*Supplementary Materials*

# Metabolic Impact of Anticancer Drugs Pd2Spermine and Cisplatin on the Brain of Healthy Mice

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## Content of Supplementary Materials:

**Figure S1.** PCA and PLS-DA scores scatter plots for the ^{1}H NMR spectra of polar extracts from brain of healthy BALB/c mice after exposure to cDDP at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls.

**Figure S2.** PCA and PLS-DA scores scatter plots for the ^{1}H NMR spectra of polar extracts from brain of healthy BALB/c mice after exposure to Pd2Spm at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls.

**Figure S3.** Examples of ^{1}H NMR spectral regions illustrating differences in a) polar and b) nonpolar metabolites between cDDP-exposed (blue trace) and Pd2Spm-exposed (red trace), after 48 h of exposure.
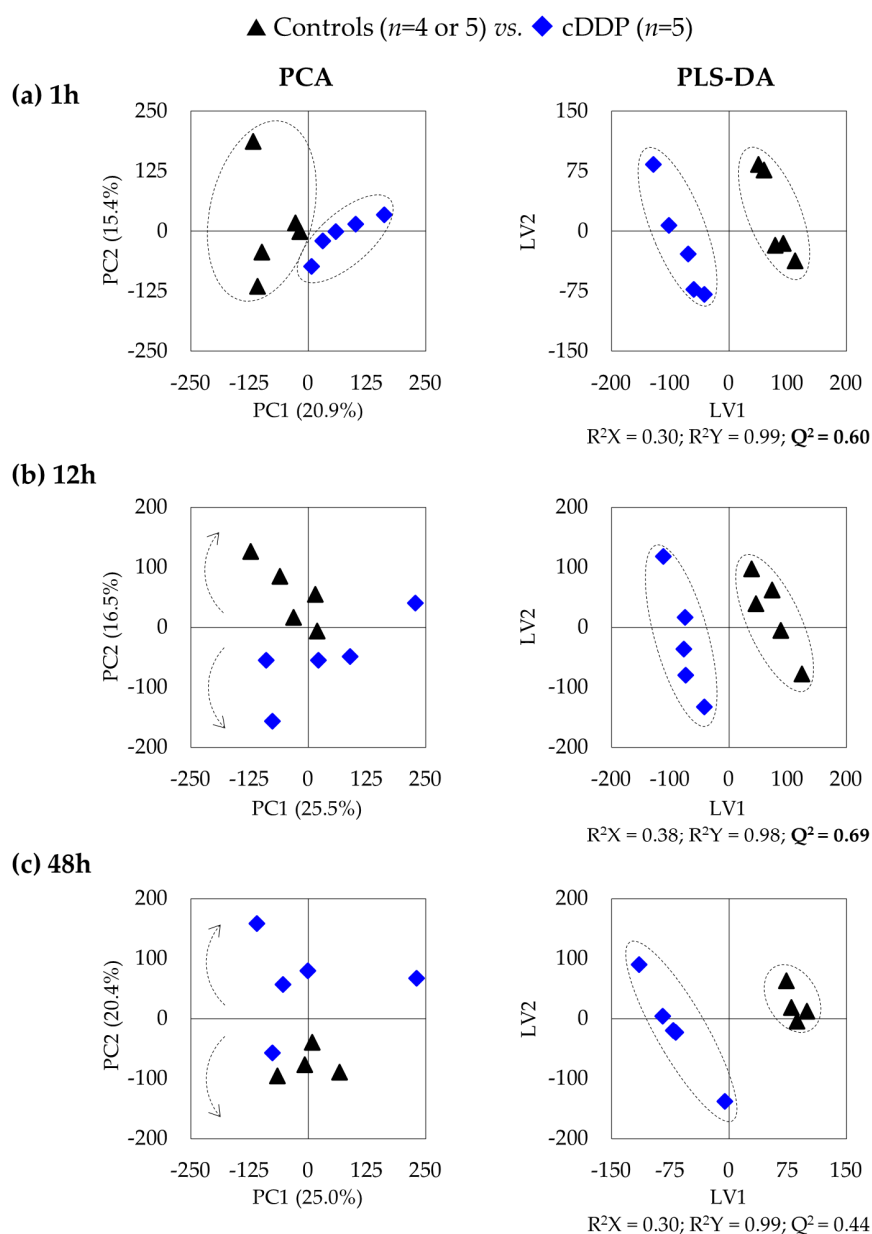
**Figure S4.** PCA and PLS-DA scores scatter plots for the ^{1}H NMR spectra of nonpolar extracts from brain of healthy BALB/c mice after exposure to cDDP at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls.

**Figure S5.** PCA and PLS-DA scores scatter plots for the ^{1}H NMR spectra of nonpolar extracts from brain of healthy BALB/c mice after exposure to Pd2Spm at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls.

**Table S1.** List of metabolites and corresponding spin systems identified in the 500MHz ^{1}H NMR spectra of polar and nonpolar extracts of brain from healthy BALB/c mice, at 1 h post-injection time with phosphate buffer saline.

**Table S2.** Significant metabolite variations (expressed in effect size, ES) in the polar (top) and nonpolar (bottom) metabolomes of mice brain exposed to Pd2Spm, compared to cDDP, at 1, 12 and 48 h post-injection times.

## Figure S1



**Figure S1.** PCA and PLS-DA scores scatter plots for the $^1$H NMR spectra of polar extracts from brain of healthy BALB/c mice after exposure to cDDP at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls. Validation parameters ($R^2$ and $Q^2$) are indicated for each PLS-DA model; $Q^2$ values > 0.5 are highlighted in bold.

