**Supporting Information**

# Machine Learning-enabled Renal Cell Carcinoma Status Prediction Using Multi-Platform Urine-based Metabolomics

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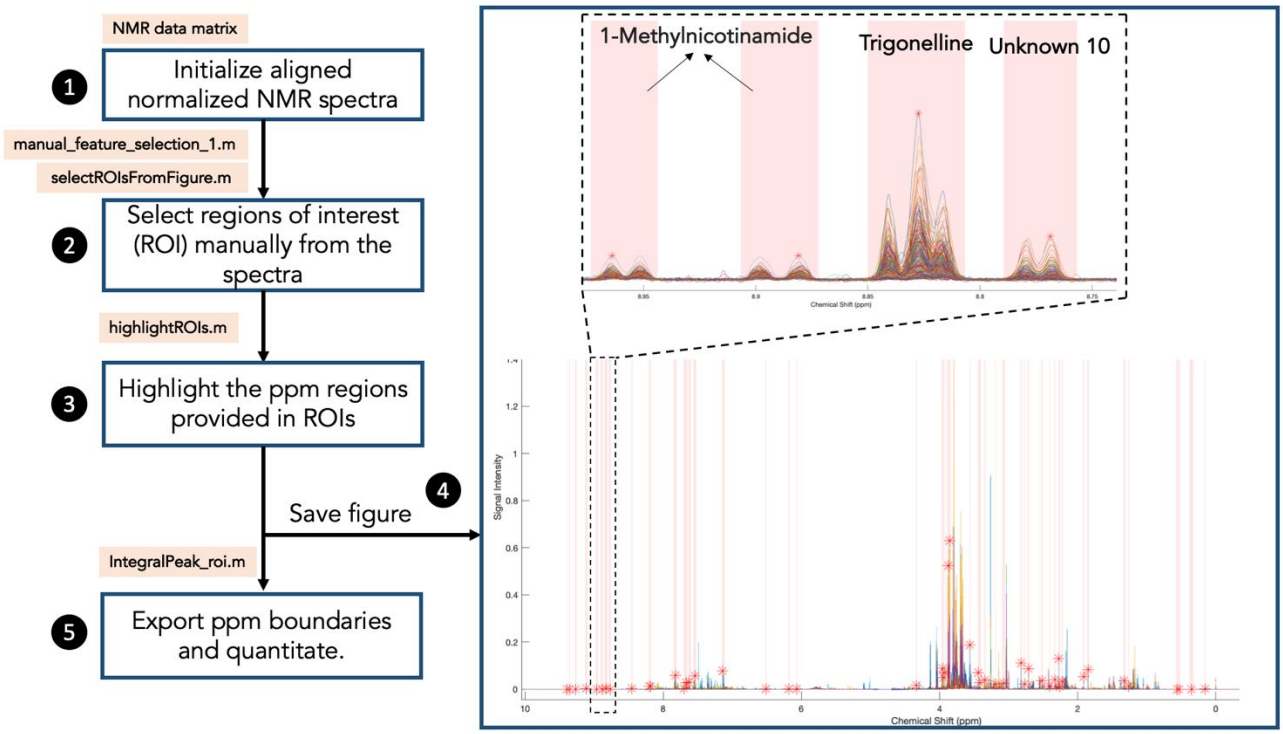
**Supporting information section S1. NMR quality assurance and quality control and Spectral Binning**

All study cohort urine samples were randomized prior to running NMR experiments. Twelve NMR buffer blanks, 3 per run, were added to ensure there is no carry-over of samples between urine samples. One buffer blank was added at the beginning, middle, and end of each run. Thirty-two external controls (Nicotine, Ethanol & Drug Free Human Urine, Female; Golden West Diagnostics, LLC), 8 in each of the four NMR racks (run) used, were added to the study to ascertain the reliability of the sample acquisition process particularly for comparing across runs. One external control was added after the first blank at the beginning the run, and one before the last blank at the end of the run, while the remaining six external controls were randomized with the study's urine samples. The external pooled controls served its purpose as the controls were clustered tightly together on a PCA plot (data not shown). Also, 27 µL were taken out of each study urine sample for two internal pooled controls. 1D $^{1}$H NMR experiment was conducted on all samples, while HSQC was carried out on one internal pooled control sample, and HSQC-TOCSY was carried out on the other. The NMR data for the internal pooled samples were used for metabolite annotation using AssureNMR and COLMARm[1] as described in the manuscript. Fifty metabolomic features in the aligned and normalized 1D $^{1}$H NMR spectra were manually binned and quantified by taking spectral areas for integration in regions without overlap, and combined with MS features for downstream analysis. A manual binning workflow in the Edison laboratory in-house metabolomics toolbox MATLAB scripts was used for binning (https://github.com/artedison/Edison_Lab_Shared_Metabolomics_UGA). **Scheme S1**

shows the steps involved in this process: aligned normalized NMR spectra were initialized, the functions manual_feature_selection1.m and selectROIsFromFigure.m allowed us to manually select multiple regions in the displayed spectra, where a rectangle is drawn around the region of interest (ROI) for binning in an interactive fashion. The highlighROIs.m function highlights the ppm regions provided in ROI, and the binned, highlighted NMR spectra figure is saved. Finally, ppm boundaries are exported, and IntegralPeak_roi.m function was used to integrate metabolomic features in the binned spectra.

**Supporting information scheme S1. NMR Peak Picking Methods**



**Supporting information section S2. Model evaluation metrics**

The following metrics were used for model evaluation, where TP is true positive, TN is true negative, FP is false positive, and FN is false negative:

Accuracy measures the percentage of all correctly predicted samples.

$$Accuracy = \frac{TP + TN}{TP + TN + FP + FN}$$

Sensitivity (or recall) measures the percentage of correctly predicted RCC patients out of the total RCC samples.

$$Sensitivity = \frac{TP}{TP + FN}$$

Specificity measures the percentage of correctly predicted controls out of the total control samples.

