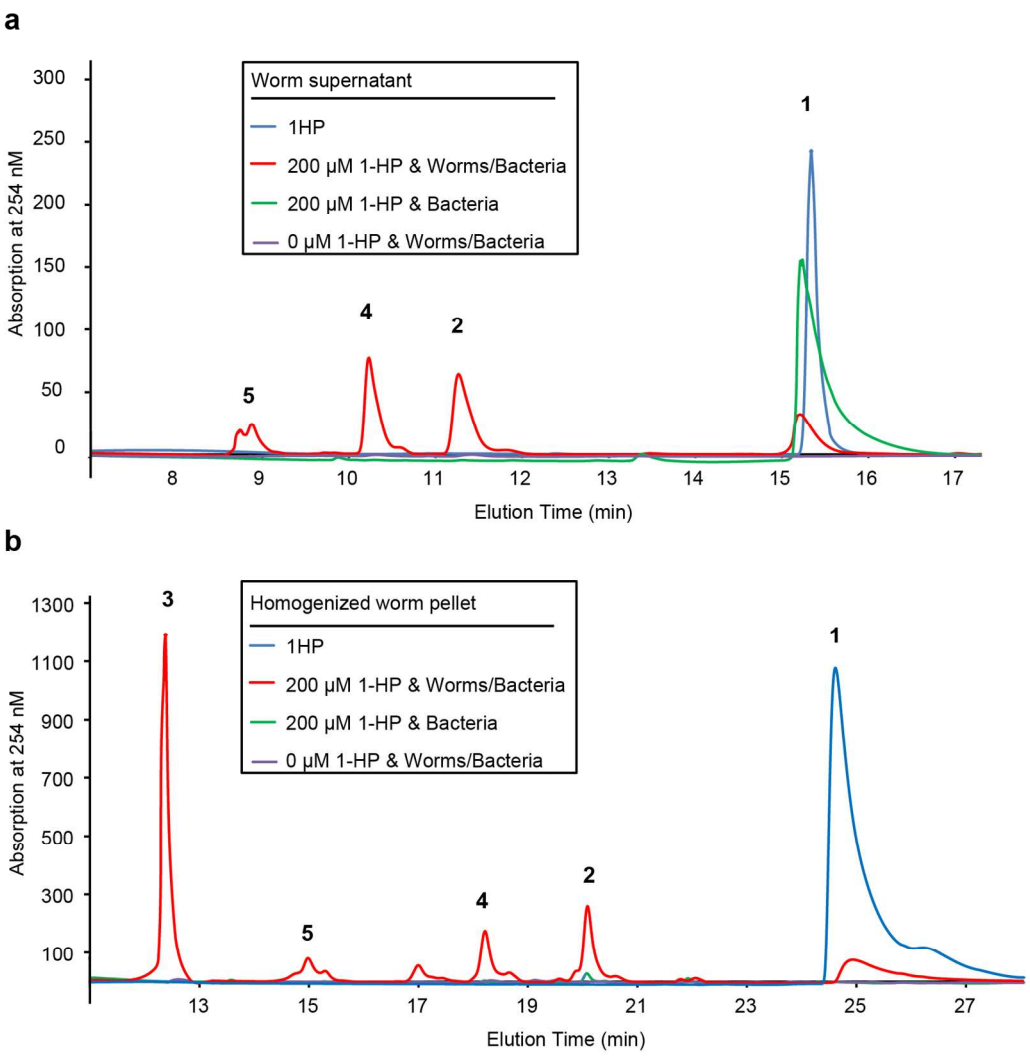
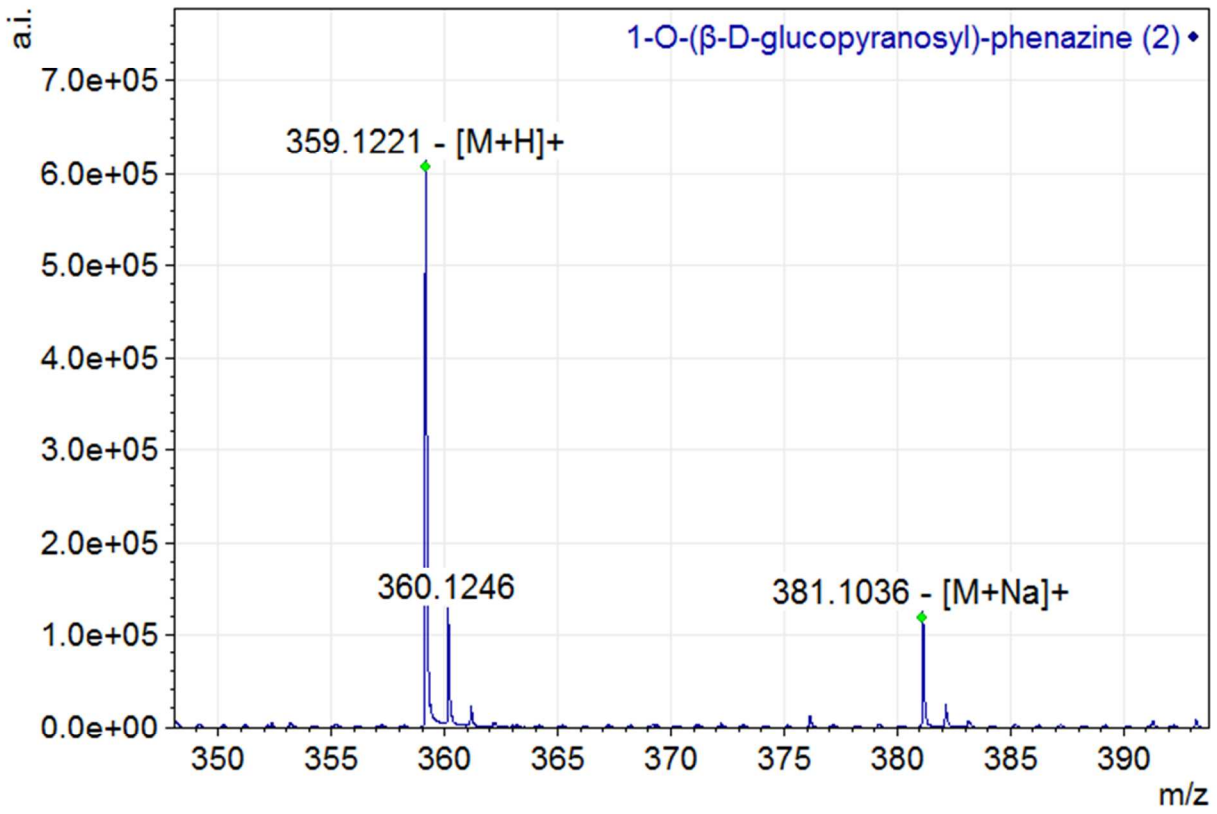
**Supplementary Table 1.** High-resolution MS data for 1-HP and indole-derived *C. elegans* metabolites acquired using negative-ion and positive-ion electrospray ionization (ESI).

| Compound | Ion | Ion formula | Calculated *m/z* | Observed *m/z* |
|---|---|---|---|---|
| **2** | $[M+H]^+$ | $C_{18}H_{19}N_2O_6^+$ | 359.1238 | 359.1221 |
| **3** | $[M-H]^-$ | $C_{18}H_{18}N_2O_9P^-$ | 437.0755 | 437.0755 |
| **4** | $[M+H]^+$ | $C_{24}H_{29}N_2O_{11}^+$ | 521.1766 | 521.1757 |
| **5a-b** | $[M+H]^+$ | $C_{30}H_{39}N_2O_{16}^+$ | 683.2294 | 683.2222 |
| **7** | $[M-H]^-$ | $C_{14}H_{16}NO_5^-$ | 278.1034 | 278.1071 |
| **8** | $[M-H]^-$ | $C_{14}H_{17}NO_8P^-$ | 358.0697 | 358.0701 |

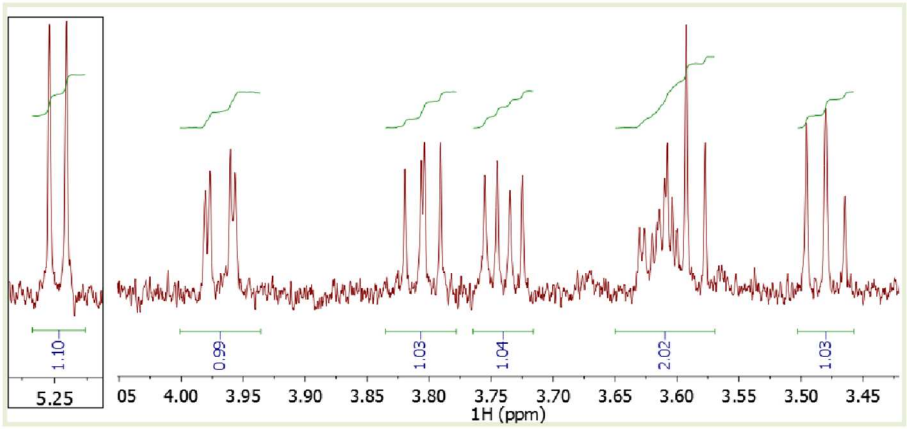



**Supplementary Figure 1.** HPLC-UV-Chromatograms of *C. elegans* supernatants and homogenized worm pellets after treatment with 1-HP. (a) UV Chromatogram of 1-HP (dark blue), worm media from 1 million wild type (N2) young adults exposed to 0 µM 1-HP (purple) or 200 µM 1-HP (red) for 24 hours, and 1% *E. coli* HB101 exposed to 200 µM 1-HP for 24 hours (green). (b) UV chromatogram of 1-HP (blue), the homogenates of 1 million wild type (N2) young adults exposed to 0 µM 1-HP (purple) or 200 µM 1-HP (red) for 24 hours, and 1% *E. coli* HB101 exposed to 200 µM 1-HP for 24 hours (green). The 1-HP curves should be interpreted for elution time only and may not indicate relative amounts of the compound. In both (a) and (b) peaks are labeled to indicate compounds as in Figure 1.

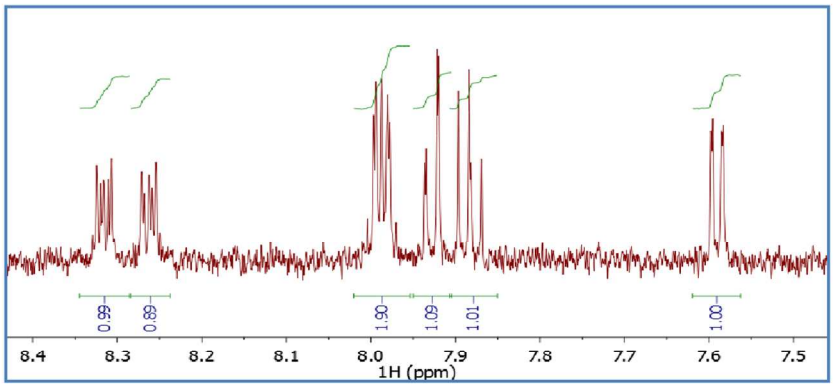



**Supplementary Figure 2.1.** Mass spectrum of purified 1-*O*-( $\beta$ -D-glucopyranosyl)-phenazine (2) from the supernatants of 1-HP exposed young-adult worms in ESI+ mode with annotation of the $[M+H]^+$ peak at *m/z* 359 and the $[M+Na]^+$ peak at *m/z* 381.