## Figure S2
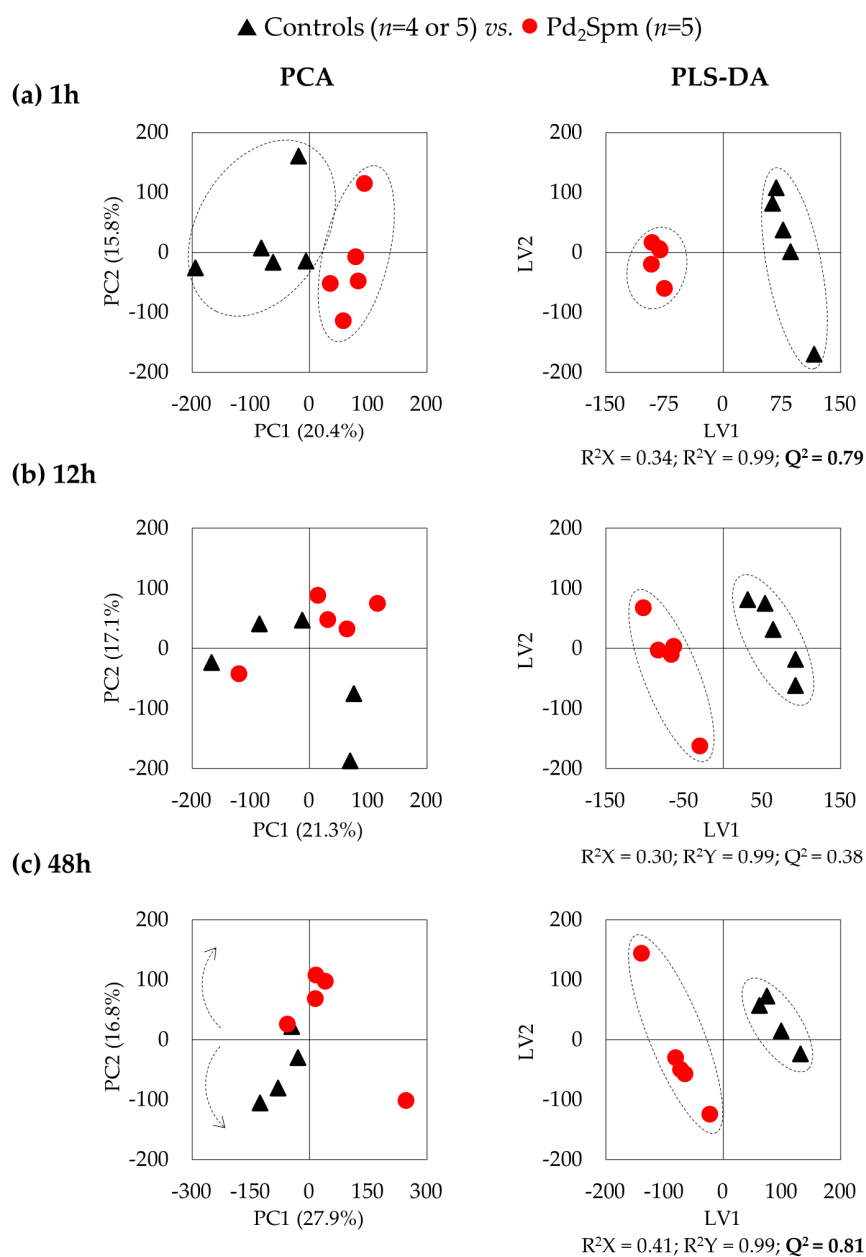
▲ Controls (*n*=4 or 5) *vs.* ● Pd$_2$Spm (*n*=5)



**Figure S2.** PCA and PLS-DA scores scatter plots for the $^1$H NMR spectra of polar extracts from brain of healthy BALB/c mice after exposure to Pd$_2$Spm at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls. In PLS-DA models, Q$^2$ values > 0.5 (in bold) represent robust models.
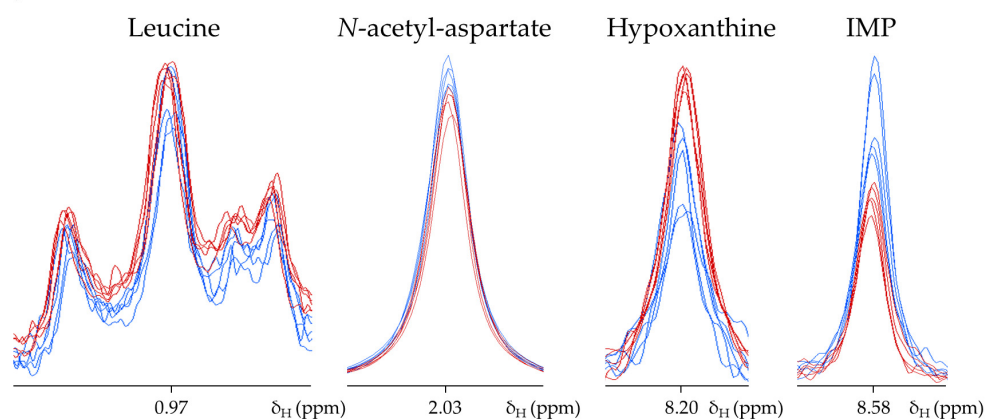
## Figure S3

**a) Polar metabolites**

Leucine    *N*-acetyl-aspartate    Hypoxanthine    IMP

0.97    $\delta_H$ (ppm)    2.03    $\delta_H$ (ppm)    8.20    $\delta_H$ (ppm)    8.58    $\delta_H$ (ppm)

**b) Nonpolar metabolites**

Total cholesterol C6$\underline{H}$    PUFAs =CHC$\underline{H}_2$CH=    MUFAs & PUFAs $\underline{H}$C=C$\underline{H}$

5.45    $\delta_H$ (ppm)    2.82    $\delta_H$ (ppm)    5.36    $\delta_H$ (ppm)

PTE, C$\underline{H}_2$[(NH$_3$)$^+$]    PL, glyceryl C2$\underline{H}$

3.15    $\delta_H$ (ppm)    5.20    $\delta_H$ (ppm)

**Figure S3.** Examples of $^1$H NMR spectral regions illustrating differences in a) polar and b) nonpolar metabolites between cDDP-exposed (blue trace) and Pd₂Spm-exposed (red trace), after 48 h of exposure.