$$Specificity = \frac{TN}{TN + FP}$$

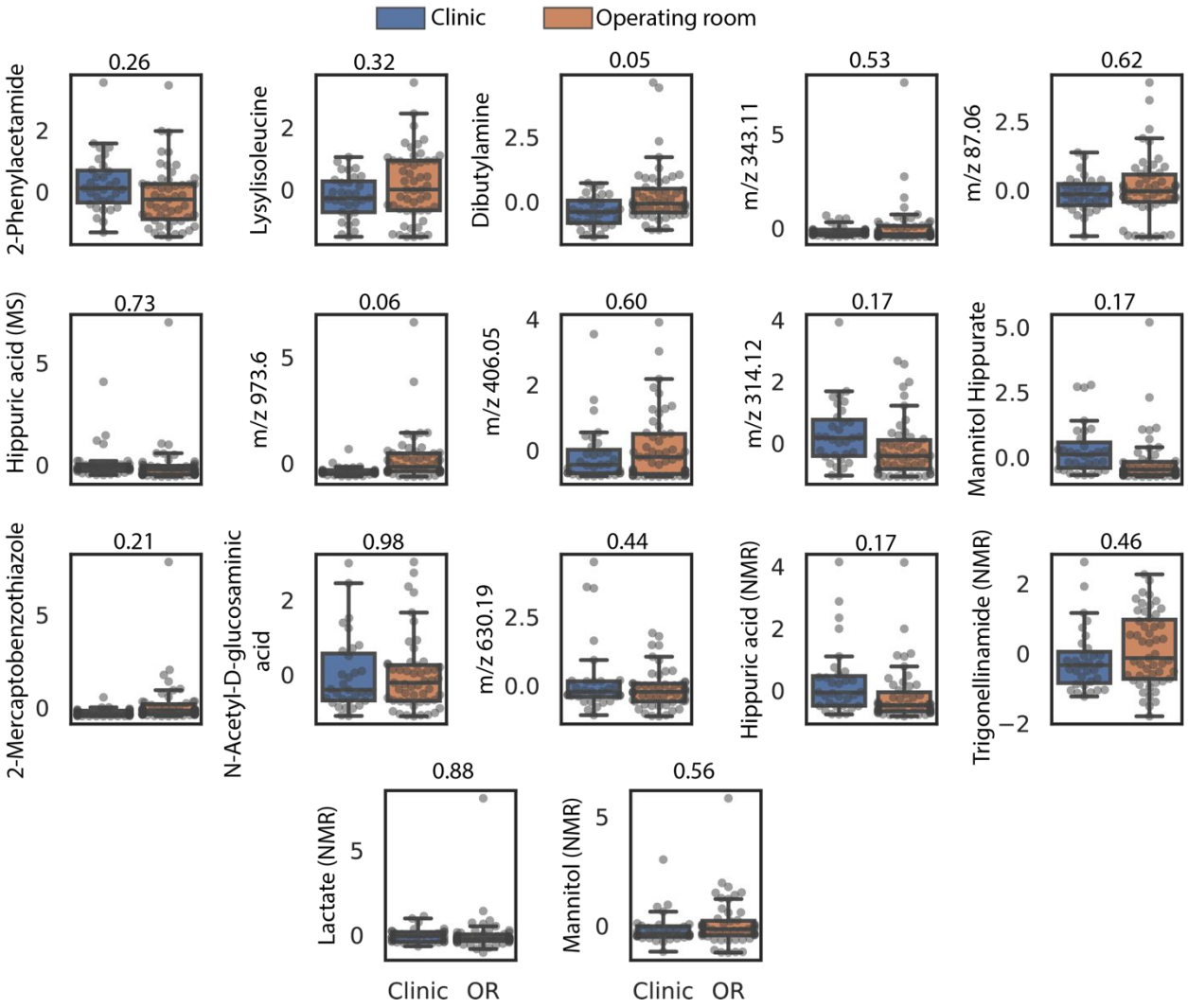



**Figure S1:** Relative quantification of all discriminating metabolomic features identified in the study, for RCC samples collected in the clinic *vs.* operating room. *q*-values were computed by taking the FDR correction (Benjamini-Hochberg) after an independent *t*-test. All features were statistically insignificant. Raw data were transformed *via* autoscaling for visualization.

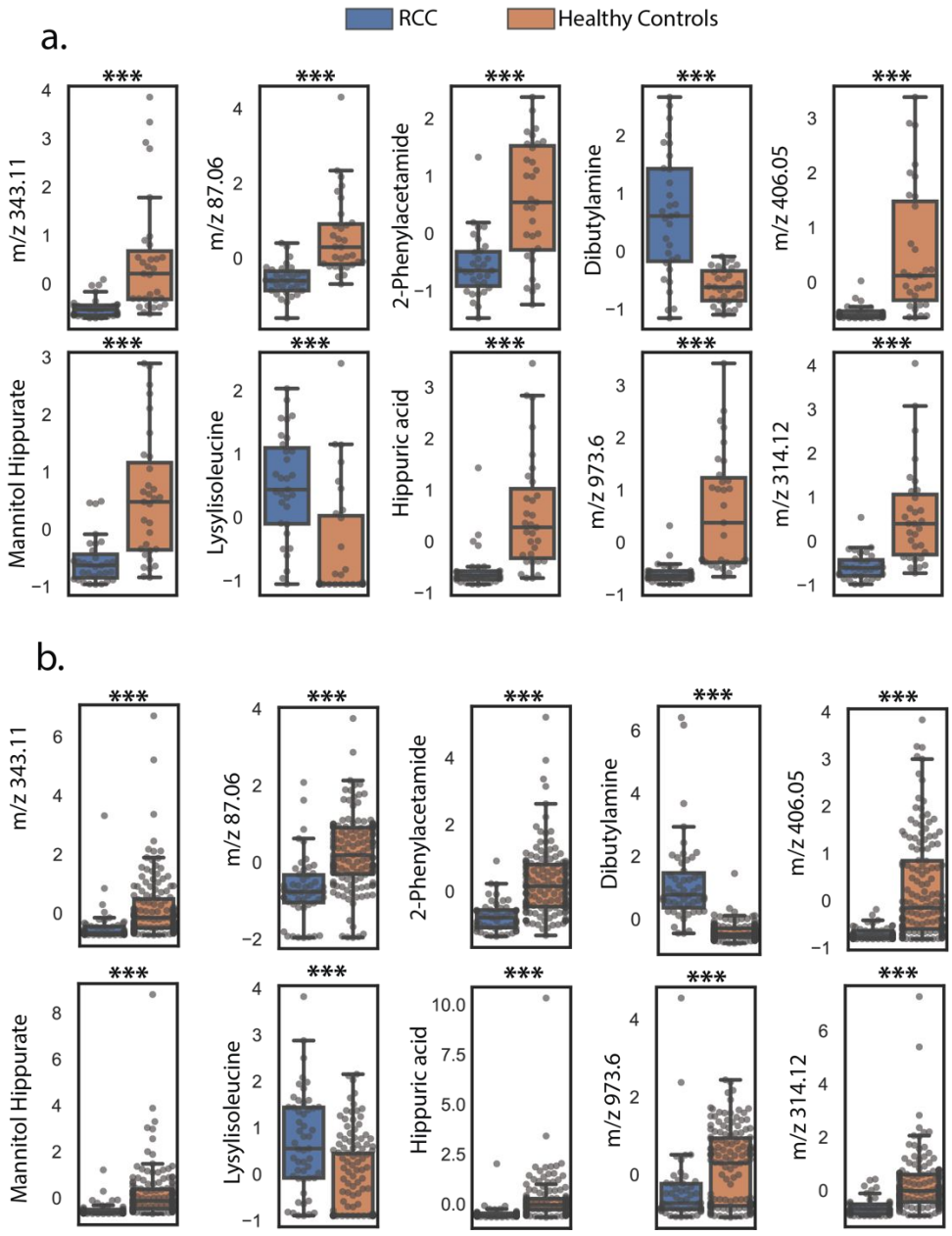



**Figure S2:** Relative quantification of the 10-metabolite panel in (a) the model cohort. After selecting features with greater than a one-fold change between control and RCC groups, *q*-values were computed by taking the FDR correction (Benjamini-Hochberg) after an independent *t*-test. ( $* \, q \leq 0.05, \;\; ** \, q \leq 0.01, \;\; *** \, q \leq 0.001$). (b) Test cohort, *p*-values from the Welch t-test were reported (unequal sample size). (

$* p \leq 0.05$, $** p \leq 0.01$, $*** p \leq 0.001$). Raw data were transformed *via* autoscaling for visualization

**a.**

| ID | Metabolite ID | Mode | RT [min] | FC | *q*-value |
|---|---|---|---|---|---|
| **2102** | dibutylamine, N-butylisobutylamine, diisobutylamine (isomer) | positive | 3.449 | 1.57 | 7.87E-05 |
| **3872** | 3872 | positive | 4.049 | -2.58 | 1.90E-04 |
| **3675** | 3675 | positive | 1.184 | -1.08 | 2.62E-04 |
| **3757** | 3757 | positive | 4.003 | -2.39 | 3.26E-04 |
| **6261** | 6261 | negative | 2.591 | -1.88 | 3.68E-04 |
| **5383** | 5383 | negative | 4.067 | -2.62 | 3.85E-04 |
| **720** | 2-phenylacetamide | positive | 2.562 | -1.07 | 3.86E-04 |
| **4401** | 4401 | positive | 4.039 | -2.69 | 4.81E-04 |
| **6276** | 6276 | negative | 2.636 | -1.65 | 4.81E-04 |