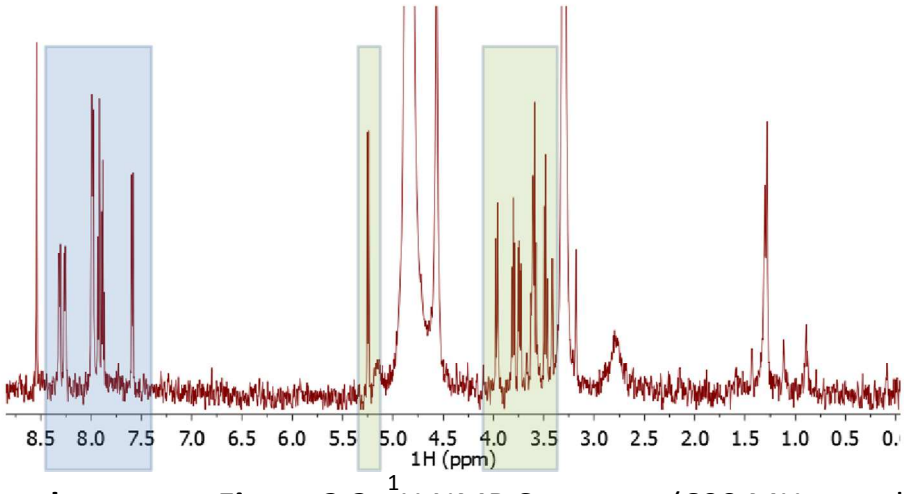
**a**



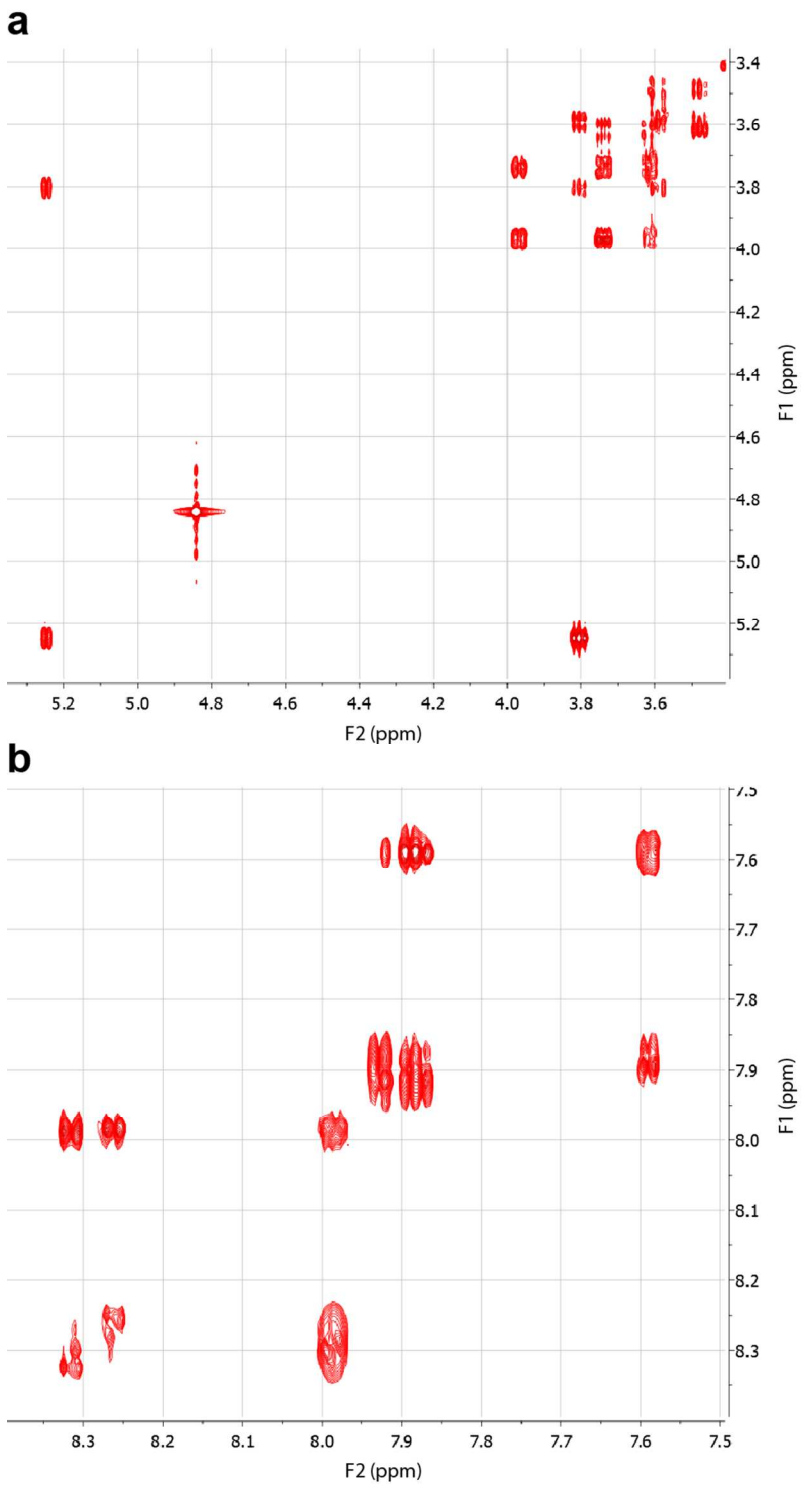

**b**




**c**




**Supplementary Figure 2.2.** $^{1}$H NMR Spectrum (600 MHz, methanol-$d_4$) of purified 1-*O*-(β-D-glucopyranosyl)-phenazine (2). Expansions of the $^{1}$H-NMR spectrum highlighting the carbohydrate (a) and aromatic (b) regions. (c) Full $^{1}$H-NMR spectrum with regions shown in (a) and (b) highlighted in blue and green, respectively.

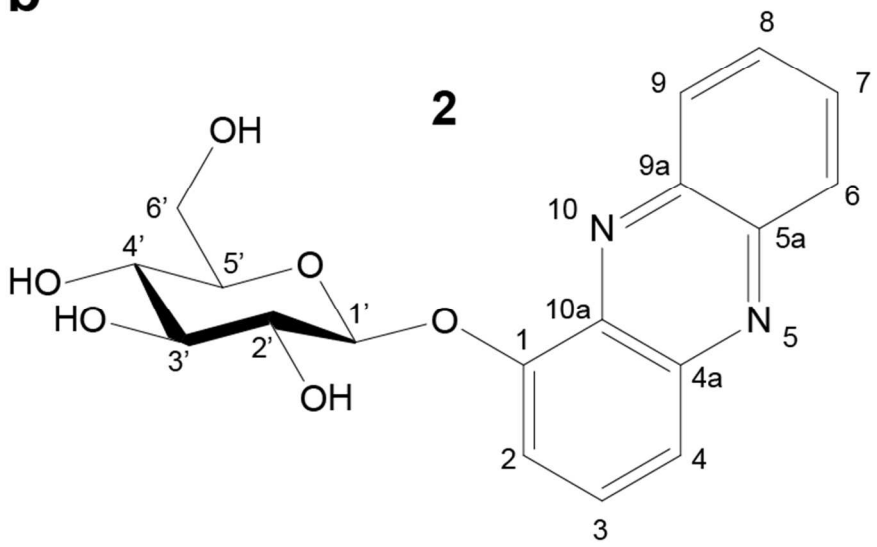



**Supplementary Figure 2.3.** gCOSY Spectrum (600 MHz, methanol-$d_4$) of purified 1-*O*-( $\beta$ -D-glucopyranosyl)-phenazine (2). Expansions showing the carbohydrate (a) and aromatic (b) regions.

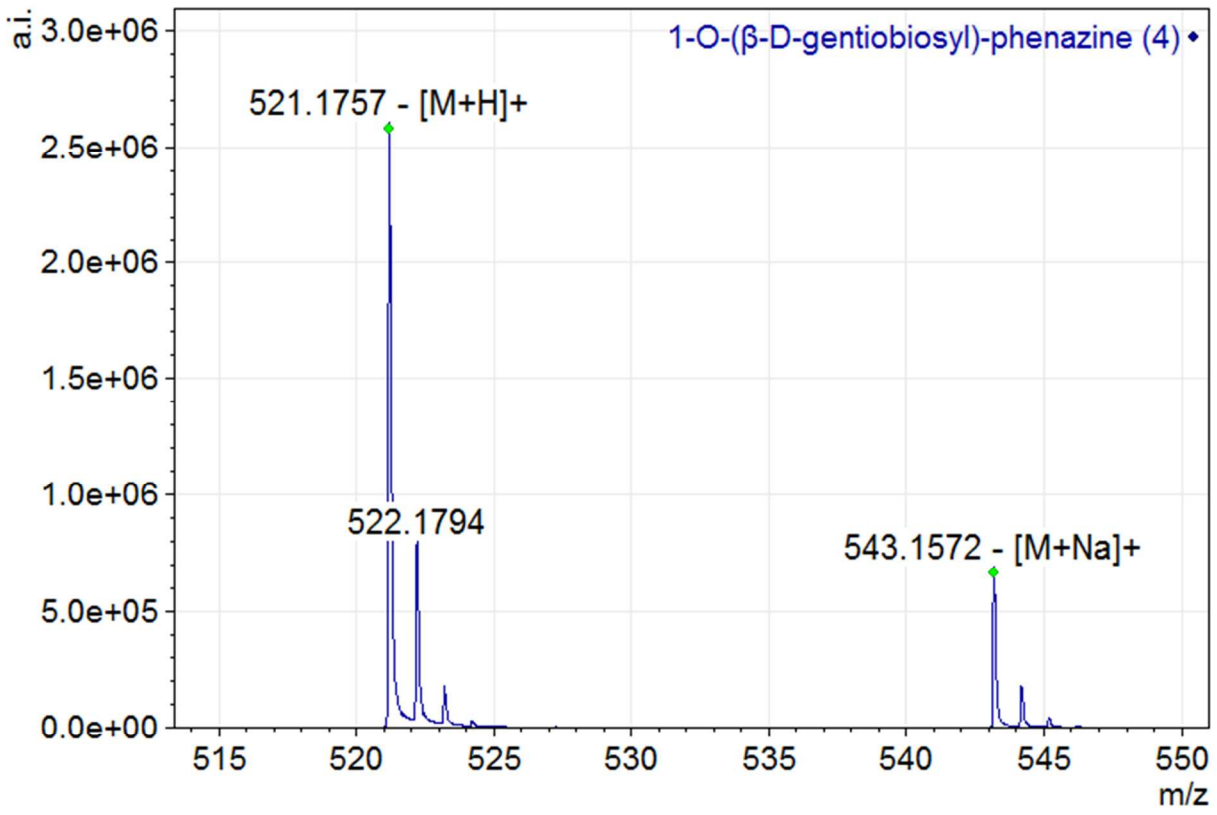
**a**

| # | Type | $\delta_H$ [ppm] | J [Hz] |
|---|---|---|---|
| 2 | dd | 7.59 | 7.5, 1.2 |
| 3 | dd | 7.88 | 8.9, 7.5 |
| 4 | dd | 7.93 | 8.8, 1.2 |
| 6 | ddd | 8.32 | |
| 7 | ddd | 7.98 | |
| 8 | ddd | 7.98 | |
| 9 | ddd | 8.26 | |
| 1’ | d | 5.25 | 7.8 |
| 2’ | dd | 3.81 | 9.4, 7.8 |
| 3’ | dd | 3.59 | 9.2, 9.2 |
| 4’ | dd | 3.48 | 9.3, 9.3 |
| 5’ | ddd | 3.62 | 9.9, 6.1, 2.4 |
| 6’a | dd | 3.74 | 12.1, 6.0 |
| 6’b | dd | 3.97 | 12.0, 2.2 |

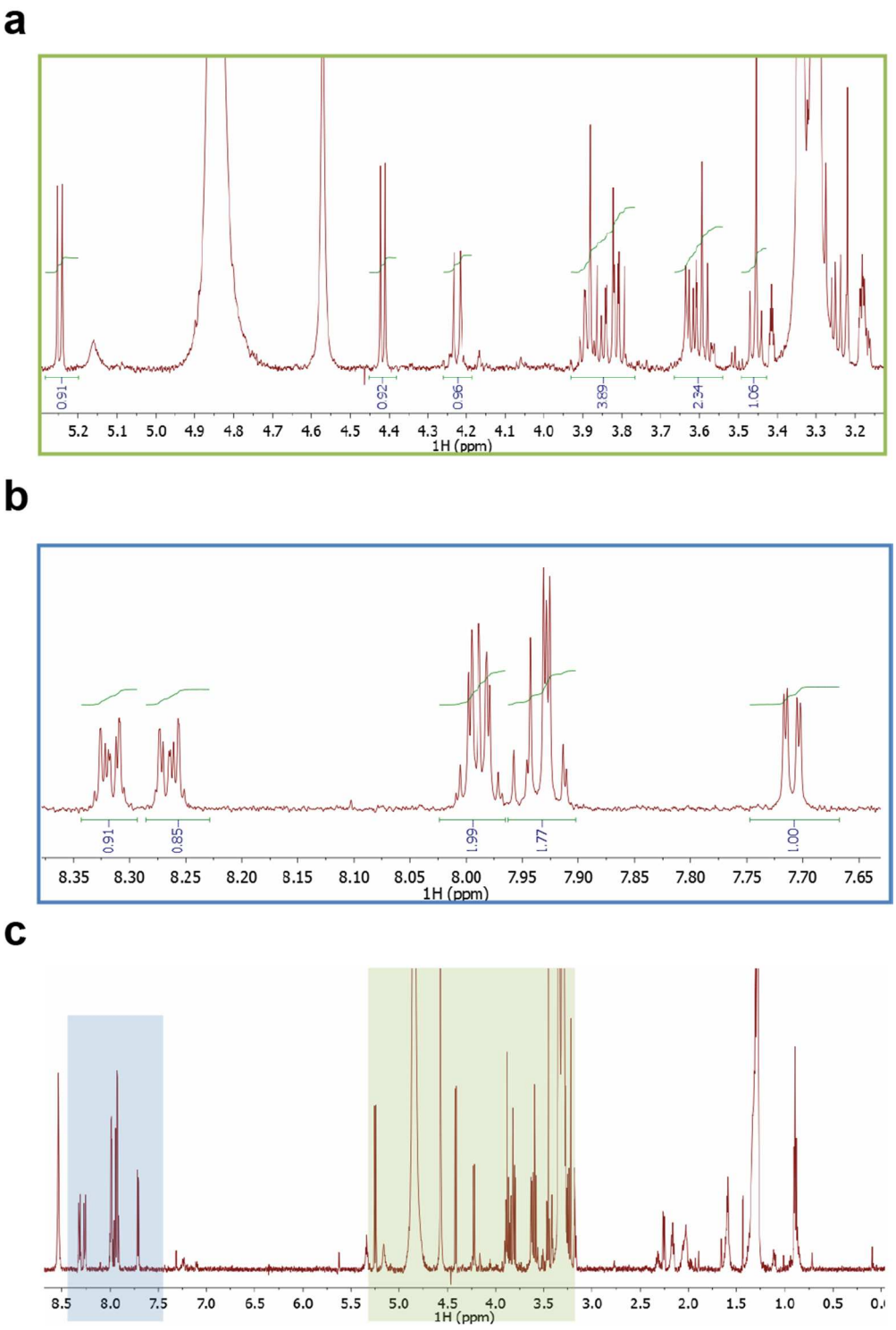
**b**



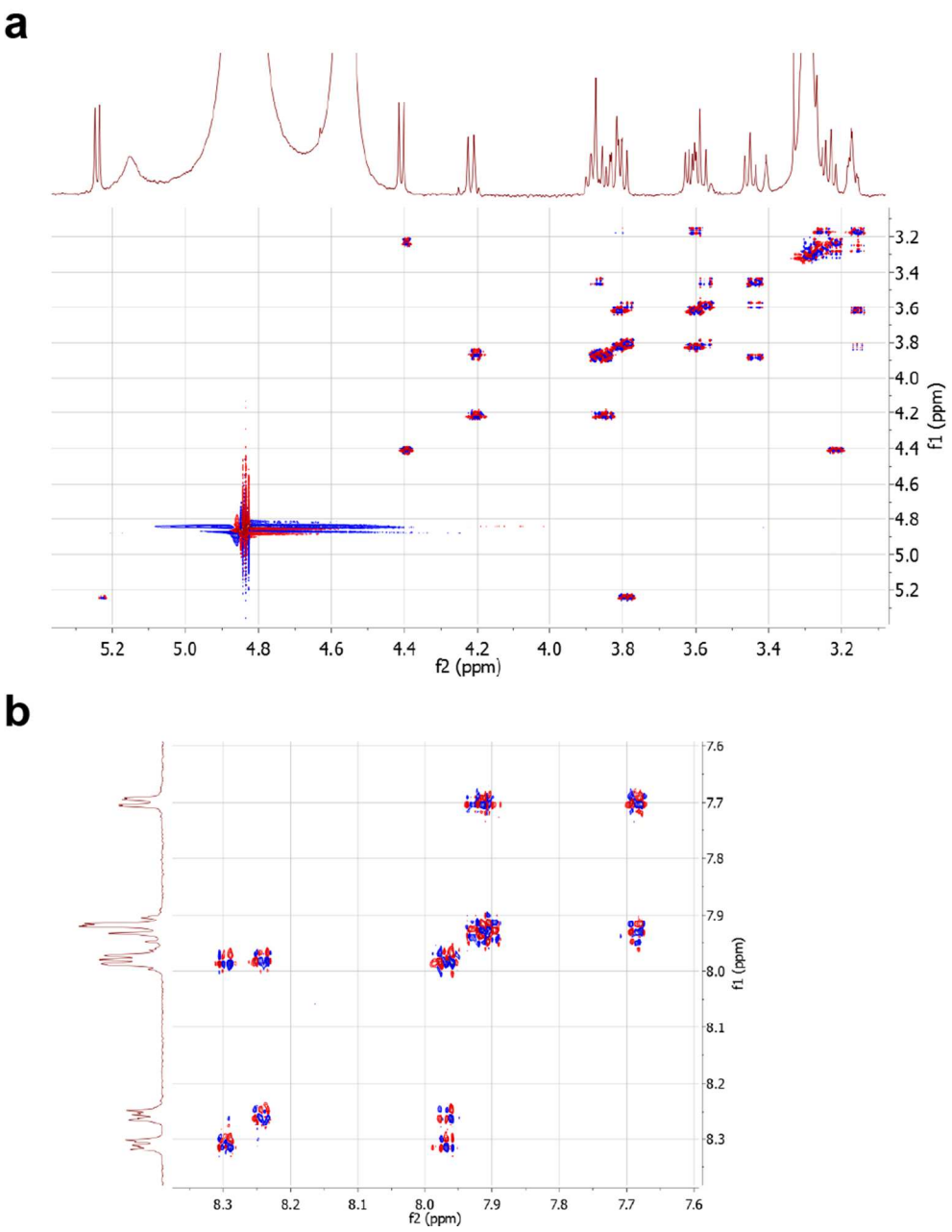

**Supplementary Figure 2.4.** NMR data and structure of purified 1-*O*-( $\beta$ -D-glucopyranosyl)-phenazine (2) in methanol-$d_4$. (a) Table showing assignments (b) Structure of 1-*O*-(β-D-glucopyranosyl)-phenazine with atom numbers.