# Figure S4

▲ Controls (*n*=4 or 5) *vs.* ◆ cDDP (*n*=5)



**Figure S4.** PCA and PLS-DA scores scatter plots for the $^1$H NMR spectra of nonpolar extracts from brain of healthy BALB/c mice after exposure to cDDP at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls. In PLS-DA models, $Q^2$ values > 0.5 (in bold) represent robust models.
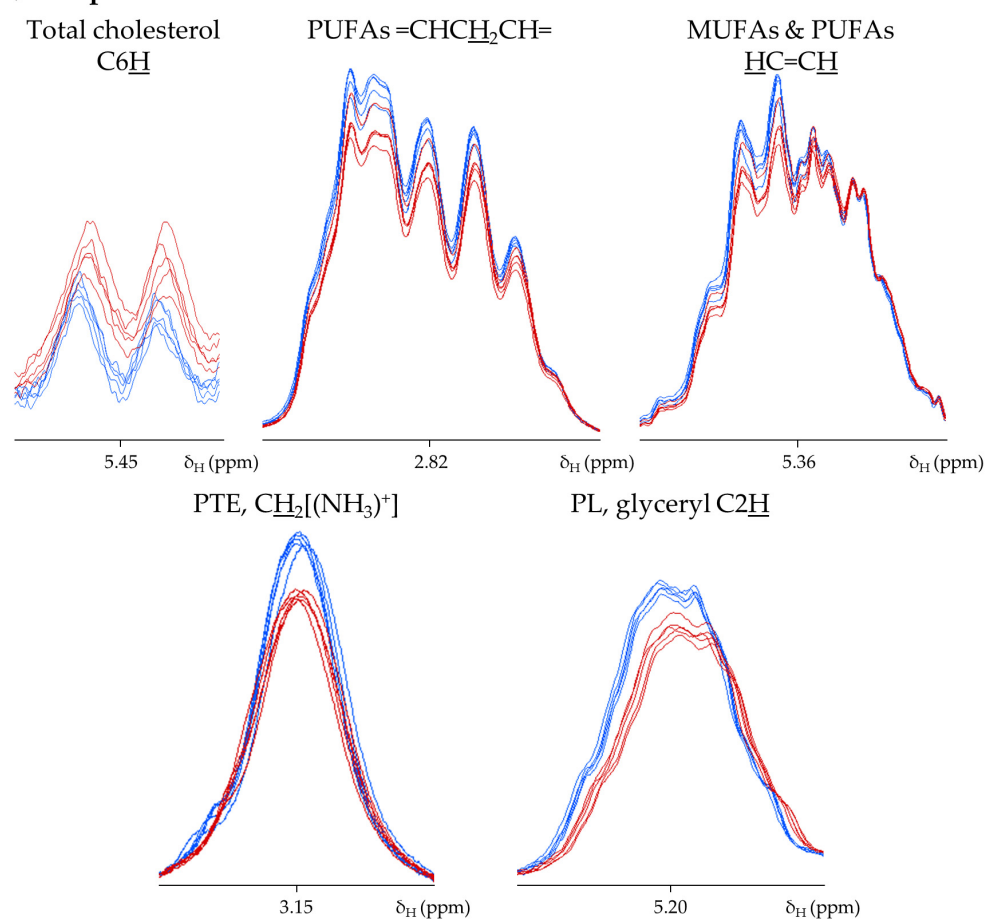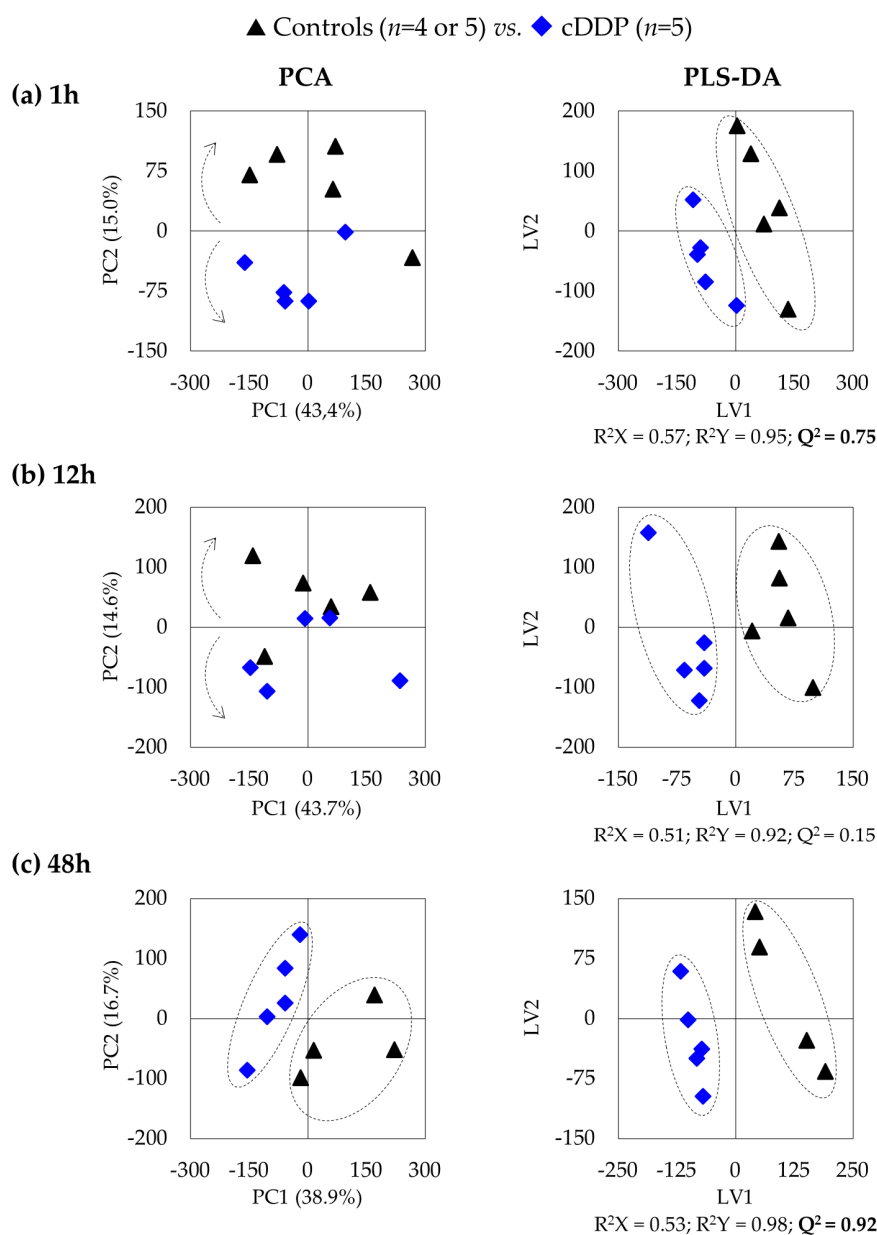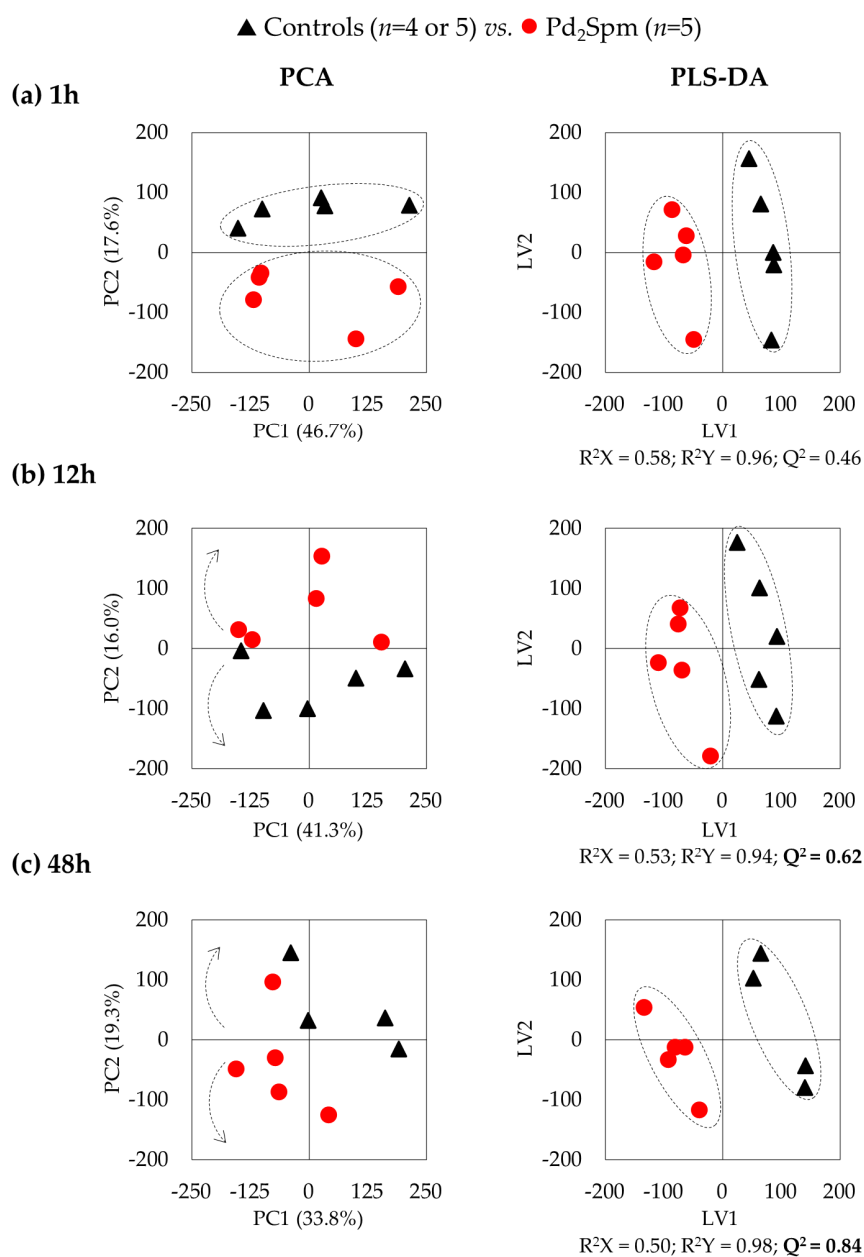
## Figure S5