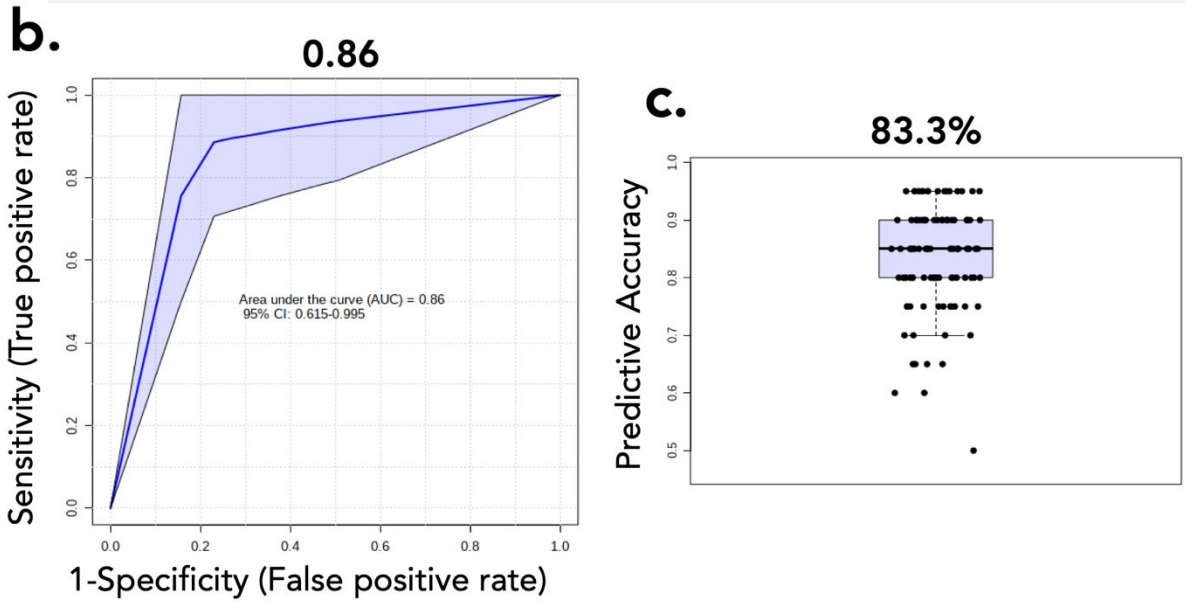



**Figure S3**: Selection of metabolomic features with *q*-values and classifying with logistic regression using the Metaboanalyst 5.0 biomarker analysis platform. (a) Metabolomic features with the top ten highest *q*-values from univariate analysis. (b) ROC-AUC (c) Predictive accuracy. Analysis was carried out using the model cohort.

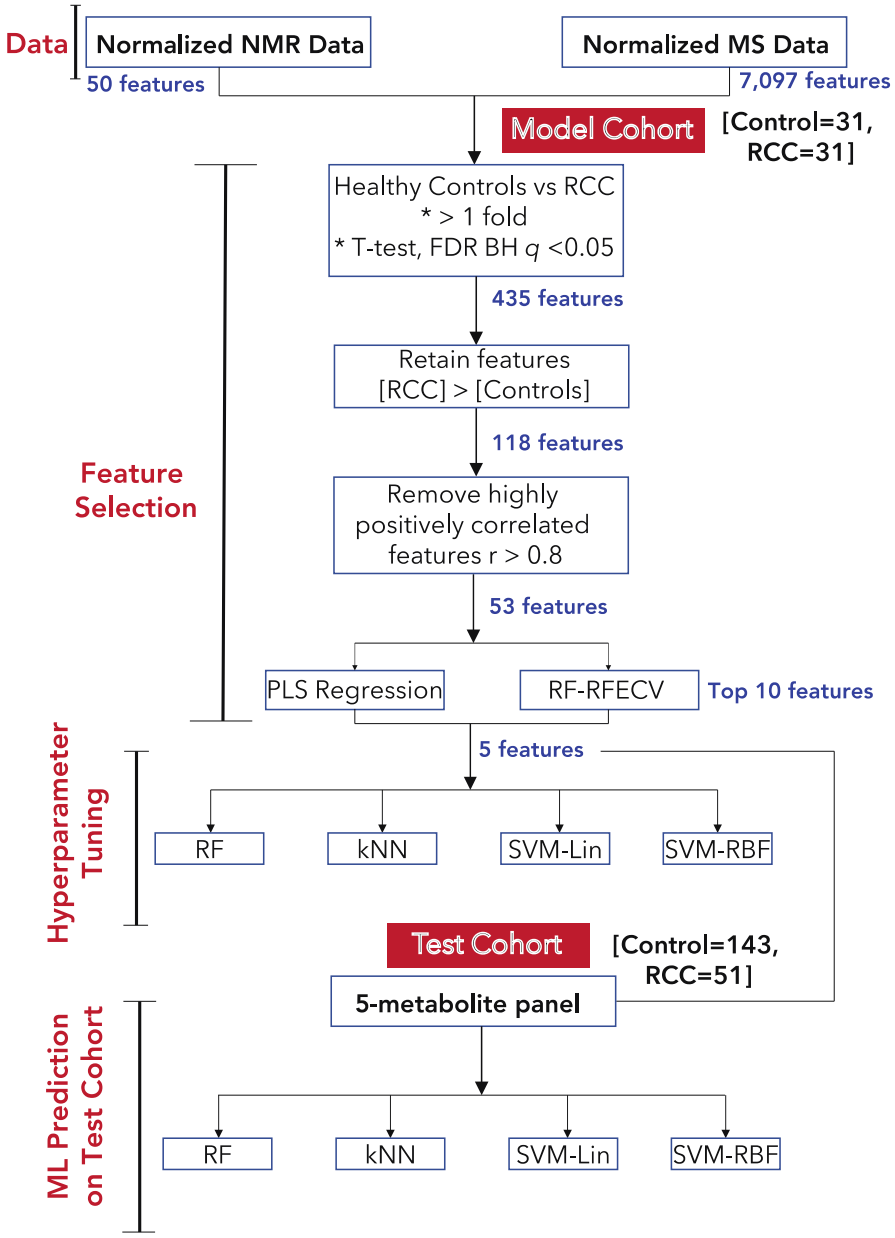



**Figure S4:** Machine learning pipeline focused on upregulated features in RCC *vs*. controls. **PLS:** partial least squares, **RF- RFECV:** random forest recursive feature elimination – cross validation, **FDR-BH:** false discovery rate Benjamini-Hochberg procedure, ***k*-NN:** *k*-nearest neighbors.

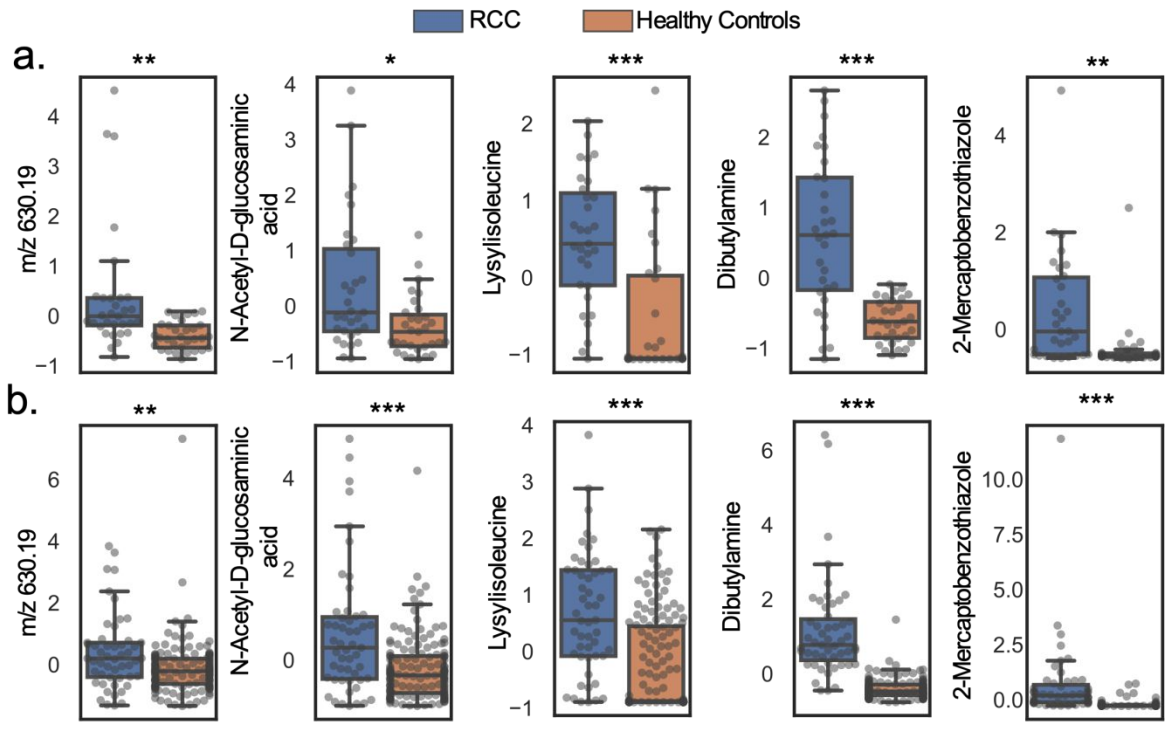



**Figure S5:** Relative abundances for the panel of upregulated metabolites. (a) Model cohort. *q*-values were computed by taking the FDR correction (Benjamini-Hochberg) after an independent *t*-test. ( $* \, q \leq 0.05$, $** \, q \leq 0.01$, $*** \, q \leq 0.001$). (b) In the test cohort, *p*-values from the Welch t-test were reported (unequal sample size). ( $* \, p \leq 0.05$, $** \, p \leq 0.01$, $*** \, p \leq 0.001$). Raw data were transformed *via* autoscaling for visualization