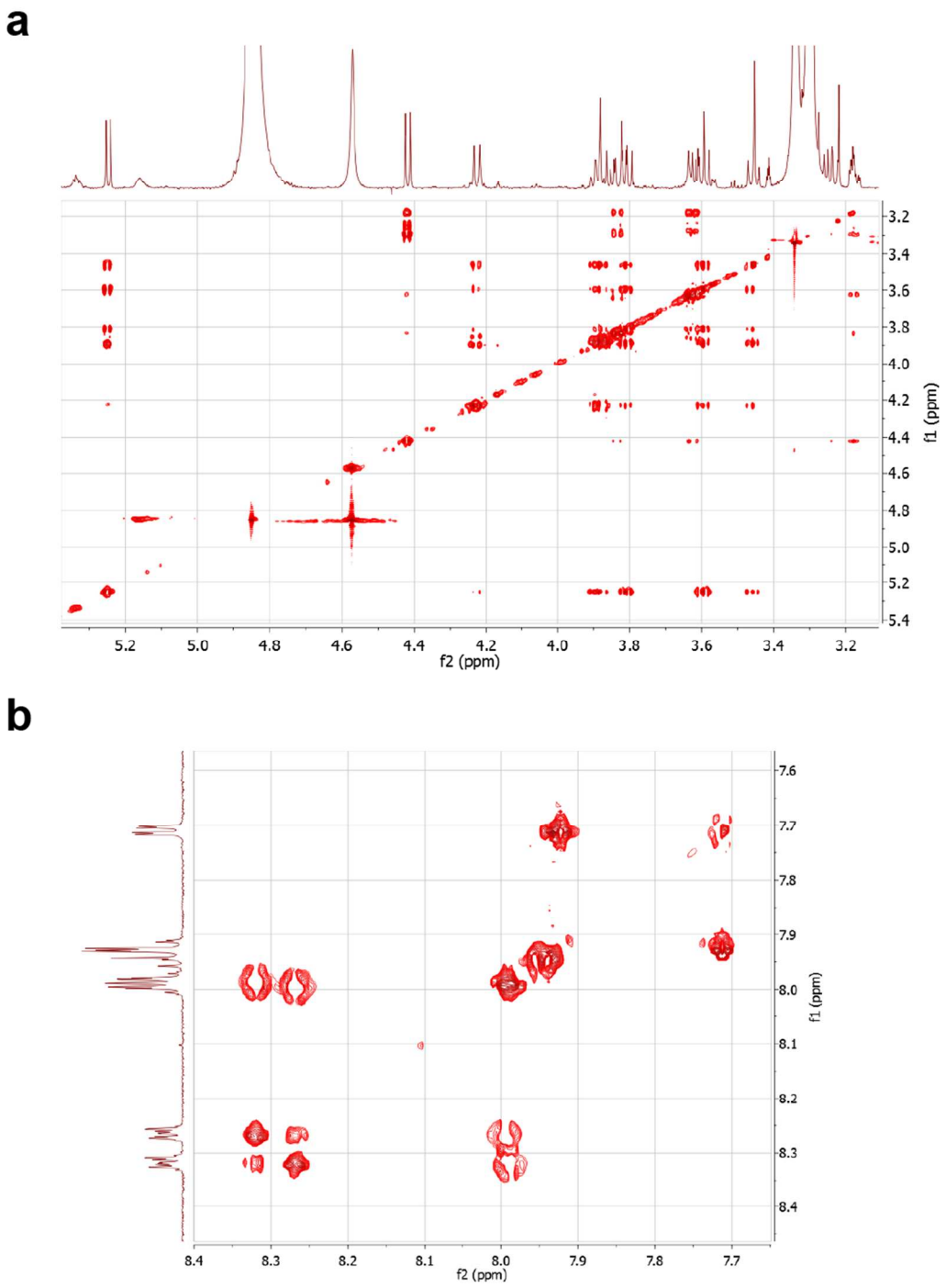

**Supplementary Figure 3.1.** Mass spectrum of purified 1-O-( $\beta$ -D-gentiobiosyl)-phenazine (4) from the supernatants of 1-HP exposed young-adult worms in ESI+ mode with annotation of the $[M+H]^+$ peak at *m/z* 521 and the $[M+Na]^+$ peak at *m/z* 543.



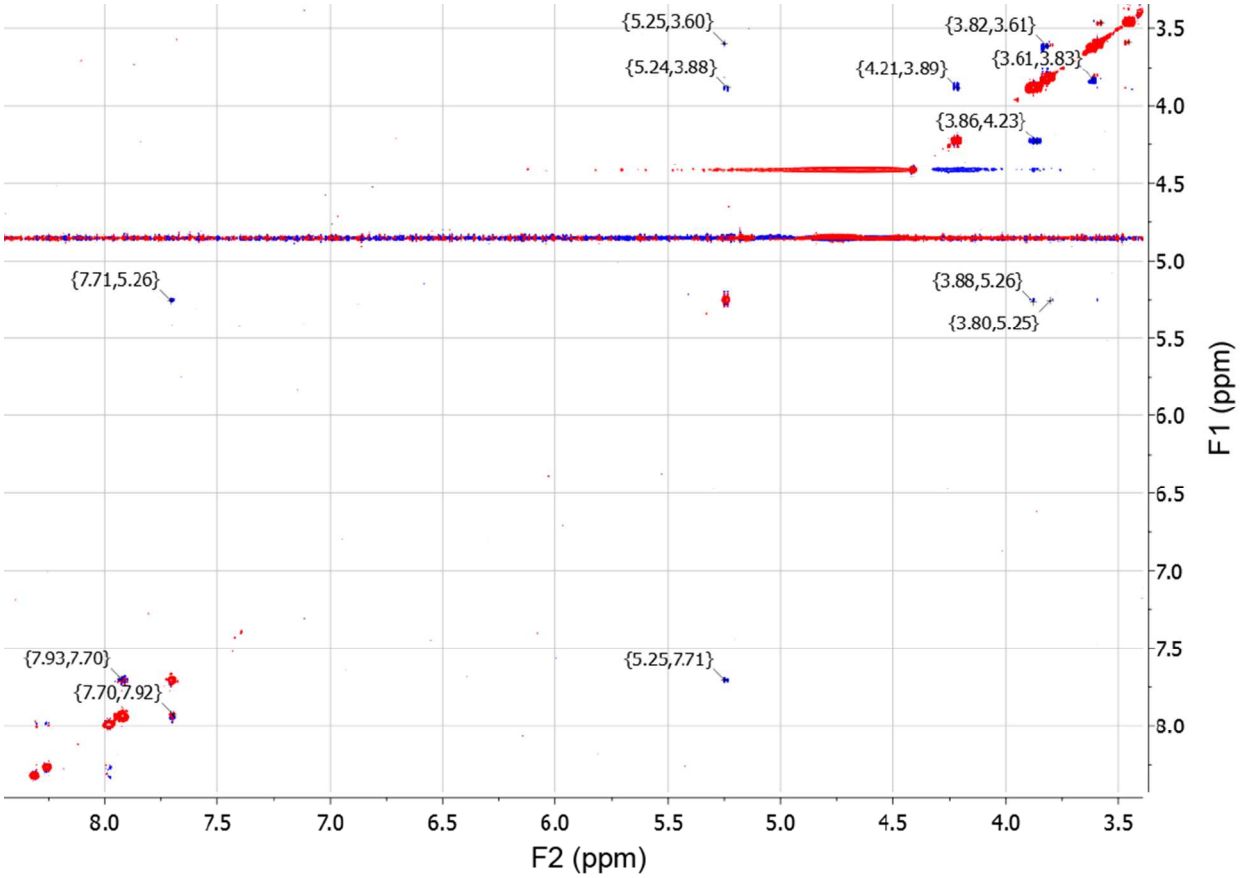

**Supplementary Figure 3.2.** $^{1}$H NMR Spectrum (600 MHz, methanol-$d_4$) of purified 1-O-( $\beta$ -D-gentiobiosyl)-phenazine (4). Expansions showing the carbohydrate (a) and aromatic (b) regions. (c) Full spectrum. Regions shown in (a) and (b) are highlighted in blue and green, respectively.



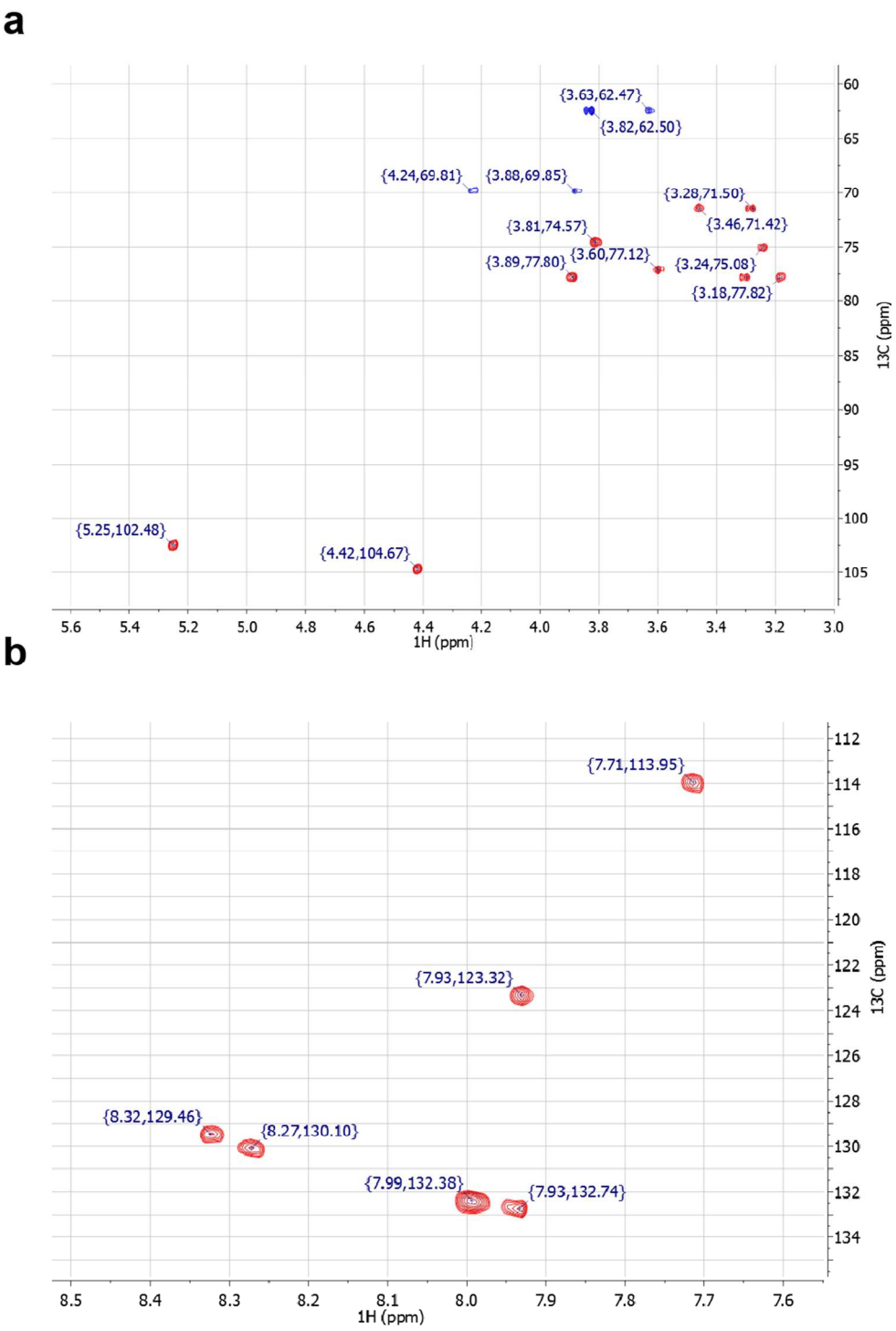

**Supplementary Figure 3.3.** dqfCOSY Spectrum (600 MHz, methanol-$d_4$) of purified 1-O-( $\beta$ -D-gentiobiosyl)-phenazine (4). Expansions showing the carbohydrate (a) and aromatic (b) regions.




**Supplementary Figure 3.4.** TOCSY Spectrum (600 MHz, methanol-$d_4$) of purified 1-O-( $\beta$ -D-gentiobiosyl)-phenazine (4). Expansions showing the carbohydrate (a) and aromatic (b) regions.