▲ Controls (*n*=4 or 5) *vs.* ● $Pd_2Spm$ (*n*=5)

**(a) 1h**

**PCA**          **PLS-DA**

$R^2X = 0.58; R^2Y = 0.96; Q^2 = 0.46$

**(b) 12h**

$R^2X = 0.53; R^2Y = 0.94; \mathbf{Q^2 = 0.62}$

**(c) 48h**

$R^2X = 0.50; R^2Y = 0.98; \mathbf{Q^2 = 0.84}$

**Figure S5.** PCA and PLS-DA scores scatter plots for the ^{1}H NMR spectra of nonpolar extracts from brain of healthy BALB/c mice after exposure to Pd2Spm at a) 1 h, b) 12 h, and c) 48 h post-injections times, compared to controls. In PLS-DA models, $Q^2$ values > 0.5 (in bold) represent robust models.

**Table S1.** List of metabolites and corresponding spin systems identified in the 500MHz ^{1}H NMR spectra of polar and nonpolar extracts of brain from healthy BALB/c mice, at 1 h post-injection time with phosphate buffer saline. [a] Predominant metabolites; [b] Metabolites identified, for the first time to our knowledge, by ^{1}H NMR in brain extracts of roedent models; [*] Possible contamination due to the extraction procedure; [†] Tentative assignment. Human Metabolome Database (HMDB ID) metabolite identidication codes are indicated whenever possible. Abbreviations: 3-HBA, 3-hydroxybutyrate; ADP, adenosine diphosphate; AMP, adenosine monophosphate; ATP, adenosine triphosphate; DMA, dimethylamine; DMSO$_2$, dimethyl sulfone; GABA, $\gamma$-aminobutyrate; GPC, glycerophosphocholine; GSH, glutathione (reduced); HX, hypoxanthine; IMP, inosine monophosphate; MUFAs, monounsaturated fatty acids; NAD^{+}, nicotinamide adenine dinucleotide; PC, phosphocholine; PE, phosphoethanolamine; PL, phospholipid; PTC, phosphatidylcholine; PTE, phosphatidylethanolamine; PUFAs, polyunsaturated fatty acids; SM, sphingomyelin; TMAO, Trimethylamine *N*-oxide; s, singlet; d, doublet; dd, double doublet; t, triplet; q, quartet; m, multiplet; br, broad signal.

| | Metabolite | δ$_H$ ppm (multiplicity, assignment) | HMDB ID [61] |
|---|---|---|---|
| **Polar extracts** | | | |
| Amino acids and derivatives | Alanine | 1.48 (d, $\beta$CH$_3$); 3.78 (q, $\alpha$CH) | HMDB0000161 |
| | Asparagine [†] | 2.83 (m, $\beta$CH$_2$) | HMDB0000168 |
| | Aspartate | 2.68 (dd, $\beta$CH); 2.81 (dd, $\beta$'CH); 3.90 (dd, $\alpha$CH) | HMDB0000191 |
| | Creatine [a] | 3.04 (s, N-CH$_3$); 3.93 (s, N-CH$_2$) | HMDB0000064 |
| | GABA [a] | 1.90 (m, $\beta$CH$_2$); 2.30 (t, $\gamma$CH$_2$); 3.01 (t, $\alpha$CH$_2$) | HMDB0000112 |
| | Glutamate [a] | 2.04 (m, $\beta$CH); 2.11 (m, $\beta$'CH); 2.36 (m, $\gamma$CH$_2$); 3.76 (dd, $\alpha$CH) | HMDB0000148 |
| | Glutamine | 2.14 (m, $\beta$CH$_2$); 2.45 (m, $\gamma$CH$_2$); 3.77 (t, $\alpha$CH) | HMDB0000641 |
| | Glycine | 3.55 (s, $\alpha$CH$_2$) | HMDB0000123 |
| | Histidine | 7.06 (s, C4H ring); 8.00 (s, C2H ring) | HMDB0000177 |
| | Isoleucine | 0.94 (t, $\delta$CH3); 1.01 (d, $\beta$'CH3); 1.99 (m, $\beta$CH) | HMDB0000177 |
| | Leucine | 0.96 (t, $\delta$CH$_3$/ $\delta$'CH$_3$); 1.71 (m, $\beta$CH$_2$/ $\gamma$CH) | HMDB0000687 |
| | Lysine | 1.45 (m, $\gamma$CH$_2$); 1.73 (m, $\delta$CH$_2$); 1.92 (m, $\beta$CH$_2$) | HMDB0000182 |
| | *N*-acetylaspartate [a] | 2.02 (s, CH); 2.50, 2.70 (m, CH$_2$); 4.40 (m, CH$_3$) | HMDB0000812 |
| | Phenylalanine | 7.33 (d, C2H/ C6H ring); 7.38 (m, C4H ring); 7.42 (t, C3H/ C5H ring) | HMDB0000159 |
| | Taurine [a] | 3.27 (t, S-CH$_2$); 3.43 (t, N-CH$_2$) | HMDB0000251 |
| | Tyrosine | 6.90 (d, C3H/ C5H ring); 7.20 (d, C2H/ C6H ring) | HMDB0000158 |
| | Valine | 0.99 (d, $\gamma$CH$_3$); 1.05 (d, $\gamma$'CH$_3$); 2.27 (m, $\beta$CH); 3.61 (d, $\alpha$CH) | HMDB0000883 |
| Choline deri- | Choline [a] | 3.21 (s, N(CH$_3$)$_3$) | HMDB0000097 |
| | GPC | 3.23 (s, N(CH$_3$)$_3$); 3.92 (m, $\alpha$CH$_2$); 4.33 (m, PO$_3$-$\alpha$CH$_2$) | HMDB0000086 |
| | PC [a] | 3.22 (s, N(CH$_3$)$_3$); 4.17 (m, PO$_3$-CH$_2$) | HMDB0001565 |
| Nucleotides and derivatives | Adenine [b, †] | 8.23 (s, C2H ring) | HMDB0000034 |
| | Adenosine | 4.29 (q, C4'H ribose); 4.44 (dd, C3'H ribose); 6.10 (d, C1'H ribose); 8.12 (s, C8H ring); 8.27 (s, C2H ring) | HMDB0000050 |
| | ADP | 6.15 (d, C1'H ribose); 8.27 (s, C2H ring); 8.54 (s, C8H ring) | HMDB0001341 |
| | AMP | 4.51 (dd, C2'H ribose); 6.14 (d, C1'H ribose); 8.27 (s, C2H ring); 8.61 (s, C8H ring) | HMDB0000045 |
| | ATP | 6.15 (d, C1'H ribose); 8.28 (s, C2H ring); 8.545 (s, C8H ring) | HMDB0000538 |
| | Hypoxanthine [b] | 8.18 (s, C2H); 8.20 (s, C8H) | HMDB0000157 |
| | IMP | 8.24 (s, C8H ring); 8.58 (s, C8H ring) | HMDB0000175 |
| | Inosine | 6.10 (d, C1'H); 8.24 (s, C8H ring); 8.35 (s, C2H ring) | HMDB0000195 |
| | NAD^{+} [†] | 8.43 (s, A8) | HMDB0000902 |
| | Uridine | 5.90 (d, C5H ring); 5.92 (d, C1'H ring); 7.88 (d, C6H ring) | HMDB0000285 |
| Organi | 3-HBA | 1.20 (d, CH$_3$); 2.31 (m, CH$_2$) | HMDB0000357 |
| | Acetate | 1.92 (s, CH$_3$) | HMDB0000042 |