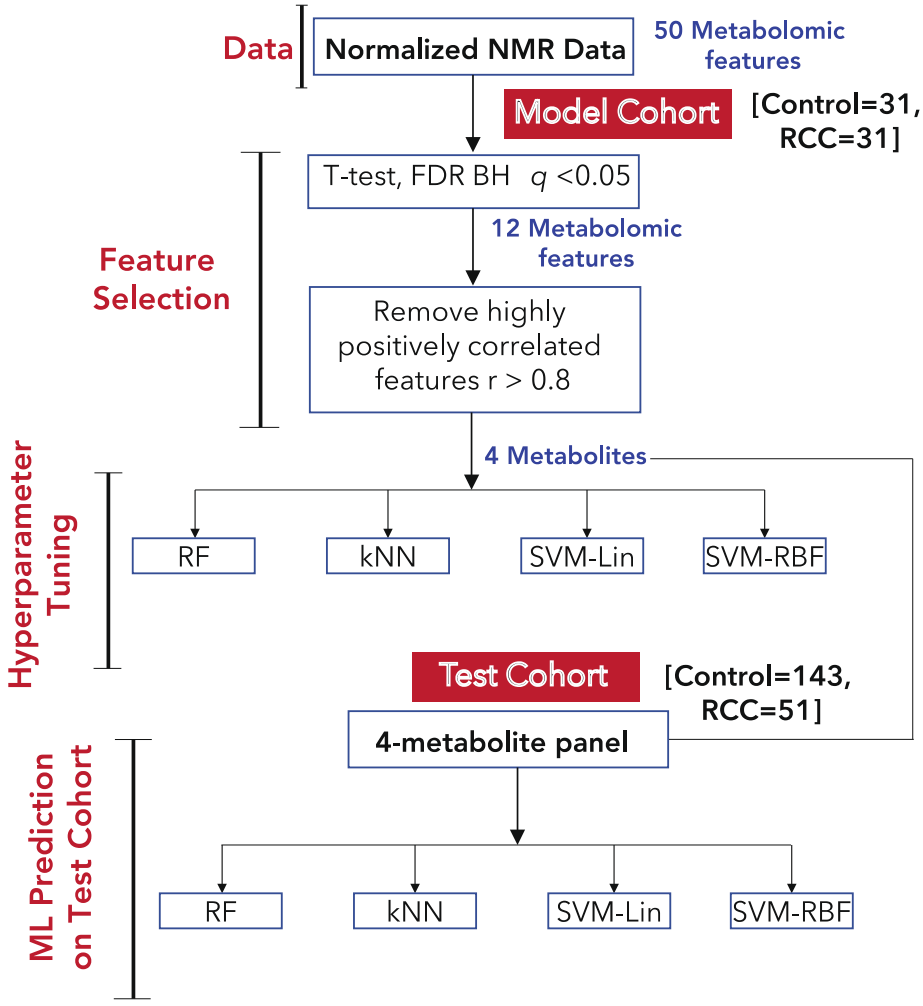



**Figure S6:** Machine learning pipeline focused only on NMR features. Using the model cohort, all NMR features were subjected to feature selection strategies culminating in four selected metabolites (hippurate, trigonellinamide, lactate, and mannitol). Hyperparameters for four different machine learning models were tuned using the model cohort and the 4-metabolite panel. Final predictions were made using the test cohort under cross-validated conditions. **PLS:** partial least squares, **RF- RFECV:** random forest recursive feature elimination – cross validation, **FDR-BH:** false discovery rate Benjamini-Hochberg procedure, ***k*-NN:** *k*-nearest neighbors, **SVM:** support vector machines (Lin: linear, RBF: radial basis function).

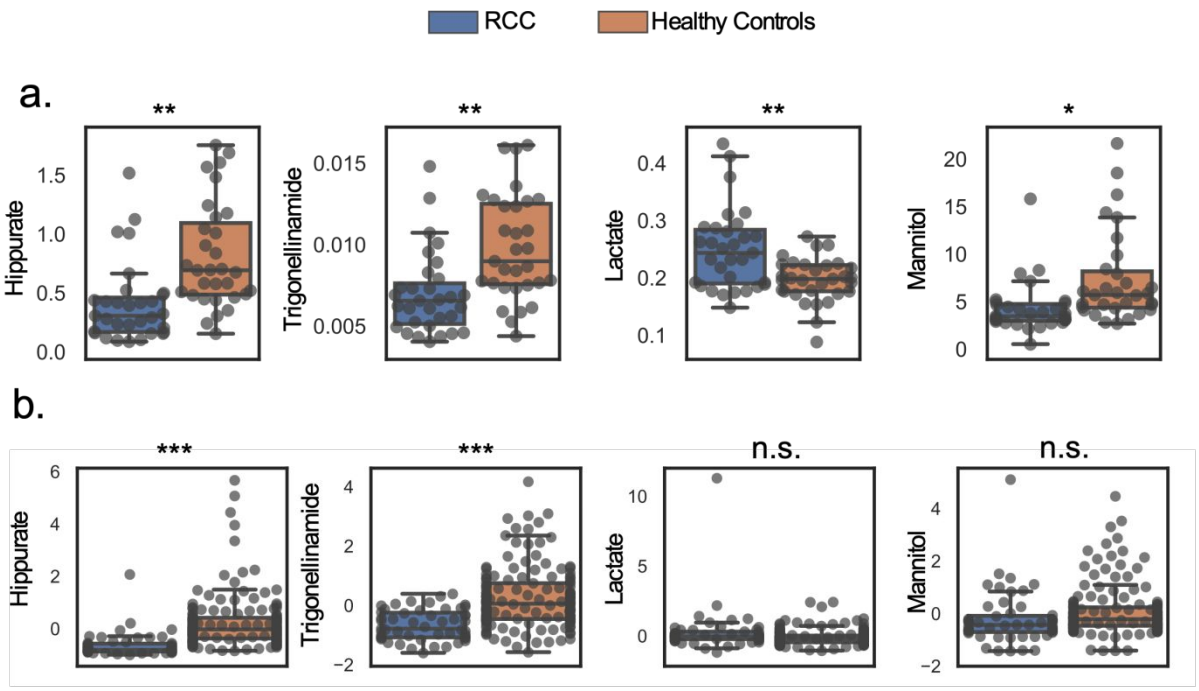



**Figure S7:** Relative quantification of features in the NMR RCC metabolic panel. (a) Model cohort. *q*-values were computed by taking the FDR correction (Benjamini-Hochberg) after an independent *t*-test. ( $* \, q \leq 0.05, \;\; ** \, q \leq 0.01, \;\; *** \, q \leq 0.001$). (b) In the test cohort, *p*-values from the Welch t-test were reported (unequal sample size). (n.s. not significant $* \, p \leq 0.05, \;\; ** \, p \leq 0.01, \;\; *** \, p \leq 0.001$). Raw data were transformed *via* autoscaling for visualization