**Supplementary Figure 3.5.** NOESY Spectrum (600 MHz, methanol-$d_4$) of purified 1-O-(β-D-gentiobiosyl)-phenazine (4).

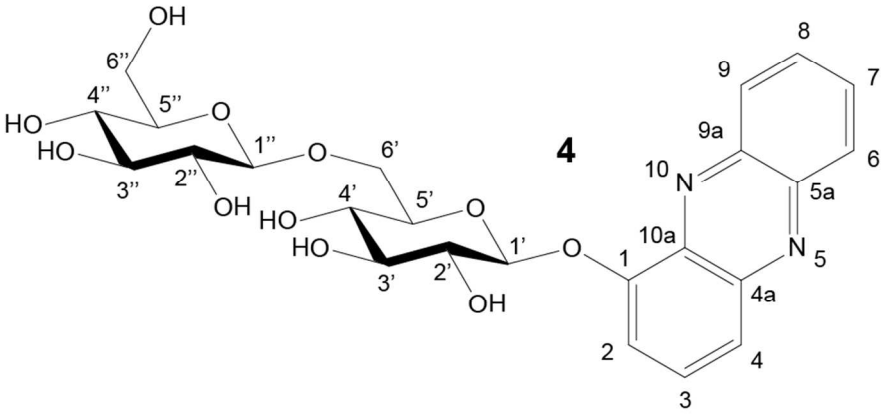



**Supplementary Figure 3.6.** HSQC Spectrum (600 MHz for $^{1}$H, 150 MHz for $^{13}$C, methanol-$d_4$) of purified 1-O-( $\beta$ -D-gentiobiosyl)-phenazine (4). Expansions showing the carbohydrate (a) and aromatic (b) regions.

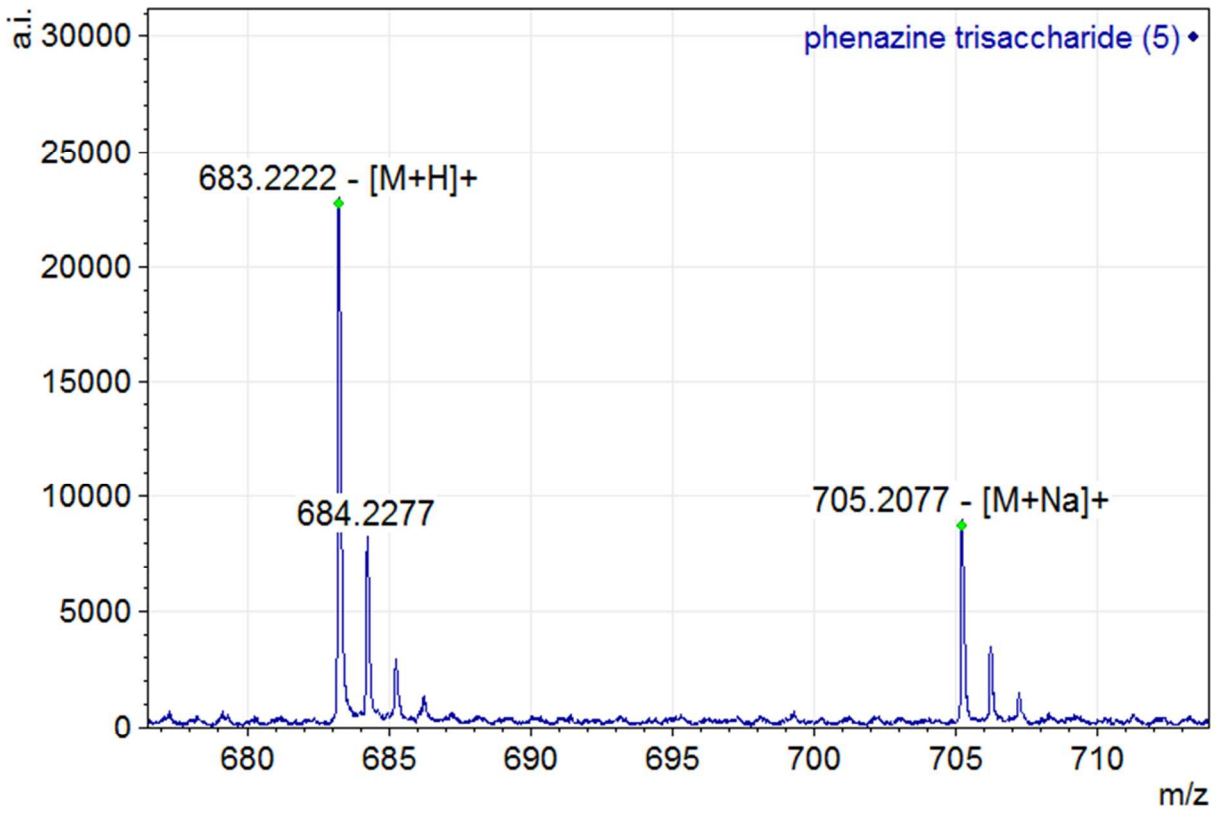
**a**

| # | Type | $\delta_H$ [ppm] | J [Hz] | $\delta_C$ | NOESY |
|---|---|---|---|---|---|
| 2 | dd | 7.7 | 6.9, 1.8 | 114 | 1' |
| 3-4 | dd | 7.93 | | 132.7, 123.3 | |
| 6 | ddd | 8.31 | | 129.5 | |
| 7-8 | ddd | 7.99 | | 132.4, 132.4 | |
| 9 | ddd | 8.26 | | 130.1 | |
| 1' | d | 5.24 | 7.8 | 102.5 | 2 |
| 2' | dd | 3.8 | 9.3, 7.7 | 74.6 | |
| 3' | dd | 3.59 | 9.2, 9.2 | 77.1 | |
| 4' | dd | 3.45 | 9.2, 9.2 | 71.4 | |
| 5' | ddd | 3.89 | | 77.8 | |
| 6a' | dd | 3.87 | | 69.9 | 6b' |
| 6b' | dd | 4.22 | 10.2 | 69.8 | 6a' |
| 1" | d | 4.41 | 7.6 | 104.7 | |
| 2" | dd | 3.24 | 8.3, 8.3 | 75.1 | |
| 3" | dd | 3.3 | | 77.8 | |
| 4" | dd | 3.27 | | 71.5 | |
| 5" | ddd | 3.17 | | 77.8 | |
| 6a" | dd | 3.61 | 11.9, 6.0 | 62.5 | |
| 6b" | dd | 3.82 | 11.6, 2.6 | 62.5 | |

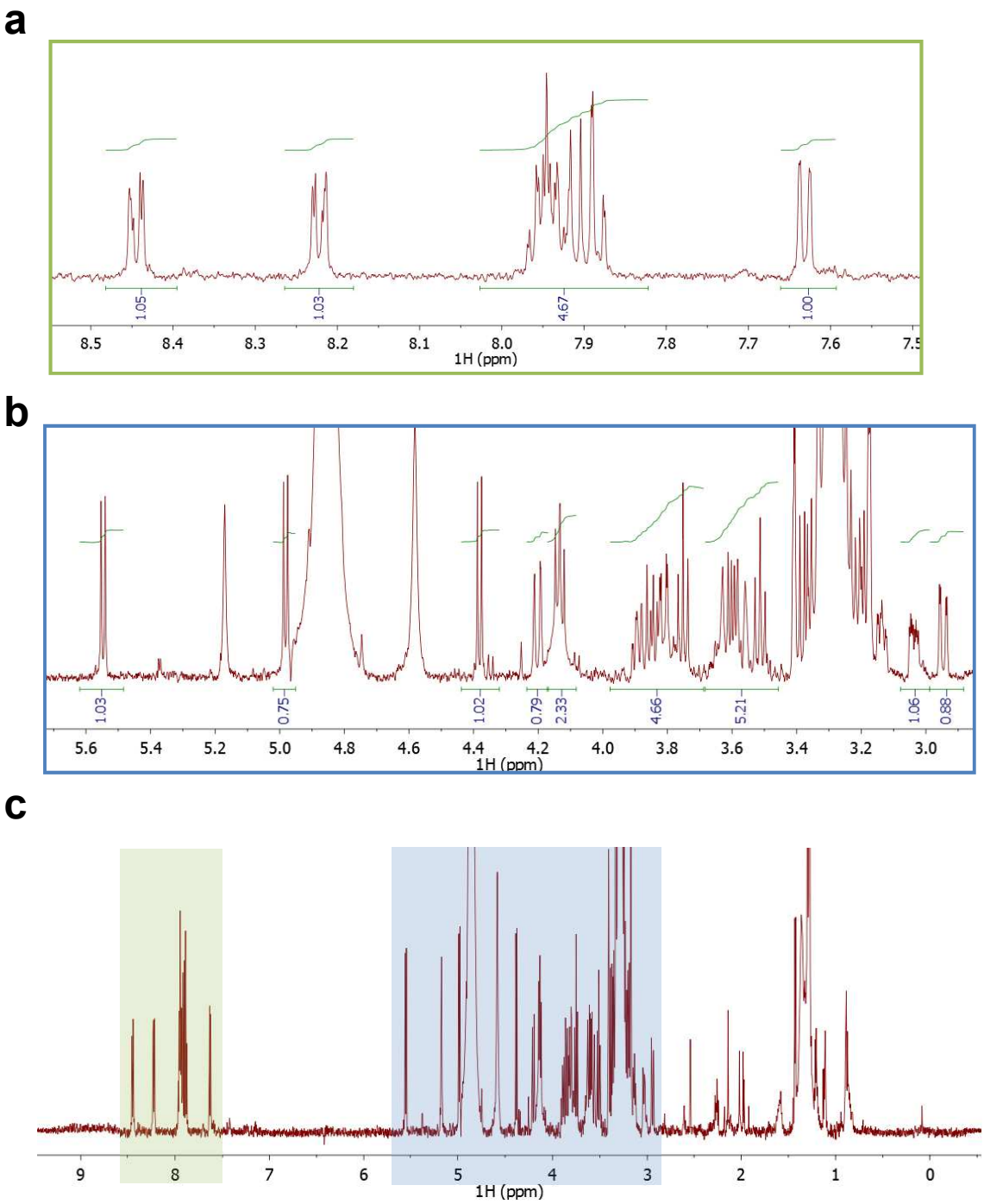
**b**



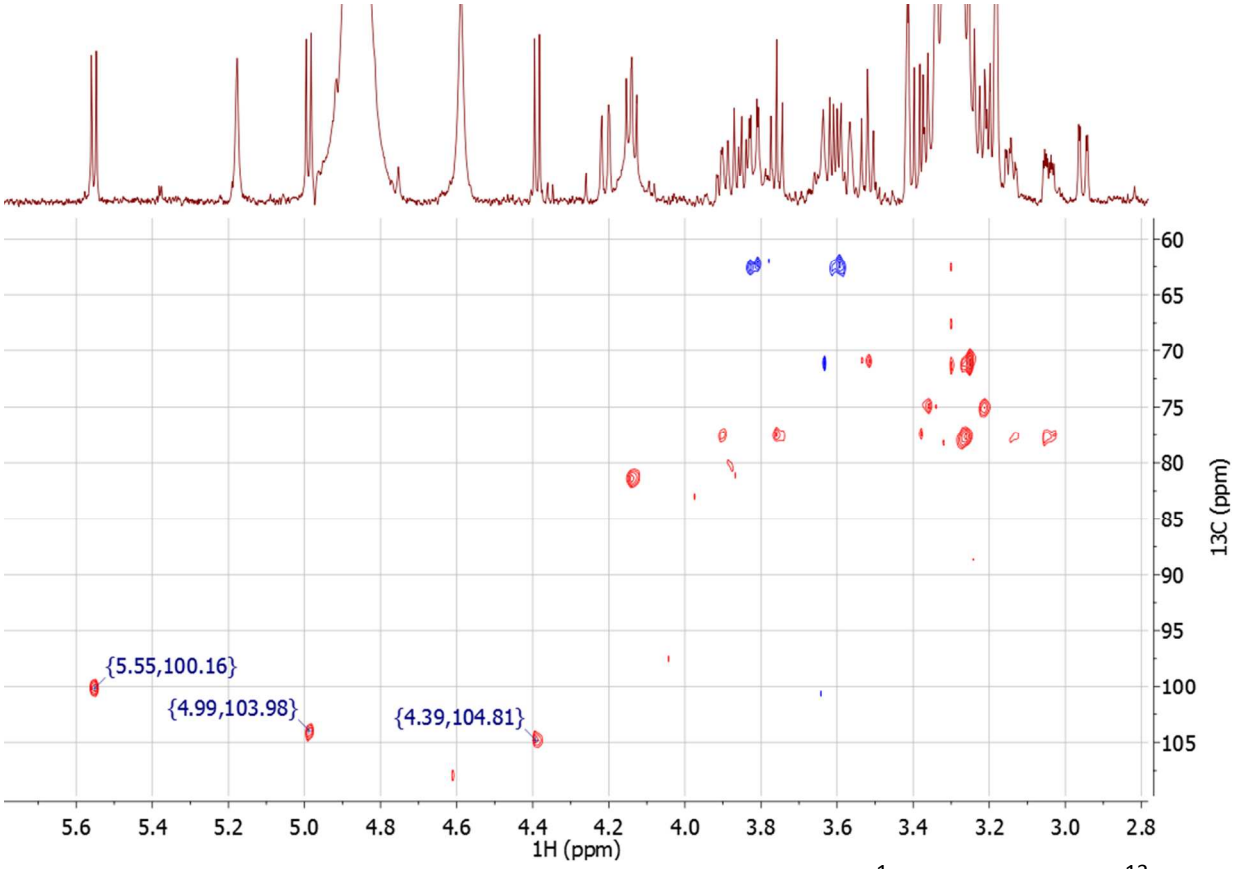

**Supplementary Figure 3.7.** NMR data of purified 1-O-( $\beta$ -D-gentiobiosyl)-phenazine (4) in methanol-$d_4$ . (a) Table showing assignments (b) Structure



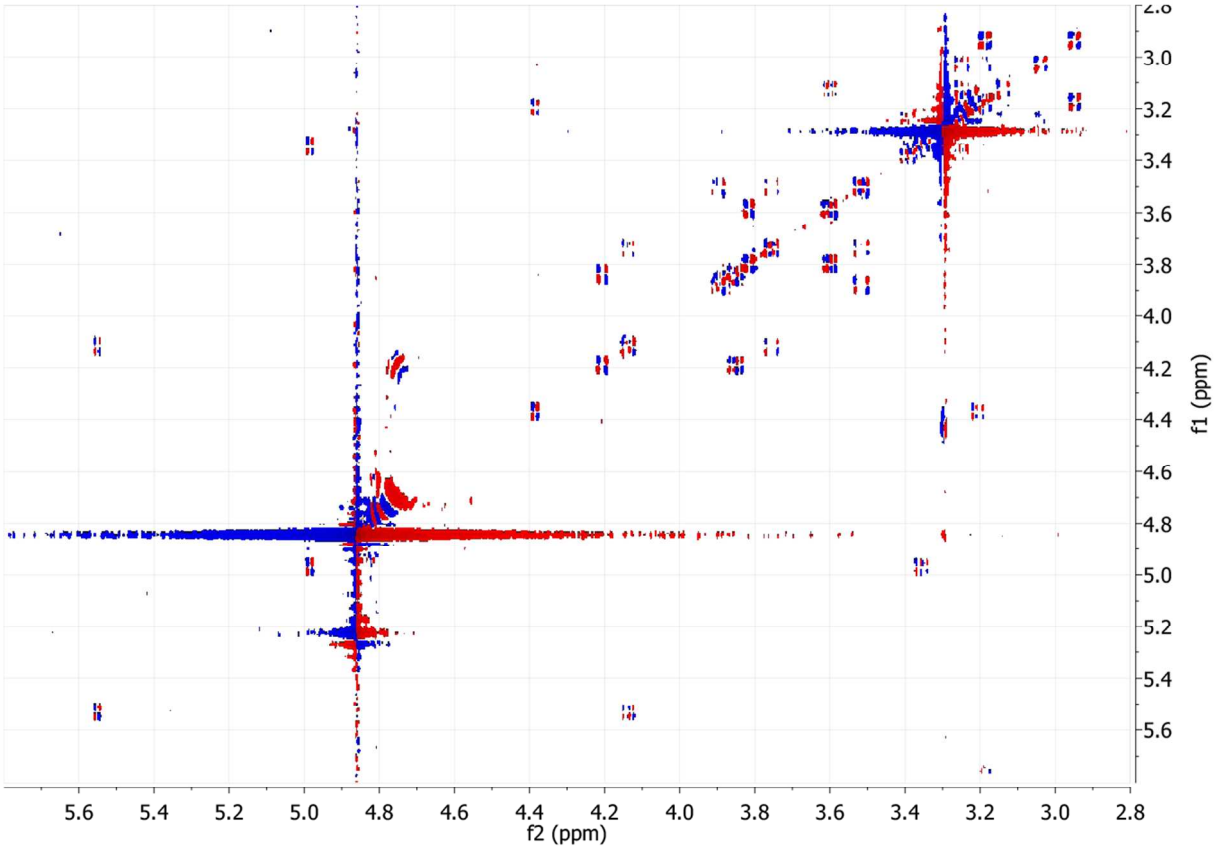

**Supplementary Figure 4.1.** Mass spectrum of purified phenazine trisaccharide (5) from the supernatants of 1-HP exposed young-adult worms in ESI+ mode with annotation of the $[M+H]^+$ peak at *m/z* 683 and the $[M+Na]^+$ peak at *m/z* 705.