| | | | |
|---|---|---|---|
| | Ascorbate | 4.52 (d, C4H ring) | HMDB0000044 |
| | Formate | 8.46 (s, CH) | HMDB0000142 |
| | Fumarate | 6.52 (s, CH) | HMDB0000134 |
| | Lactate [a] | 1.33 (d, $CH_3$); 4.10 (q, CH) | HMDB0000190 |
| | Succinate | 2.41 (s, $CH_2$) | HMDB0000254 |
| Other compounds | Acetone [b] | 2.24 (s, $CH_3$) | HMDB0001659 |
| | DMA [b] | 2.73 (s, $(CH_3)_2$) | HMDB0000087 |
| | $DMSO_2$ [b, †] | 3.15 (s, $CH_3$) | HMDB0004983 |
| | Ethanol * | 1.19 (t, $CH_3$); 3.65 (q, $CH_2$) | HMDB0000108 |
| | *m*-Inositol [a] | 3.28 (t, C5H); 3.54 (dd, C1H/ C3H) 3.62 (t, C4H/ C6H); 4.06 (t, C2H) | HMDB0000211 |
| | Niacinamide | 7.60 (m, C5H); 8.72 (m, C4H, C6H); 8.94 (m, C2H) | HMDB0001406 |
| | PE [b, †] | 3.99 (m, $PO_3$-$CH_2$) | HMDB0000224 |
| | Propylene glycol * | 1.15 (d, $CH_3$); 3.87 (m, CH) | HMDB0001881 |
| | TMAO [b] | 3.27 (s, $CH_3$) | HMDB0000925 |
| **Nonpolar extracts** | | | |
| Cholesterol | Free [a] | 0.68 (s, $C18H_3$); 0.86 (d, $C26H_3$); 0.87 (d, $C27H_3$); 0.91 (s, $C27H_3$); 0.95 (m, C9H); 0.99 (m, C14H); 1.01 (s, $C19H_3$); 1.07 (m, C1H); 1.10 (m, C17H); 1.12 (m, C23H/ C24H); 1.13 (m, C22H); 1.49 (m, C11H); 1.51 (m, C2H); 1.84 (m, C4H); 1.87 (t, C16H); 1.95 (t, C7H); 1.99 (t, C7H′); 2.26 (t, C4H′); 3.53 (m, C3H); 5.45 (d, C6H) | HMDB0000067 |
| | Esterified | 0.68 (s, $C18H_3$); 0.86 (d, $C26H_3$); 0.87 (d, $C27H_3$); 0.91 (s, $C27H_3$); 0.95 (m, C9H); 0.99 (m, C14H); 1.01 (s, $C19H_3$); 1.10 (m, C17H); 1.12 (m, C23H/ C24H); 1.13 (m, C22H); 1.15 (m, C1H); 1.84 (t, C4H); 1.87 (t, C15H′); 1.95 (t, C7H); 1.99 (t, C7H′); 2.31 (t, C4H′); 5.45 (d, C6H) | Undefined |
| Fatty acids (FA) | Saturated | 0.89 (br, $CH_3$); 1.25 (br, $(CH_2)_n$); 2.30 (m, $CH_2CO$) | Undefined |
| | Monounsaturated | 2.02 ($\omega7/\omega9$ $CH_2CH_2CH=$); 2.30 (m, $\omega7/\omega9$ $CH_2CO$); 5.34 (m, **HC=CH**) | 16:1 ($\Delta^9$;$\omega7$), HMDB0031053 / 18:1 ($\Delta^9$;$\omega9$), HMDB0000207 |
| | Polyunsaturated [a] | 0.93 (t, $\omega6$ $CH_3$); 0.98 (t, $\omega3$ $CH_3$); 1.30 (m, $(CH_2)_n$); 1.33 (m, $\omega3/\omega6$ $=CHCH_2(CH_2)_n$); 2.05 (m, $CH_2CH=$); 2.30 (m, $CH_2CO$); 2.77 (t, 18:2 $=CHCH_2CH=$); 2.81 (m, $\omega3/\omega6$ $=CHCH_2CH=$); 2.82 (m, 22:6 $=CHCH_2CH=$); 2.84 (m, 22:6 $CH_2CO$); 5.37 (m, **HC=CH**) | 18:2 ($\Delta^{9,12}$; $\omega6$), HMDB0000673 22:6 ($\Delta^{4,7,10,13,16,19}$; $\omega3$), HMDB0002183 and other undefined $\omega3/\omega6$ FAs |
| Phospholipids | PL resonances | 3.75 (br, $CH_2N(CH_3)_3$); 3.94 (br, Glyceryl $C3H_2$); 4.38 (br, $POCH_2$); 4.40 (br, Glyceryl $C1H_2$); 5.20 (br, Glyceryl C2H) | Undefined |
| | PTC [a] | 3.31 (s, $N(CH_3)_3$); 4.12 (m, Glyceryl, $C1H_2$) | Undefined |
| | PTE [a] | 3.15 (br, $CH_2[(NH_3)^+]$); 8.60 (br, $[(NH_3)^+]$) | Undefined |
| | SM | 3.29 (s, $N(CH_3)_3$) | Undefined |