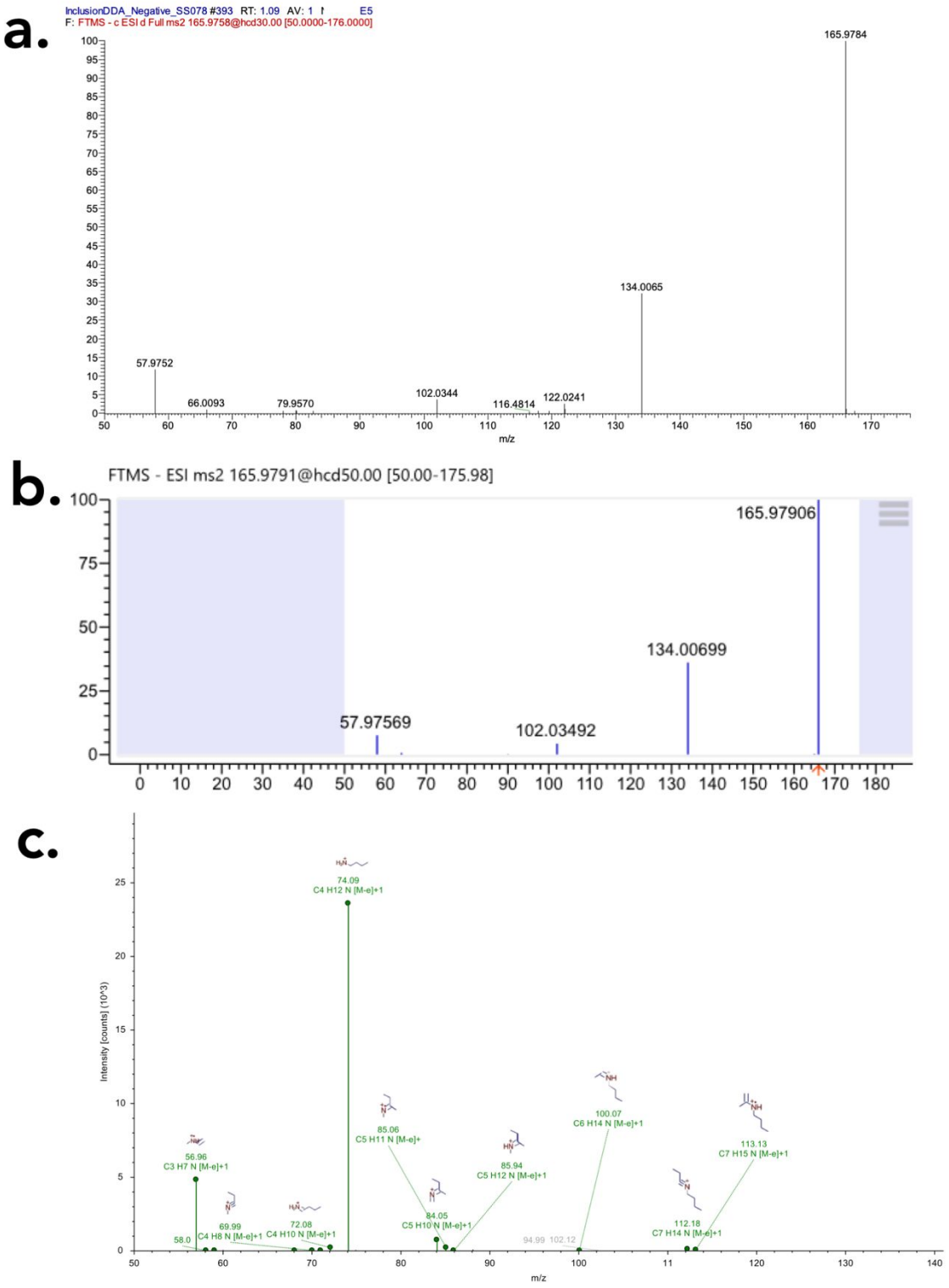



**Figure S8:** MS/MS annotation of 2-mercaptobenzothiazole and dibutylamine/ n-butylisobutylamine/disobutylamine. a) Experimental spectra of 2-mercaptobenzothiazole

b) MS/MS of spectrum of 2-mercaptobenzothiazole from mzCloud database. c) Annotated MS/MS spectrum of feature identified as dibutylamine/n-butylisobutylamine/disobutylamine. Annotations are obtained from in silico fragmentation in Compound Discoverer (Thermo Fisher).

**Table S1:** Propensity score matching and model cohort characteristics**.** *p*-Values were calculated using Welch and Student *t*-test, for unequal and equal sample sizes, respectively. Race: unknown/missing (6), mixed (1), and Asian (1) were all classified as others (8); Smoker: former (46) and current (9) were all classified as former/current (55). **RCC:** Renal Cell Carcinoma.

| | Pre-match Groups | | | Post-match Groups | | |
|---|---|---|---|---|---|---|
| Characteristic | **Controls** | **RCC** | ***p*-Value** | **Controls** | **RCC** | ***p*-Value** |
| No of Urine Samples | 174 | 31 | | 31 | 31 | |
| *Mean Age ± SD* | 54.4 ± 10.3 | 59.5 ± 12.4 | 0.03 | 58.0 ± 13.0 | 59.5 ± 12.1 | 0.64 |
| *BMI* | 27.3 ± 4.5 | 29.1 ± 5.8 | 0.11 | 26.5 ± 4.9 | 29.1 ± 5.8 | 0.06 |
| *Race* | | | | | | |
| **Caucasian** | 162 (93.1%) | 21 (67.7%) | | 25 (80.6%) | 21 (67.7%) | |
| **Black/African American** | 5 (4.0%) | 9 (29.0%) | | 3 (9.7%) | 9 (29.0%) | |
| **Others** | 7 (2.8%) | 1 (3.2%) | | 3 (9.7%) | 1 (3.2%) | |
| *Smoker* | | | | | | |
| **Never** | 131 (75.3%) | 19 (61.3%) | | 17 (54.8%) | 19 (61.3%) | |
| **Former/Current** | 43 (24.7%) | 12 (38.7%) | | 14 (45.2%) | 12 (38.7%) | |
| *Gender* | | | | | | |
| **Male** | 145 (83.3%) | 14 (45.2%) | | 14 (45.2%) | 14 (45.2%) | |
| **Female** | 29 (16.6%) | 17 (54.8%) | | 17 (54.8%) | 17 (54.8%) | |

**Table S2:** Model Cohort RCC Characteristics.

| Characteristic | Frequency | Percentage |
|---|---|---|
| *Metastasis* | | |
| Yes | 7 | 22.6% |
| No | 24 | 77.4% |
| *Histological Subtypes* | | |
| Pure Clear Cell | 23 | 71% |
| Papillary | 3 | 9.7% |
| Chromophobe | 2 | 6.5% |
| Clear Cell Papillary | 2 | 6.5% |
| Unclassified | 2 | 6.5% |
| [a] *Nuclear Grade* | | |
| 1 | 0 | 0% |
| 2 | 10 | 33.3% |
| 3 | 8 | 26.7% |
| 4 | 12 | 40% |
| *T-Stage* | | |
| T1a | 11 | 35.4% |
| T1b | 5 | 16.1% |
| T2a | 2 | 6.4% |
| T2b | 1 | 3.2% |
| T3a | 11 | 35.4% |
| T4 | 1 | 3.2% |
| *M-Stage* | | |
| M0 | 24 | 77.4% |
| M1 | 7 | 22.6% |
| *N-Stage* | | |
| N0 | 25 | 80.6% |
| N1 | 4 | 12.9% |
| NX | 2 | 6.5% |
| [b] *RCC Stage* | | |
| I | 13 | 44.8% |
| II | 3 | 10.3% |
| III | 6 | 20.7% |
| IV | 7 | 24.1% |

[a] One nuclear grade missing. [b] Two individuals cancer stages are not reported due to inconclusive TNM staging.