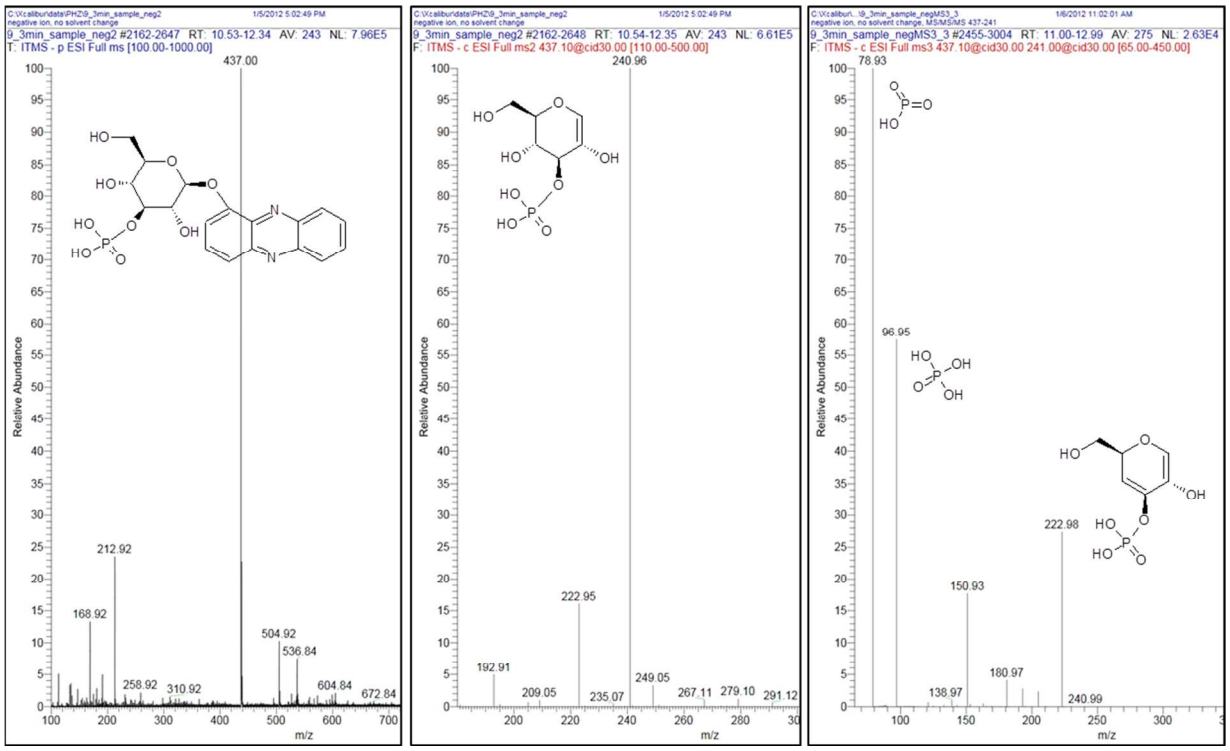

**Supplementary Figure 4.2.** $^{1}$H NMR Spectrum (600 MHz, methanol-$d_4$) of purified phenazine trisaccharide (5). Expansions showing the aromatic (a) and carbohydrate (b) regions. (c) Full spectrum. Regions shown in (a) and (b) are highlighted in blue and green, respectively.



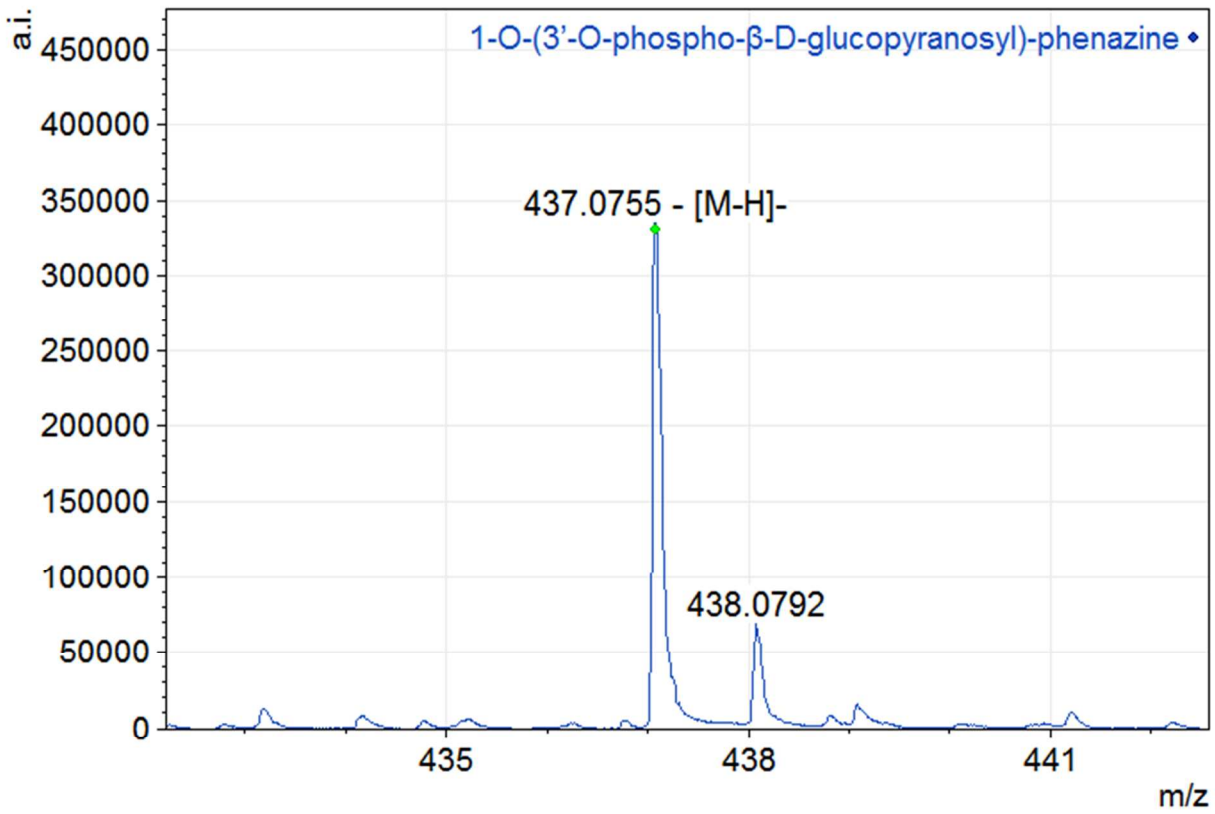

**Supplementary Figure 4.3.** HSQC Spectrum (600 MHz for $^{1}H$, 150 MHz for $^{13}C$, methanol-$d_4$) of purified phenazine trisaccharide (5). Expansion showing the carbohydrate region. The three anomeric carbons are labeled.



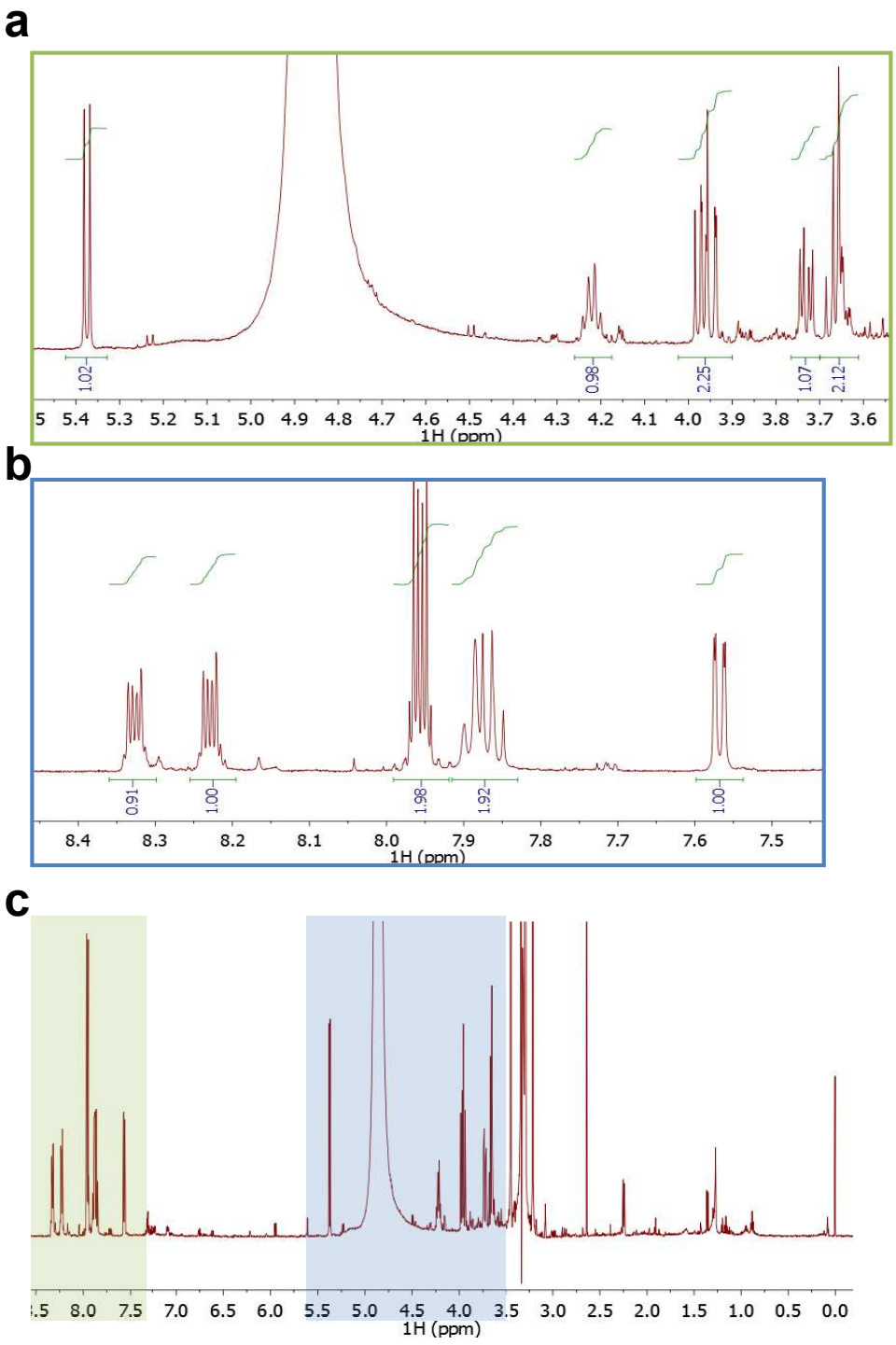

**Supplementary Figure 4.4.** dqfCOSY Spectrum (600 MHz, methanol-$d_4$) of purified phenazine trisaccharide (5). Expansion showing the carbohydrate region.