**Table S2.** Significant metabolite variations (expressed in effect size, ES) in the polar (top) and nonpolar (bottom) metabolomes of mice brain exposed to Pd$_2$Spm, compared to cDDP, at 1, 12 and 48 h post-injection times. Only variations with |ES| > ES Error and *p*-value < 0.05 are shown. [†] Tentative assignment. [‡] Partial integration of resonance peak. [a] Metabolic variation statistically significant after False Discovery Rate (FDR) correction; [b] Metabolite observed to vary only in the direct comparison of the two drugs (and not in comparison with controls). Abbreviations as defined in Tables 1 and S1.

| | | | Pd$_2$Spm *vs.* cDDP | | | | | |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| | | | **1h** | | **12h** | | **48h** | |
| Family/ assignment | Metabolite | $\delta_H$/ ppm (multiplicity) | ES ±Error | *p*-value | ES ±Error | *p*-value | ES ±Error | *p*-value |
| Amino acids and derivatives | Alanine | 1.48 (d) | -3.2 ± 1.9 | $2.4 \times 10^{-3}$ [a] | — | — | 1.5 ± 1.4 | $4.9 \times 10^{-2}$ |
| | Leucine | 0.96 (t) | -1.8 ± 1.5 | $2.4 \times 10^{-2}$ | — | — | 3.1 ± 1.8 | $5.1 \times 10^{-3}$ [a] |
| | *N*-acetylaspartate | 2.03 (s) | — | — | — | — | -3.0 ± 1.8 | $1.7 \times 10^{-3}$ [a] |
| | Taurine [b] | 3.43 (t) | — | — | — | — | -2.4 ± 1.6 | $1.3 \times 10^{-2}$ |
| | Valine | 1.05 (d) | -3.0 ± 1.8 | $1.7 \times 10^{-3}$ [a] | — | — | — | — |
| Choline derivatives | Glycerophosphocholine | 3.23 (s) | 2.5 ± 1.7 | $1.4 \times 10^{-2}$ | — | — | — | — |
| | Phosphocholine [b] | 3.21 (s) | — | — | — | — | -2.1 ± 1.5 | $1.2 \times 10^{-2}$ |
| Nucleotides and derivatives | Adenine [†,b] | 8.23 (s) | — | — | — | — | -2.2 ± 1.6 | $2.0 \times 10^{-2}$ |
| | Adenosine | 4.29 (q) | — | — | — | — | 1.5 ± 1.4 | $1.6 \times 10^{-2}$ |
| | ADP | 8.54 (s) | 1.9 ± 1.5 | $1.8 \times 10^{-2}$ | — | — | -1.6 ± 1.4 | $3.9 \times 10^{-2}$ |
| | HX | 8.20 (s) | — | — | — | — | 2.9 ± 1.8 | $6.2 \times 10^{-3}$ [a] |
| | IMP | 8.58 (s) | — | — | — | — | -3.0 ± 1.8 | $4.8 \times 10^{-3}$ [a] |
| | Inosine | 8.35 (s) | -1.8 ± 1.5 | $2.8 \times 10^{-2}$ | — | — | — | — |
| Organic acids | Formate | 8.46 (s) | — | — | -1.8 ± 1.5 | $3.4 \times 10^{-2}$ | — | — |
| Other compounds | Acetone | 2.24 (s) | -5.7 ± 2.8 | $1.7 \times 10^{-3}$ [a] | — | — | — | — |
| | DMA | 2.73 (s) | 4.4 ± 2.3 | $2.3 \times 10^{-3}$ [a] | — | — | 2.2 ± 1.6 | $1.7 \times 10^{-2}$ |
| | DMSO$_2$ [†] | 3.15 (s) | 14.5 ± 6.5 | $7.9 \times 10^{-3}$ | 3.1 ± 1.8 | $7.9 \times 10^{-3}$ | — | — |
| Unassigned resonances | U1 | 0.80 (t) | -2.2 ± 1.6 | $2.1 \times 10^{-2}$ | — | — | — | — |
| | U4 [b] | 2.06 (s‡) | — | — | — | — | 2.4 ± 1.6 | $3.2 \times 10^{-2}$ |
| | U5 [b] | 6.08 (s) | — | — | — | — | -1.8 ± 1.5 | $4.5 \times 10^{-2}$ |
| Cholesterol | Free, C3H | 3.53 (m) | — | — | — | — | 3.1 ± 1.8 | $7.9 \times 10^{-3}$ [a] |
| | Total, C6H | 5.45 (d) | — | — | — | — | 3.2 ± 1.9 | $2.7 \times 10^{-3}$ [a] |
| | Total, C7H & C7H′ | 1.95-1.97 | — | — | — | — | 1.9 ± 1.5 | $2.5 \times 10^{-2}$ |
| | Total, C26H$_3$ | 0.86 (d) | — | — | — | — | 1.8 ± 1.5 | $2.8 \times 10^{-2}$ |
| | Total, C27H$_3$ | 0.87 (d) | — | — | — | — | 4.3 ± 2.3 | $1.6 \times 10^{-4}$ [a] |
| Fatty acids | **CH$_3$** | 0.89 (br) | — | — | — | — | 2.8 ± 1.7 | $7.9 \times 10^{-3}$ [a] |
| | Saturated, (**CH$_2$**)$_n$ | 1.25 (br) | 2.8 ± 1.7 | $7.9 \times 10^{-3}$ [a] | — | — | 2.3 ± 1.6 | $7.2 \times 10^{-3}$ [a] |
| | MUFAs ($\Delta^9$; $\omega$7/$\omega$9), **CH$_2$**CH$_2$CH= | 2.02 (m) | — | — | — | — | 1.9 ± 1.5 | $2.1 \times 10^{-2}$ |
| | MUFAs, **HC=CH** | 5.34 (m) | — | — | — | — | 2.5 ± 1.6 | $7.9 \times 10^{-3}$ [a] |
| | PUFAs ($\omega$3), **CH$_3$** | 0.98 (t) | — | — | — | — | -1.6 ± 1.4 | $3.6 \times 10^{-2}$ |
| | PUFAs, (**CH$_2$**)$_n$ | 1.30 (m) | — | — | — | — | 2.3 ± 1.6 | $1.1 \times 10^{-2}$ [a] |
| | PUFAs ($\omega$3/$\omega$6), =CHCH$_2$(CH$_2$)$_n$ | 1.33 (m) | — | — | — | — | 3.4 ± 1.9 | $9.9 \times 10^{-4}$ [a] |
| | PUFAs, **CH$_2$**CH= | 2.05 (m) | — | — | — | — | -1.9 ± 1.5 | $1.6 \times 10^{-2}$ [a] |
| | PUFAs ($\omega$3/$\omega$6), =CH**CH$_2$**CH= | 2.81 (m) | — | — | — | — | -2.4 ± 1.6 | $6.5 \times 10^{-3}$ [a] |
| | PUFAs, **HC=CH** | 5.37 (m) | — | — | — | — | -2.7 ± 1.7 | $3.0 \times 10^{-3}$ [a] |
| | 18:2 ($\Delta^{9,12}$; $\omega$6), =CH**CH$_2$**CH= | 2.77 (t) | -2.5 ± 1.7 | $4.29 \times 10^{-3}$ [a] | -1.3 ± 1.4 | $1.6 \times 10^{-2}$ [a] | -2.0 ± 1.5 | $1.5 \times 10^{-2}$ [a] |
| | 22:6 ($\Delta^{4,7,10,13,16,19}$; $\omega$3), **CH$_2$**CO | 2.38 (m) | — | — | — | — | -3.8 ± 2.1 | $3.3 \times 10^{-4}$ [a] |
| | 22:6 ($\Delta^{4,7,10,13,16,19}$;$\omega$3), =CH**CH$_2$**CH= | 2.82 (m) | — | — | — | — | -3.1 ± 1.8 | $1.4 \times 10^{-3}$ [a] |
| Phospholipids | **CH$_2$**N(**CH$_3$**)$_3$ | 3.75 (br) | 2.3 ± 1.6 | $8.9 \times 10^{-3}$ [a] | 1.7 ± 1.4 | $3.4 \times 10^{-2}$ | — | — |
| | PO**CH$_2$** | 4.38 (br) | — | — | — | — | -5.9 ± 2.9 | $1.2 \times 10^{-4}$ [a] |
| | Glyceryl C3**H$_2$** | 3.94 (br) | — | — | — | — | -5.9 ± 2.9 | $5.8 \times 10^{-5}$ [a] |
| | Glyceryl C1**H$_2$** | 4.40 (br) | — | — | — | — | -4.1 ± 2.2 | $2.0 \times 10^{-4}$ [a] |
| | Glyceryl C2**H** | 5.20 (br) | — | — | — | — | -4.9 ± 2.5 | $8.9 \times 10^{-5}$ [a] |
| | PTC & SM, N(**CH$_3$**)$_3$ | 3.29 - 3.31 | — | — | — | — | -5.6 ± 2.7 | $5.7 \times 10^{-5}$ [a] |
| | PTC, Glyceryl C1**H$_2$** | 4.12 (m) | — | — | — | — | -3.4 ± 2.0 | $7.0 \times 10^{-4}$ [a] |