**Table S3:** Test Cohort Characteristics**.** *p*-Values were calculated using the Welch *t*-test.

| Characteristic | Controls | RCC | *p*-Value |
|---|---|---|---|
| No. of Urine Samples | 143 | 51 | |
| *Mean Age ± SD* | 53.6 ± 9.5 | 61.7 ± 13.7 | 0.0002 |
| *BMI* | 27.5 ± 4.4 | 29.1 ± 5.6 | 0.03 |
| *Race* | | | |
| Caucasian | 136(95.1%) | 35(70.0%) | |
| Black/African American | 5(3.4%) | 11(22.0%) | |
| Others | 2(1.4%) | 4(8.0%) | |
| *Smoker* | | | |
| Never | 113 (79%) | 32 (62.7%) | |
| Former/Current | 30 (21%) | 19 (37.3%) | |
| *Gender* | | | |
| Male | 131 (91.6%) | 31 (62.0%) | |
| Female | 12 (8.4%) | 19 (38.0%) | |
| *Histological Subtypes* | | | |
| Pure Clear Cell | | 35 (68.6%) | |
| Papillary | | 7 (13.7%) | |
| Clear Cell Papillary | | 4 (7.8%) | |
| Chromophobe | | 3 (5.9%) | |
| Unclassified | | 2 (3.9%) | |
| *Metastasis* | | | |
| No | | 41 (80.4%) | |
| Yes | | 10 (19.6%) | |
| [a] *Nuclear Grade* | | | |
| 1 | | 0 (0%) | |
| 2 | | 20 (41.7%) | |
| 3 | | 21 (43.8%) | |
| 4 | | 7 (14.6%) | |
| [b] *RCC Stage* | | | |
| I | | 20 (48.8%) | |
| II | | 5 (12.2%) | |
| III | | 9 (22.0%) | |
| IV | | 7 (17.1%) | |

[a] For nuclear grades, three samples were missing. [b] Ten samples have missing RCC staging because of inconclusive TNM staging.

**Table S4:** Quantified NMR features**.** ppm values, confidence score, fold changes, and *q*-values.

| ***Metabolite/ Features*** | ***$^{1}$H (ppm)*** | ***$^{13}$C(ppm)*** | ***Peak patterns*** | ***Confidence Score*** | ***Fold Change*** | ***q-value*** |
|---|---|---|---|---|---|---|
| *unknown 1* | 0.15 | - | (s) | - | 0.01 | 0.957 |
| *unknown 2* | 0.36 | - | (m) | - | 0.42 | 0.050 |
| *[**]bile acid 1* | 0.53 | - | (s) | 1 | 0.32 | 0.119 |
| *[**]bile acid 2* | 0.56 | - | (s) | 1 | 0.12 | 0.560 |
| *3-hydroxyisovaleric acid* | 1.26 | 30.84 | (s) | 3 | 0.07 | 0.704 |
| *lactate* | 1.31 | 22.97 | (d) | 4 | 0.34 | 0.003 |
| *unknown 3* | 1.85 | - | (s) | - | 0.59 | 0.560 |
| *acetate* | 1.90 | 26.04 | (s) | 3 | 0.57 | 0.196 |
| *acetone* | 2.23 | 32.40 | (s) | 3 | -0.12 | 0.196 |
| *unknown 4* | 2.26 | - | (s) | - | -0.06 | 0.704 |
| *acetoacetate* | 2.27 | 32.19 | (s) | 3 | -0.07 | 0.634 |
| *unknown 5* | 2.33 | - | (s) | - | -0.03 | 0.860 |
| *¨pyruvate* | 2.41 | - | (s) | 2 | 0.05 | 0.560 |
| *citrate* | 2.53 | 48.52 | (d) | 3 | -0.05 | 0.811 |
| *dimethylamine (DMA)* | 2.71 | 37.5 | (s) | 3 | 0.22 | 0.119 |
| *unknown 6* | 2.77 | - | (s) | - | 0.05 | 0.827 |
| *methylguanidine* | 2.82 | 30.21 | (s) | 3 | 0.16 | 0.256 |
| *unknown 7* | 3.08 | - | (t) | - | -0.34 | 0.126 |
| *choline* | 3.19 | 56.69 | (s) | 3 | -0.06 | 0.686 |
| *[a]scyllo-inositol* | 3.35 | 76.4 | (s) | 3 | -1.14 | 0.002 |
| *taurine* | 3.42 | 38.07 | (t) | 4 | -0.03 | 0.811 |
| *acetoacetate* | 3.44 | 56.22 | (s) | 3 | 0.23 | 0.368 |
| *4-hydroxyphenylacetate (4-HPA)* | 3.44 | 46.34 | (s) | 4 | 0.23 | 0.368 |
| *glycine* | 3.56 | 44.18 | (s) | 3 | 0.48 | 0.368 |
| *mannitol* | 3.86 | 65.94 | (d) | 4 | -0.78 | 0.012 |
| *mannitol* | 3.88 | 65.94 | (d) | 4 | -0.68 | 0.022 |
| *creatine* | 3.92 | - | (s) | 3 | 0.07 | 0.811 |
| *[a]glycolate* | 3.94 | 64.32 | (s) | 3 | 0.07 | 0.663 |

| | | | | | | |
|---|---|---|---|---|---|---|
| *hippurate* | 3.96 | 46.46 | (d) | 4 | -0.68 | 0.004 |
| *4-hydroxyhippuric acid* | 3.96 | 46.58 | (d) | 3 | -0.68 | 0.004 |
| *tartrate* | 4.34 | 76.55 | (s) | 3 | 0.13 | 0.728 |
| *unknown 8* | 6.07 | - | (s) | - | 0.09 | 0.415 |
| *unknown 9* | 6.18 | - | (s) | - | 0.59 | 0.267 |
| *fumarate* | 6.52 | - | (s) | 2 | 0.17 | 0.560 |
| *4-hydroxyphenylacetate (4-HPA)* | 7.13 | 133.15 | (d) | 4 | 0.83 | 0.168 |
| *hippurate* | 7.55 | 131.50 | (t) | 4 | -0.98 | 0.002 |
| *hippurate* | 7.65 | 134.92 | (m) | 4 | -0.96 | 0.002 |
| [a]*4-aminohippuric acid* | 7.67 | 133.02 | (d) | 3 | -1.64 | 0.002 |
| *indoxyl sulfate (IS)* | 7.70 | 127.07 | (d) | 3 | 0.18 | 0.492 |
| *hippurate* | 7.83 | 129.85 | (dd) | 4 | -0.91 | 0.003 |
| *hypoxanthine* | 8.18 | 148.27 | (s) | 3 | 0.21 | 0.811 |
| *hypoxanthine* | 8.20 | 144.75 | (s) | 3 | 0.86 | 0.368 |
| *formate* | 8.45 | 173.71 | (s) | 3 | 0.22 | 0.488 |
| *unknown 10* | 8.77 | - | (d) | - | 0.19 | 0.791 |
| *trigonelline* | 8.83 | 147.46 | (t) | 3 | -0.2 | 0.686 |
| *trigonellinamide* | 8.89 | - | (d) | 2 | -0.49 | 0.002 |
| *trigonellinamide* | 8.97 | - | (d) | 2 | -0.49 | 0.002 |
| *trigonelline* | 9.11 | 148.50 | (s) | 3 | -0.33 | 0.524 |
| *trigonellinamide* | 9.27 | - | (s) | 2 | -0.51 | 0.009 |
| *unknown 11* | 9.36 | - | (s) | - | -0.14 | 0.686 |

[a] Quantification may be unreliable because of spectral overlaps. Tentative assignment (Monteiro *et al* 2016[1]) s=singlet, d=doublet, dd=doublet of doublet, m=multiplet. Fold change (FC) was calculated as the base 2 logarithm of the average integral ratios between RCC and controls samples. Positive FC values indicate increased abundance in RCC, while negative values indicate higher abundance in control samples. *q*-values were computed by taking the FDR correction (Benjamini-Hochberg) after an independent *t*-test.