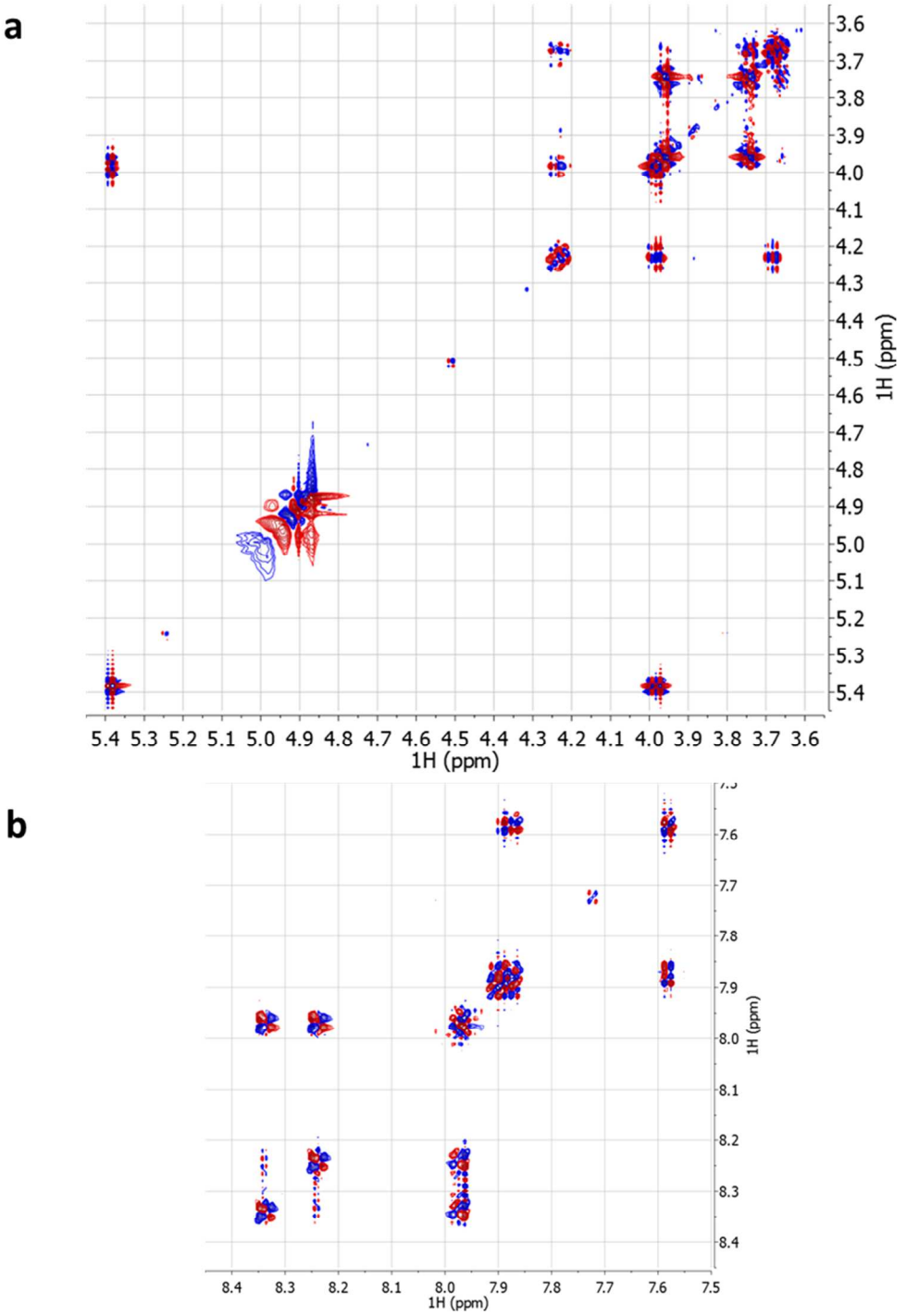

**Supplementary Figure 5.1.** Mass Chromatograms of purified 1-*O*-(3'-*O*-phospho-$\beta$-D-glucopyranosyl)-phenazine (3) from the homogenates of 1-HP exposed young-adult worms. (left) Total ion scan showing the $[M-H]^-$ peak at *m/z* 437 (center) MS-MS of *m/z* 437 showing the $[M-H]^-$ peak at *m/z* 241 (right) $MS^3$ of *m/z* 241 showing $[M-H]^-$ peaks at *m/z* 223, 96, and 79



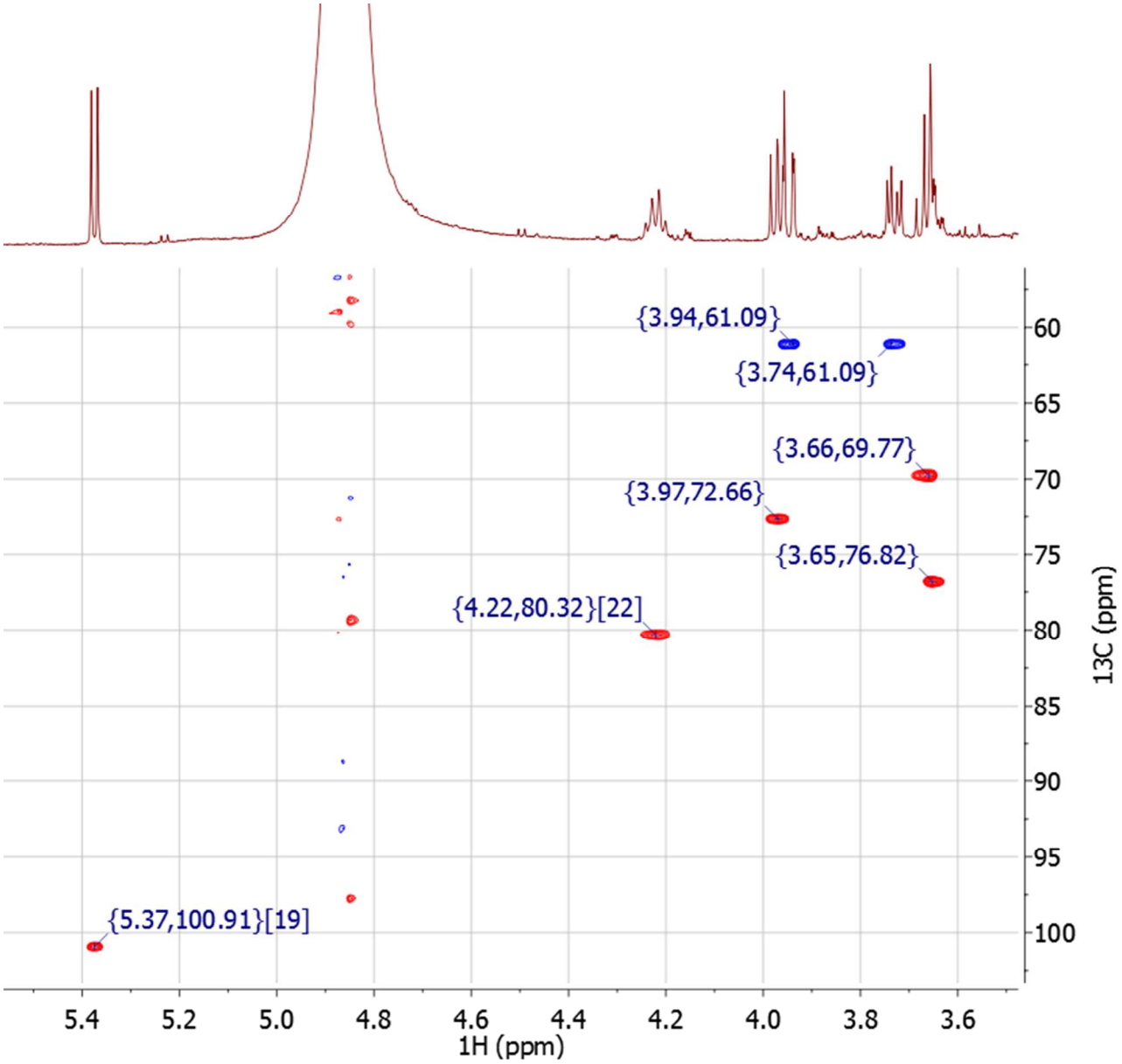

**Supplementary Figure 5.2.** Mass spectrum of purified 1-*O*-(3'-*O*-phospho- $\beta$ -D-glucopyranosyl)-phenazine (3) from the homogenates of 1-HP exposed young-adult worms in ESI- mode with annotation of the $[M-H]^-$ peak at *m/z* 437.




**Supplementary Figure 5.3.** $^{1}$H NMR Spectrum (600 MHz, methanol-$d_4$) of purified 1-*O*-(3'-*O*-phospho-β-D-glucopyranosyl)-phenazine (3). Expansions showing the carbohydrate (a) and aromatic regions (b). (c) Full spectrum. Regions shown in (a) and (b) are highlighted in green and blue, respectively.




**Supplementary Figure 5.4.** dqfCOSY Spectrum (600 MHz, methanol-$d_4$) of purified 1-*O*-(3'-*O*-phospho- $\beta$ -D-glucopyranosyl)-phenazine (3). (a) Expansions showing the carbohydrate and (b) aromatic regions.

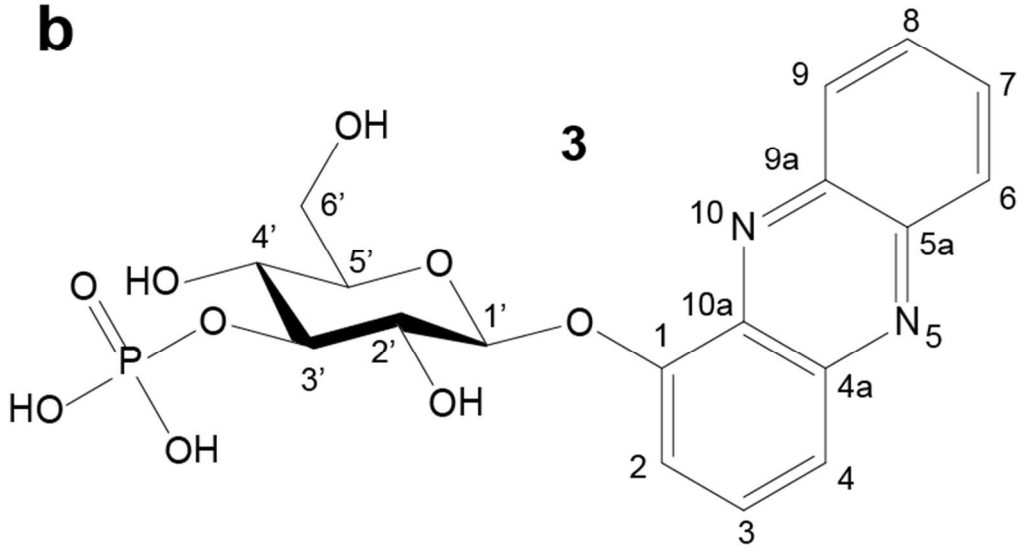



**Supplementary Figure 5.5.** HSQC Spectrum (600 MHz for $^{1}$H, 150 MHz for $^{13}$C, methanol-$d_4$) of purified 1-*O*-(3'-*O*-phospho- $\beta$ -D-glucopyranosyl)-phenazine (3). Expansion showing the carbohydrate region.

**a**

| # | Type | $\delta_H$ [ppm] | J [Hz] | $\delta_C$ |
|---|---|---|---|---|
| 1' | d | 5.37 | 7.8 | 100.9 |
| 2' | dd | 3.97 | 9.0, 7.8 | 72.7 |
| 3' | dd | 4.24 | | 80.3 |
| 4' | dd | 3.68 | 9.7, 8.0 | 69.8 |
| 5' | ddd | 3.65 | | 76.8 |
| 6a' | dd | 3.94 | 12, 1.7 | 61.1 |
| 6b' | dd | 3.74 | 12, 5 | 61.1 |
| 2 | dd | 7.57 | 7.3, 1.5 | 112.1 |
| 3 | dd | 7.89 | | |
| 4 | dd | 7.89 | | |
| 6 | ddd | | 6.7, 3.4 | |
| 7-8 | ddd | 7.96 | 6.6, 3.2 | 131.1 |
| 9 | ddd | | 6.7, 3.4 | |

**b**




**Supplementary Figure 5.6.** NMR data of purified 1-*O*-(3'-*O*-phospho- $\beta$ -D-glucopyranosyl)-phenazine (3) in methanol-$d_4$ . (a) Table showing assignments and (b) structure.

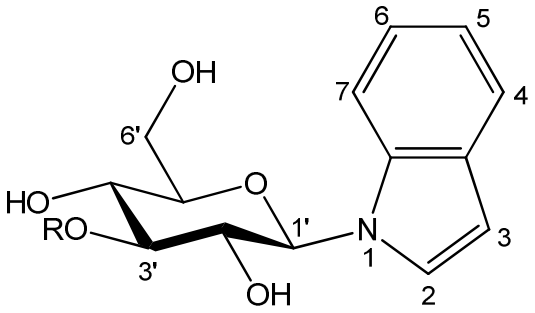
**a**

| Position | **(7)** δ $^{13}$C [ppm] | **(7)** δ $^{1}$H [ppm] | **(8)** δ $^{1}$H [ppm] | **(7)** $^{1}$H-$^{1}$H-coupling constants [Hz] | **(7)** HMBC correlations |
|---|---|---|---|---|---|
| 2 | 126.2 | 7.40 | | $J_{2',3'}$ = 3.3 | C-1 (weak), C-3', C-3a', C-7' (weak), C-7a' |
| 3 | 103.2 | 6.49 | | | C-2', C-3a', C-4' (weak), C-7a' (weak) |
| 3a | 130.3 | | | | |
| 4 | 121.3 | 7.52 | | $J_{4',5'}$ = 8.0, | C-3', C-6', C-7a' |
| 5 | 120.7 | 7.05 | | $J_{5',6'}$ = 7.4, $J_{3',5'}$ = 1.1, | C-3a', C-7' |
| 6 | 122.4 | 7.15 | | $J_{6',7'}$ = 8.0, $J_{4',6'}$ = 1.0 | C-4', C-7a' |
| 7 | 111.2 | 7.54 | | | C-3a', C-5' |
| 7a | 137.8 | | | | |
| 1' | 86.5 | 5.46 | 5.55 | J1,2 = 9.0 | C-2, C-3, C-5, C-2', C-7a' |
| 2' | 73.4 | 3.94 | 4.12 | J2,3 = 9.0, | C-1, C-3 |
| 3' | 78.9 | 3.60 | 4.24 (JH,P = 8 Hz) | J3,4 = 9.0 | C-2, C-4 |
| 4' | 71.2 | 3.50 | 3.68 | J4,5 = 9.0 | C-3, C-5, C-6 |
| 5' | 80.4 | 3.58 | | J5,6a = 5.8 | C-1, C-3, C-6 |
| 6'a | 62.5 | 3.70 | | J6a,6b = 12.1 | C-4, C-5 |
| 6'b | | 3.88 | | J5,6b = 2.2 | C-4, C-5 |

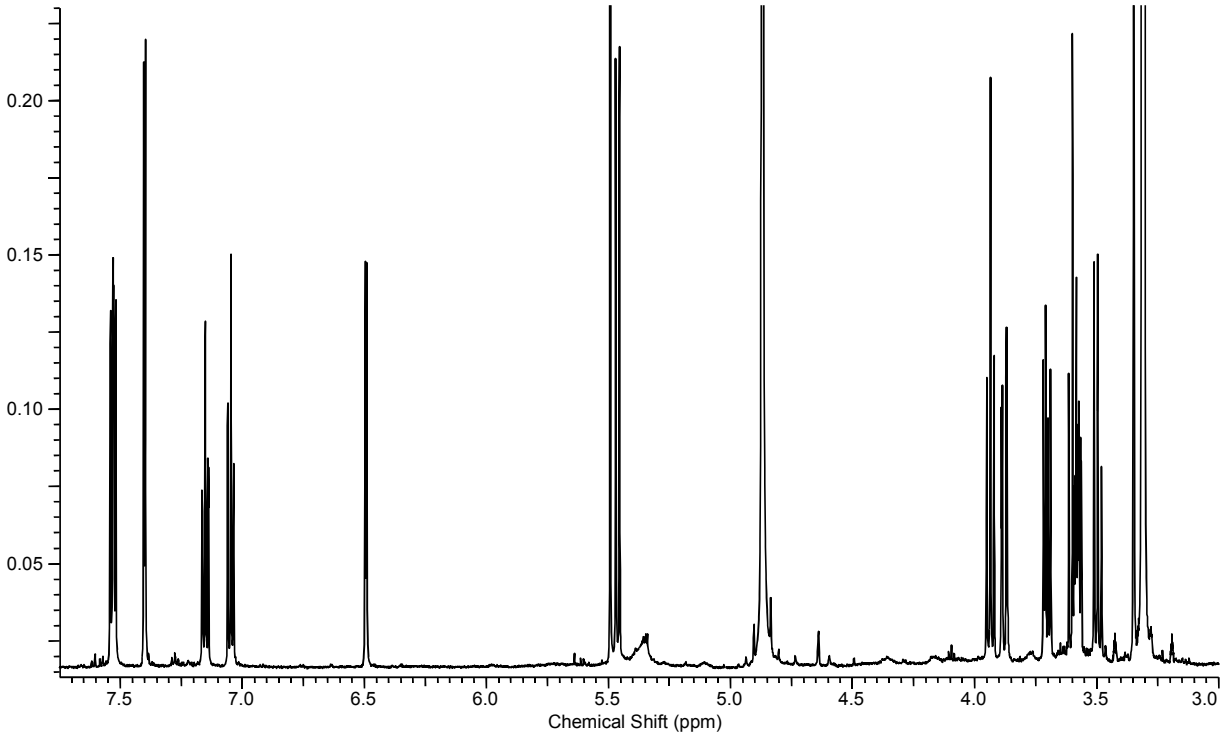
$^{1}$Characteristic $^{1}$H NMR signals of (**8**).