| Category | Resonance | Shift | Value | $p$ | Value | $p$ | Value | $p$ |
|---|---|---|---|---|---|---|---|---|
| | PTE $CH_2[(NH_3)^+]$ | 3.15 (br) | — | — | — | — | -8.8 ± 4.1 | $9.9 \times 10^{-7}$ [a] |
| | PTE $[(NH_3)^+]$ | 8.60 (br) | — | — | — | — | -2.3 ± 1.6 | $7.4 \times 10^{-3}$ [a] |
| | SM | 5.70 (m) | — | — | — | — | 2.8 ± 1.7 | $4.0 \times 10^{-3}$ [a] |
| Unassigned resonances | U2 | 0.60 (d) | — | — | — | — | -2.3 ± 1.6 | $1.1 \times 10^{-2}$ [a] |
| | U5 | 2.61 (s) | 5.2 ± 2.6 | $3.9 \times 10^{-5}$ [a] | -2.2 ± 1.6 | $9.4 \times 10^{-3}$ [a] | -1.6 ± 1.4 | $4.0 \times 10^{-2}$ |
| | U6 | 2.99 (s) | 4.4 ± 2.3 | $1.3 \times 10^{-3}$ [a] | — | — | -2.1 ± 1.5 | $1.6 \times 10^{-2}$ [a] |
| | U9 | 3.84 (d) | -1.8 ± 1.5 | $2.8 \times 10^{-2}$ | — | — | — | — |
| | U10 | 3.90 (br) | — | — | — | — | 5.5 ± 2.7 | $3.4 \times 10^{-5}$ [a] |
| | U11 | 5.29 (t) | — | — | -2.0 ± 1.5 | $1.6 \times 10^{-2}$ [a] | — | — |
| | U12 | 8.34 (br) | — | — | — | — | 4.0 ± 2.2 | $2.2 \times 10^{-4}$ [a] |
| | U13 | 0.89 (s) | — | — | — | — | 1.8 ± 1.5 | $2.6 \times 10^{-2}$ |
| | U14 | 2.20 (q) | — | — | — | — | 2.5 ± 1.7 | $4.0 \times 10^{-3}$ [a] |
| | U15 | 4.22 (br) | — | — | — | — | 3.5 ± 2.0 | $1.4 \times 10^{-3}$ [a] |
| | U16 | 7.77 (br) | — | — | — | — | 2.7 ± 1.7 | $4.4 \times 10^{-3}$ [a] |