**Table S5:** Chemical information for the 10-metabolite panel.

| *ID no.* | *Retention Time (min)* | *m/z* | | *Adduct Type* | *Mass error (ppm)* | *Elemental Formula* | *Metabolite Identity* |
|---|---|---|---|---|---|---|---|
| | | **Theoretical** | **Experimental** | | | | |
| *720* | 5.68 | 136.0757 | 136.0755 | $[M+H]^+$ | -1.47 | $C_8H_9NO$ | 2-phenylacetamide |
| *1481* | 8.83 | 260.1969 | 260.1969 | $[M+H]^+$ | 0.00 | $C_{12}H_{25}N_3O_3$ | Lys-Ile |
| *2102* | 4.39 | 130.1590 | 130.1591 | $[M+H]^+$ | 0.77 | $C_8H_{19}N$ | dibutylamine, N-butylisobutylamine, diisobutylamine (isomer) |
| *3141* | 2.27 | 343.1135 | 343.1134 | $[M+H]^+$ | -0.20 | $C_{14}H_{18}N_2O_8$ | -- |
| *3675* | 1.18 | -- | 87.0641 | $[M+H]^+$ | -- | -- | -- |
| *3804* | 2.59 | 202.0474 | 202.0478 | $[M+H]^+$ | 1.70 | $C_4H_{12}NO_6P$ | hippuric acid |
| *3872* | 4.05 | 973.6038 | 973.6027 | $[M+2H]^{2+}$ | -1.13 | $C_{100}H_{158}N_{19}O_{20}$ | -- |
| *4080* | 0.82 | 406.0597 | 406.0594 | $[M+H]^+$ | -0.78 | $C_{10}H_{21}N_3O_8P_2S$ | |
| *6261* | 2.59 | 314.1248 | 314.1244 | $[M-H]^-$ | -1.27 | $C_9H_{18}N_9O_2P$ | -- |
| *6262* | 2.67 | 376.1249, 358.1143 | 376.1246, 358.1147 | $[M+H_2O-H]^-$ [M-H] | -0.68 | $C_{15}H_{21}NO_9$ | hippurate-mannitol derivative |

**Table S6:** Machine learning hyperparameters used for binary classification using the MS-based 10-metabolite panel.

| Parameters | Initial distribution | Optimized |
|---|---|---|
| **Random Forest** | | |
| *Max_depth* | 10, 20, 30 | 10 |
| *Max_features* | 'auto', 'sqrt', 'log2' | 'auto' |
| *Min_samples_leaf* | 1, 2, 3, 4, 5 | 1 |
| *Min_samples_split* | 2, 4, 6, 8 | 2 |
| *N_estimators* | 50, 100, 150, 200 | 100 |
| **SVM-RBF** | | |
| *C* | 0.1, 1, 10, 100 | 10 |
| *gamma* | 0.01, 0.03, 0.1, 0.3, 1.0 | 0.1 |
| **Lin-SVM** | | |
| *C* | 0.001, 0.01, 0.1, 1, 5, 10 | 0.1 |
| **k-NN** | | |
| *Number of neighbors* | 2 - 30 | 4 |
| *Distance Measure* | Manhattan, Euclidean | Manhattan |

**Table S7:** Machine learning performance using the MS RCC 10 panel biomarker.

| *Algorithm* | *RF* | *K-NN* | *SVM-RBF* | *Linear SVM* |
|---|---|---|---|---|
| *AUC* | 1.0 +/- 0.0 (0.95) | 0.96 +/- 0.04 (**0.96**) | 0.99 +/- 0.01 (0.94) | 1.0 +/- 0.0 (0.97) |
| *Accuracy* | 0.95 +/- 0.04 (80%) | 0.95 +/- 0.07 (**87%**) | 0.93 +/- 0.06 (82%) | 0.95 +/- 0.07 (81%) |
| *Sensitivity* | 0.94 +/- 0.08 (100%) | 0.93 +/- 0.13 (**96%**) | 0.93 +/- 0.08 (84%) | 0.97 +/- 0.07 (100%) |
| *Specificity* | 0.97 +/- 0.07 (73%) | 0.97 +/- 0.07 (**83%**) | 0.93 +/- 0.08 (81%) | 0.93 +/- 0.13 (75%) |

**Table S8:** Compound annotation and identification for the panel of 5-metabolites upregulated in RCC.

| *ID no.* | *Retention Time (min)* | *m/z* | | *Adduct Type* | *Mass error (ppm)* | *Elemental Formula* | *Metabolite Identity* |
|---|---|---|---|---|---|---|---|
| | | *Theoretical* | *Experimental* | | | | |
| *1481* | 8.83 | 260.1969 | 260.1969 | $[M+H]^+$ | 0.00 | $C_{12}H_{25}N_3O_3$ | Lys-Ile |
| *2102* | 4.39 | 130.1590 | 130.1591 | $[M+H]^+$ | 0.77 | $C_8H_{19}N$ | dibutylamine, N-butylisobutylamine, diisobutylamine (isomer) |
| *6578* | 1.09 | 165.9790 | 165.9784 | $[M-H]^-$ | -3.61 | $C_7H_5NS_2$ | 2-mercaptobenzothiazole |
| *6594* | 6.89 | 236.0776 | 236.0777 | $[M+H]^+$ | 0.42 | $C_8H_{15}NO_7$ | N-acetyl-glucosaminic acid |
| *5698* | 3.38 | 630.1909 | 630.1895 | $[M-H]^-$ | -2.64 | $C_{24}H_{43}NO_{12}P_2S$ | -- |

**Table S9:** Hyperparameters tuned for machine learning methods used for binary classification for the upregulated RCC biomarkers.

| Parameters | Initial distribution | Optimized |
|---|---|---|
| **Random Forest** | | |
| *Max_depth* | 10, 20, 30 | 10 |
| *Max_features* | 'auto', 'sqrt', 'log2' | ‘auto’ |
| *Min_samples_leaf* | 1, 2, 3, 4, 5 | 1 |
| *Min_samples_split* | 2, 4, 6, 8 | 2 |
| *N_estimators* | 50, 100, 150, 200 | 150 |
| **SVM-RBF** | | |
| *C* | 0.1, 1, 10, 100 | 10 |
| *gamma* | 0.01, 0.03, 0.1, 0.3, 1.0 | 0.3 |
| **Lin-SVM** | | |
| *C* | 0.001, 0.01, 0.1, 1, 5, 10 | 10 |
| **k-NN** | | |
| *Number of neighbors* | 2 - 30 | 11 |
| *Distance Measure* | Manhattan, Euclidean | Euclidean |

**Table S10:** Machine learning performance using the upregulated MS RCC biomarkers.

| | MS Upregulated Biomarkers | | | |
|---|---|---|---|---|
| **Algorithm** | RF | K-NN | SVM-RBF | Linear SVM |
| **AUC** | 0.95 +/- 0.06 (0.97) | 0.92 +/- 0.1 (**0.92**) | 0.9 +/- 0.08 (0.80) | 0.9 +/- 0.08 (0.91) |
| **Accuracy** | 0.87 +/- 0.04 (70%) | 0.77 +/- 0.14 (**81%**) | 0.89 +/- 0.1 (61%) | 0.82 +/- 0.07 (71%) |
| **Sensitivity** | 0.87 +/- 0.12 (98%) | 0.7 +/- 0.27 (**86%**) | 0.87 +/- 0.16 (96%) | 0.8 +/- 0.12 (94%) |
| **Specificity** | 0.87 +/- 0.12 (59%) | 0.83 +/- 0.18 (**79%**) | 0.9 +/- 0.13 (49%) | 0.83 +/- 0.11 (62%) |