**b**

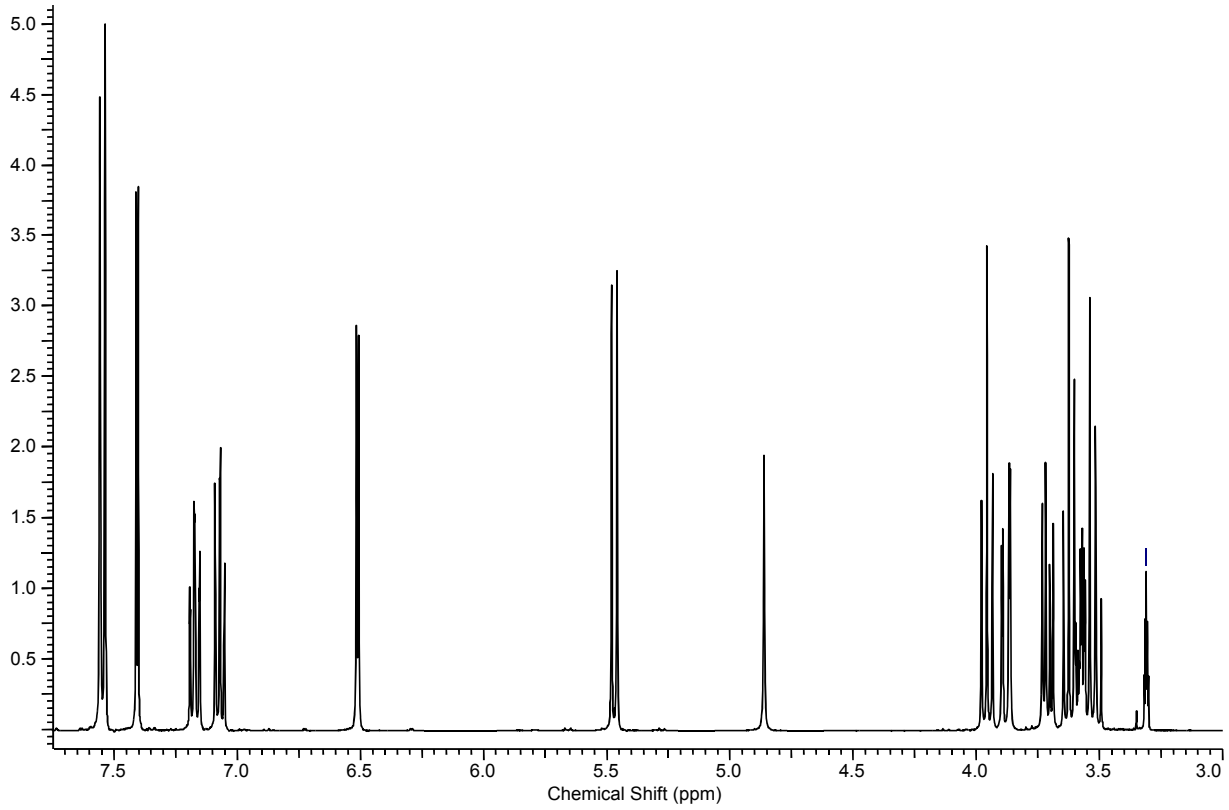
**7** (R = H)
**8** (R = $PO_3H_2$)



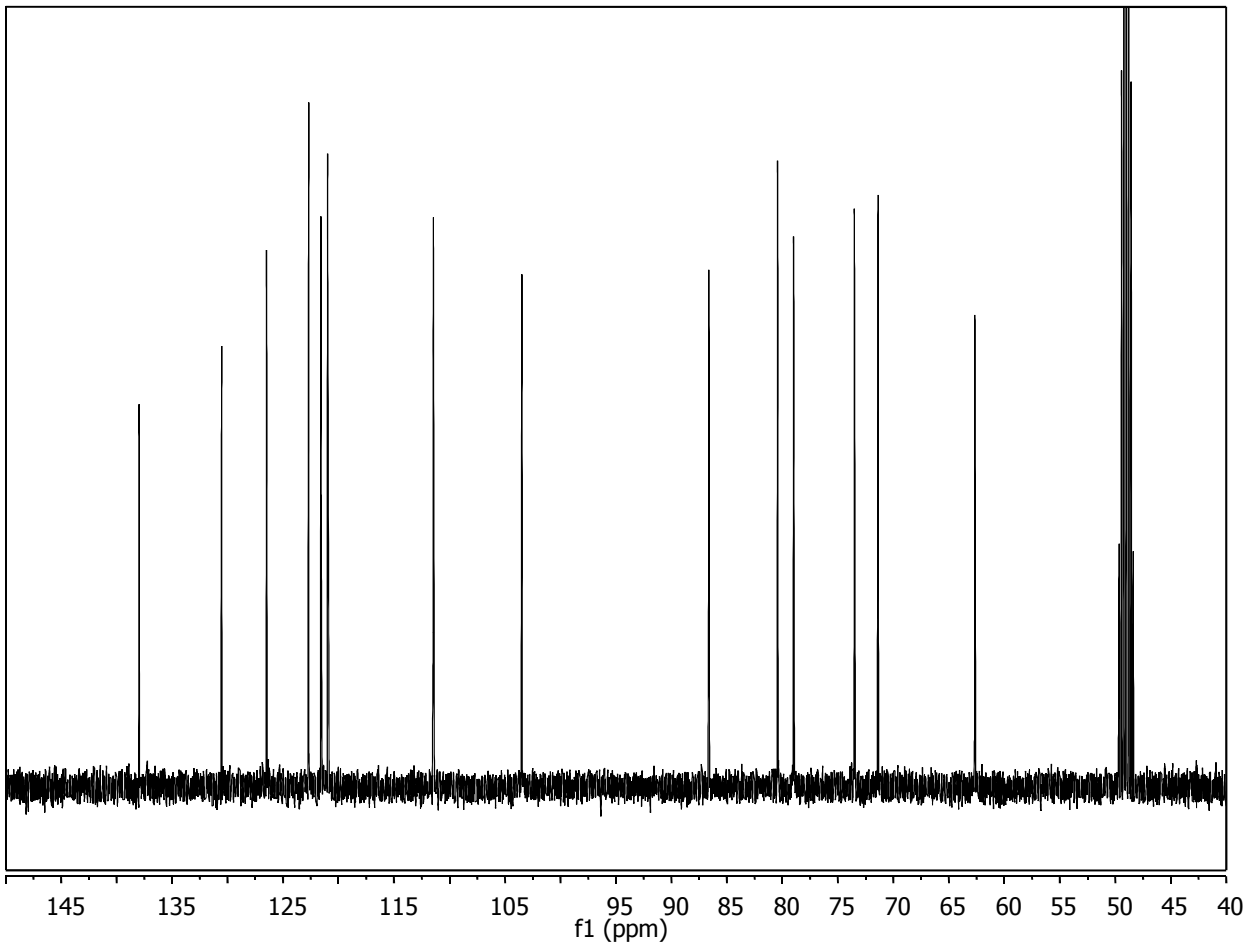

**Supplementary Figure 6.1.** NMR data of (purified or synthetic) *N*-(β-D-glucopyranosyl)-indole (7) and *N*-(3'-*O*-phospho-β-D-glucopyranosyl)-indole (8) ($^{1}$H (600 MHz), $^{13}$C (151 MHz), and HMBC NMR spectroscopic data in methanol-$d_4$. Chemical shifts were referenced to ($CD_2\underline{H}OD$) = 3.31 ppm and ($\underline{C}D_2HOD$) = 49.05 ppm). (a) Table showing assignments (b) Structures




**Supplementary Figure 6.2.** $^{1}$H NMR Spectrum (500 MHz, methanol-$d_4$) of purified *N*-(β-D-glucopyranosyl)-indole (7) from *C. elegans.*

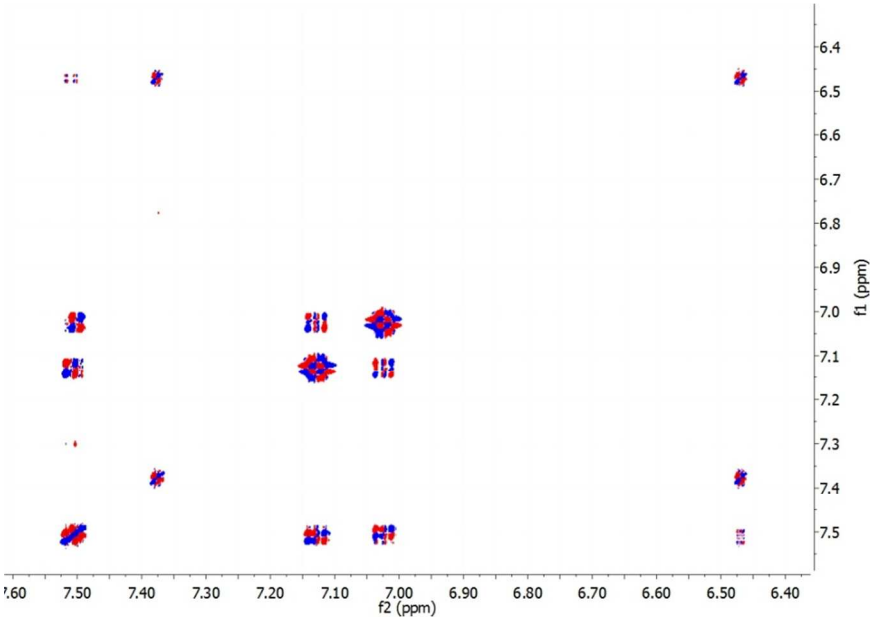



**Supplementary Figure 6.3.** $^{1}$H NMR Spectrum (400 MHz, methanol-$d_4$) of synthetic *N*-(β-D-glucopyranosyl)-indole (7).

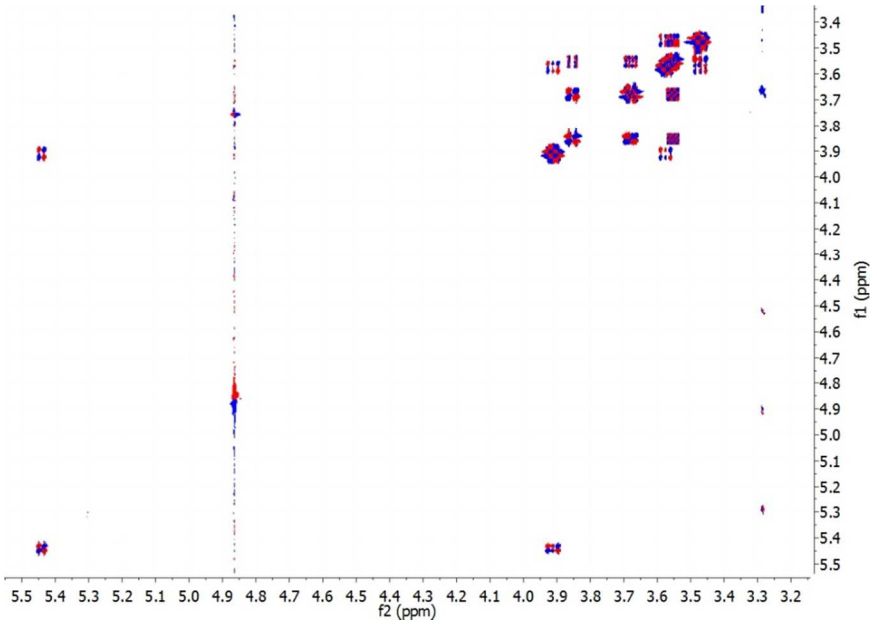



**Supplementary Figure 6.4.** $^{13}$C NMR Spectrum (100 MHz, methanol-$d_4$) of synthetic *N*-(β-D-glucopyranosyl)-indole (7).

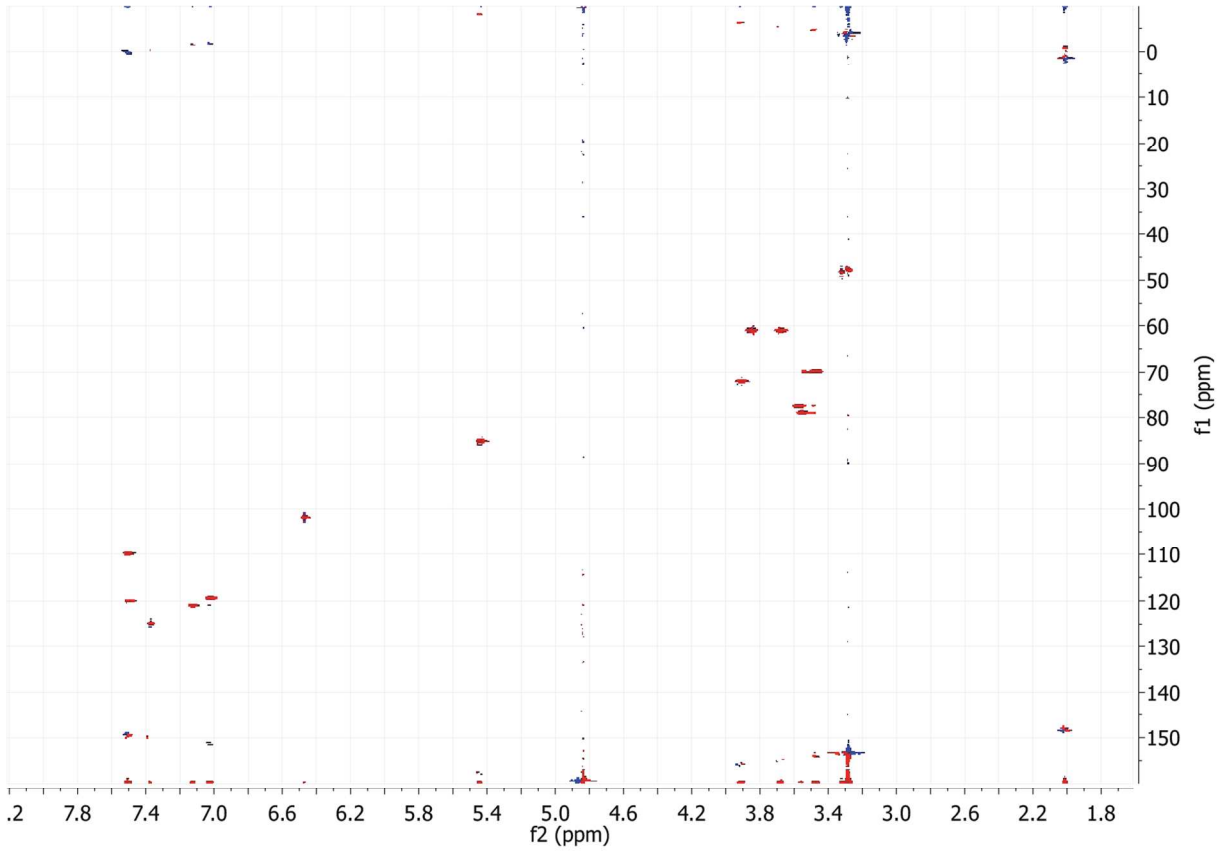
**a**



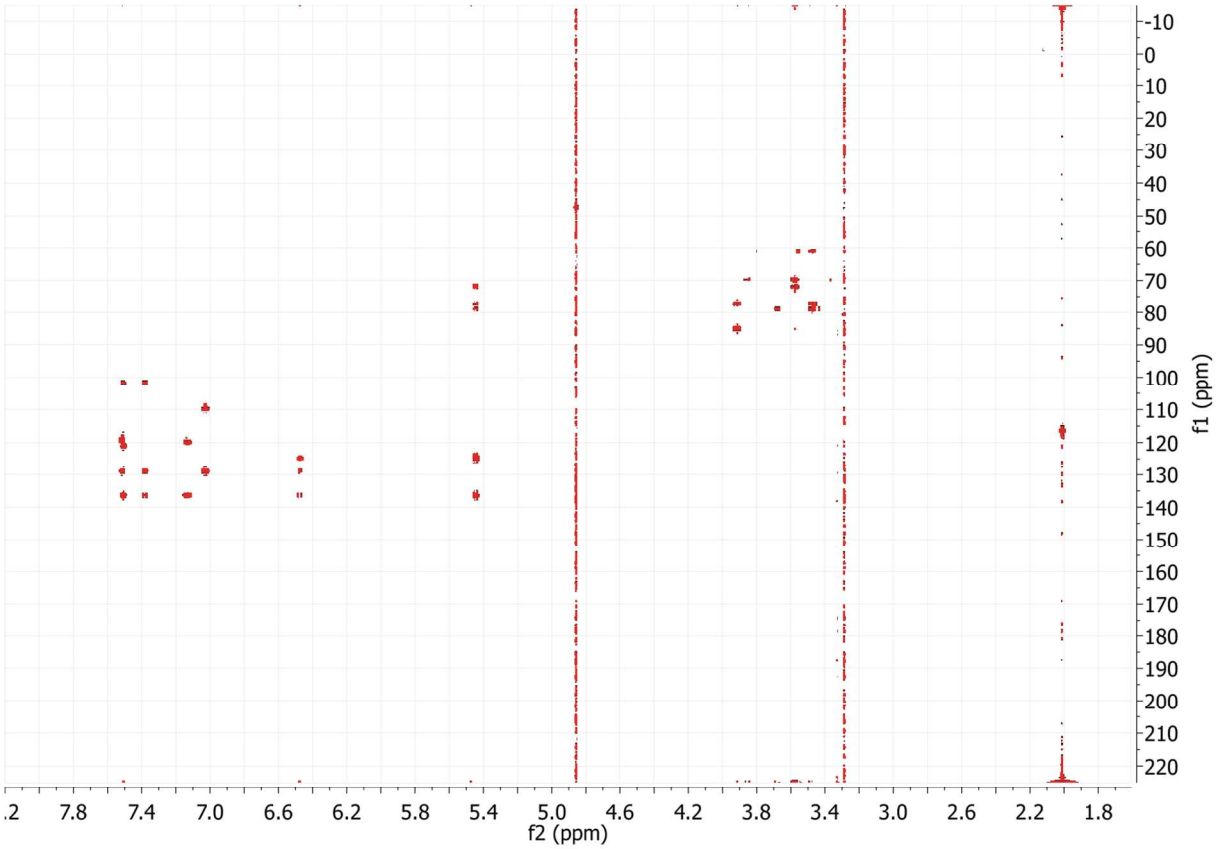

**b**



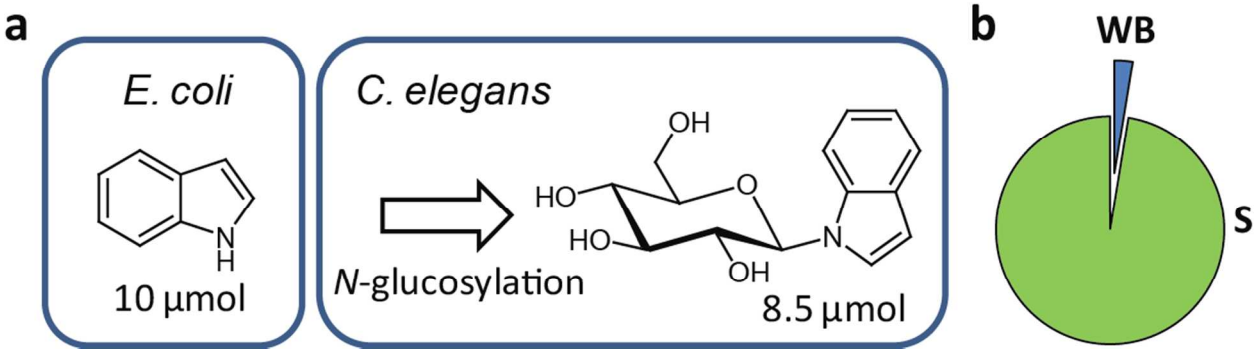

**Supplementary Figure 6.5.** dqfCOSY Spectrum (600 MHz, methanol-$d_4$) of synthetic *N*-( $\beta$ -D-glucopyranosyl)-indole (7) Expansions showing the aromatic (a) and carbohydrate (b) regions.



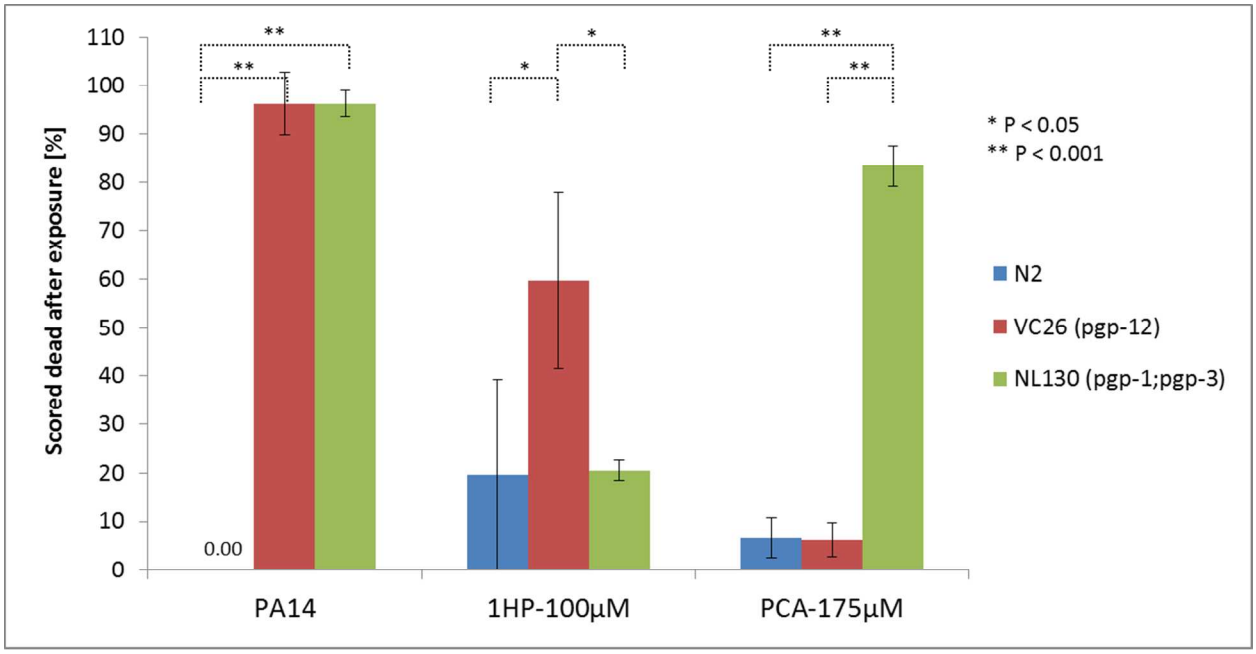

**Supplementary Figure 6.6.** HMQC Spectrum (600 MHz for $^{1}$H 151 MHz for $^{13}$C, methanol-$d_4$) of synthetic *N*-( $\beta$ -D-glucopyranosyl)-indole (7).



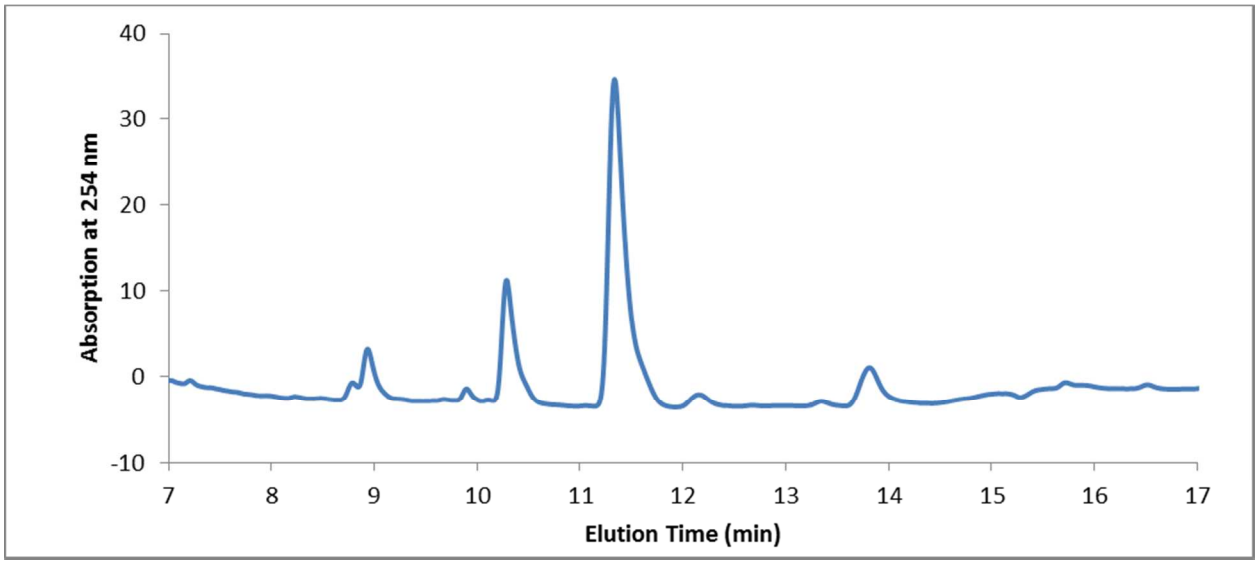

**Supplementary Figure 6.7.** HMBC Spectrum (600 MHz for $^{1}$H 151 MHz for $^{13}$C, methanol-$d_4$) of synthetic *N*-( $\beta$ -D-glucopyranosyl)-indole (7).




**Supplementary Figure 7.** Detoxification of indole via *N*-glucosylation by *C. elegans*. (a) Using HPLC-UV-ESI-MS, we found that of the 10 µmol indole present in a bacterial *E. coli* pellet provided as food, ca. 85% can be recovered after conversion to *N*-(β-D-glucopyranosyl)indole via *N*-glucosylation by *C. elegans.* (b) Comparative analysis of *C. elegans* supernatant (S) and worm body (WB) extracts indicated that *N*-(β-D-glucopyranosyl)indole is predominantly released into the media supernatant.




**Supplementary Figure 8.** Toxicity assay of wild-type (N2) and mutant (*pgp-12* knockout – VC26 or *pgp-1;pgp-3* knockout – NL130) worms. L4 staged worms after 6 hours on PG plates grown with PA14, 6 hours on M9 agar plates with 100 µM of 1HP, or 6 hours on acetic acid pH 4 plates with 175 µM of phenazine-1-carboxylic acid (PCA). N=3 or 4 with 10-30 worms each.




**Supplementary Figure 9.** HPLC-UV-Chromatogram of *C. elegans* VC26 supernatants after treatment with 100 µM 1-HP.

## Supplementary Methods

**Toxicity Assays.** *C. elegans* eggs were arrested at L1 stage for 24 hours in M9 and then grown for 42-48 hours at 20°C on NGM plates to L4 stage as verified by observation with a stereoscope. Worms were washed off plates, allowed to settle, and dispensed onto test plates. For the PG test plates with PA14, 5 µL of an overnight culture of PA14 in LB was spread on peptone-glucose plates (1% Bacto-Peptone, 1% NaCl, 1% glucose, 2% agar), incubated at 37 C for 24 hours and then placed at room temperature for 8-12 hrs. M9 agar plates were made with M9 buffer and 2% agar and were supplemented with 1-HP in DMSO. Acetic acid pH 4 plates were made with 0.1M acetic acid buffer supplemented with phenazine-1-carboxylic acid in DMSO. A small lawn of OP50 was placed on the plate to discourage worms from crawling off. After 6 hours, worms that failed to respond to physical touch with a platinum pick were scored as dead.