**Table S11:** Detailed MS/MS information for the 7-metabolite panel that distinguishes RCC from control samples. The m/z of fragment ions were obtained from DDA experiments. The corresponding collision energy (CE) is also listed in the table. Selected precursor ions are underlined. Fragment ions that matched to literature spectra or were consistent with potential structures are in bold. Metabolite identification level was assigned based on the following criteria: 1) exact mass, isotopic pattern, retention time, and MS/MS spectrum of standard matched to the feature. 2) exact mass, isotopic pattern, and MS/MS spectrum matched with literature spectra or fragmentation ions observed are consistent with the proposed structure. 3) tentative ID assignment based on elemental formula matches with literature. 4) unknowns**.**

| Feature ID no. | CE (eV), Mode | Fragment ion m/z | Metabolite identification level | Match details | Metabolite Name |
|---|---|---|---|---|---|
| 720 | 10,30,45 (+) | **136.0755**, **119.0489, 118.0648**, 107.0490, **91.0541,** 95.0602, 101.2184, 87.466, 70.9133, **65.0383** | 2 | Fragmentation consistent with spectrum (HMDB) | 2-phenylacetamide |
| 1481 | 10,30,45 (+) | 260.1977**,** 171.1495, 144.1021, 100.0757, 84.0808**,** 72.0444, 65.8753, 54.7500 | 2 | Fragmentation consistent with structure | lys-ile or lys-leu |
| 2102 | 10,30,45 (+) | 130.1592, 84.0445, 74.0965, 57.0700 | 2 | Fragmentation consistent with structure | dibutylamine (alkyl chain branching not determined) |
| 3804 | --- | **180.0880**, **105.0339, 162.0771**, 95.0497, 53.0395, 110.0345,120.0812, 138.0556 | 2 | Fragmentation consistent with spectrum (m/z cloud) | hippuric acid |
| 6262 | 10,30,45 (-) | 342.2521, 310.2751, 280.0701, **194.0449,** 181.0375**, 150.0594**, **148.0392**, 138.0297, **121.0285**, **124.0061**, **113.0230**, **93.0332, 85.0281, 89.0240, 71.0134, 73.0290** | 2 | Fragmentation consistent with spectrum (m/z cloud) | hippurate-mannitol derivative |
| 6578 | 10,30,45 (-) | **165.9784**, **134.0065**, 122.01241, 117.9193, **102.0344**, 79.9570, 66.0093, **57.9752** | 2 | Fragmentation consistent with spectrum (m/z cloud) | 2-mercaptobenzothiazole |

| | | | | | |
|---|---|---|---|---|---|
| 6594 | 10,30,45 (+) | 236.0773, 230.1947, 208.9668, 149.0452, 131.0346, 119.0349, 113.0241, 104.0350, 98.9556, 89.0240, 86.0244, 85.0291, 71.0134, 59.0134, | 2 | Fragmentation consistent with structure | N-acetyl-glucosaminic acid |

**Table S12:** Machine learning hyperparameters tuned for binary classification using only the 7-identified metabolite panel.

| Parameters | Initial distribution | Optimized |
|---|---|---|
| **Random Forest** | | |
| *Max_depth* | 10, 20, 30 | 10 |
| *Max_features* | 'auto', 'sqrt', 'log2' | 'auto' |
| *Min_samples_leaf* | 1, 2, 3, 4, 5 | 5 |
| *Min_samples_split* | 2, 4, 6, 8 | 2 |
| *N_estimators* | 50, 100, 150, 200 | 50 |
| **SVM-RBF** | | |
| *C* | 0.1, 1, 10, 100 | 1 |
| *gamma* | 0.01, 0.03, 0.1, 0.3, 1.0 | 0.03 |
| **Lin-SVM** | | |
| *C* | 0.001, 0.01, 0.1, 1, 5, 10 | 0.1 |
| **k-NN** | | |
| *Number of neighbors* | 2 - 30 | 7 |
| *Distance Measure* | Manhattan, Euclidean | Manhattan |

**Table S13:** Machine learning hyperparameters tuned for binary classification using only NMR RCC biomarkers.

| Parameters | Initial distribution | Optimized |
|---|---|---|
| **Random Forest** | | |
| *Max_depth* | 10, 20, 30 | 10 |
| *Max_features* | 'auto', 'sqrt', 'log2' | ‘auto’ |
| *Min_samples_leaf* | 1, 2, 3, 4, 5 | 3 |
| *Min_samples_split* | 2, 4, 6, 8 | 2 |
| *N_estimators* | 50, 100, 150, 200 | 50 |
| **SVM-RBF** | | |
| *C* | 0.1, 1, 10, 100 | 10 |
| *gamma* | 0.01, 0.03, 0.1, 0.3, 1.0 | 0.3 |
| **Lin-SVM** | | |
| *C* | 0.001, 0.01, 0.1, 1, 5, 10 | 1 |
| **k-NN** | | |
| *Number of neighbors* | 2 - 30 | 5 |
| *Distance Measure* | Manhattan, Euclidean | Manhattan |

**Table S14:** Machine learning performance using only NMR RCC biomarkers.

| | **NMR Biomarkers** | | | |
|---|---|---|---|---|
| **Algorithm** | *RF* | *K-NN* | *SVM-RBF* | *Linear SVM* |
| **AUC** | 0.95 +/- 0.03 (0.89) | 0.94 +/- 0.05 (0.88) | 0.94 +/- 0.06 (**0.89**) | 0.89 +/- 0.08 (0.87) |
| **Accuracy** | 0.84 +/- 0.05 (0.76) | 0.87 +/- 0.1 (0.77) | 0.86 +/- 0.06 (**0.78**) | 0.81 +/- 0.08 (0.74) |
| **Sensitivity** | 0.81 +/- 0.12 (0.86) | 0.87 +/- 0.06 (0.84) | 0.9 +/- 0.08 (**0.86**) | 0.83 +/- 0.11 (0.84) |
| **Specificity** | 0.87 +/- 0.19 (0.72) | 0.87 +/- 0.19 (0.75) | 0.81 +/- 0.12 (**0.76**) | 0.77 +/- 0.23 (0.71) |

**Table S15:** Review of some notable urine metabolomics studies comparing RCC to controls.

| ***Study*** | ***Sample Size*** | ***Race/Location of Sample Collection*** | ***Platform*** | ***Biomarker (Increase in RCC)*** | ***Biomarker (Decrease in RCC)*** |
|---|---|---|---|---|---|
| *Kind et al 2007*[2] | 6C, 6N | Tennessee, US | HILIC-LC-MS, GC-TOF-MS, RP-UHPLC-MS | No compound identification | No compound identification |
| *Kim et al 2009*[3] | 11C*, 15N | Texas, US California, US | HILIC LC-MS | No compound identification | |
| *Kim et al 2011*[4] | 29C, 33N | California, US | UHPLC-MS, GC-MS | Quinolinate, alpha-ketoglutarate | Gentisate |
| *Monteiro, M. et al 2016*[5] | 42C, 49N | Portugal | NMR | 2-ketoglutarate, N-methyl-2-pyridone-5-carboxamide, bile acids, galactose, pyruvate, succinate and valine | 4-hydroxyhippurate, 4-hydroxyphenylacetate, acetone, GAA, glycine, hippurate, malonate, phenylacetylglutamine, tartrate, trigonelline |
| *Ragone et al 2016*[6] | 40C*, 29N | Italy | NMR | Creatine, alanine, lactate and pyruvate | Hippurate, citrate, and betaine |
| *Monteiro, M. et al 2017*[7] | 30C, 37N | Portugal | GC-MS | 2-oxopropanal | 2,5,8-trimethyl-1,2,3,4-tetrahydronaphthalene-1-ol |
| *Niziol et al 2018*[8] | 7C*, 15N | Poland | LC-HRMS | Hydroxybutyrylcarnitine, decanoylcarnitine, propanoylcarnitine, carnitine, dodecanoylcarnitine, and norepinephrine sulfate. | riboflavin, acetylaspartylglutamate |
| *Liu et al 2019*[9] | 100C, 129N | China (all Chinese subjects) | LC-MS | N-Jasmonoyltyrosine, Androstenedione, Dopamine 4-sulfate, 3-Methylazelaic acid, 7alpha-hydroxy-3-oxochol-4-en-24-oic acid, Lithocholyltaurine, 11-Dodecenoic acid | Tetrahydroaldosterone-3-glucuronide, Cortolone-3-glucuronide |
| *Wang et al 2019*[10] | 117 C**, 98N | China | UPLC-MS | α-CEHC, flunisolide, glycerol tripropanoate | β-cortolone, deoxyinosine, 11b,17a,21-trihydroxypregnenolone |
| *Zhang et al 2020*[11] | 39C, 68N | China | LC-MS | – | Aminoadipic acid, 2-(formamido)-N1-(5-phosphod-ribosyl) acetamidine and alpha-N-phenylacetyl-l-glutamine |

Studies comparing benign renal tumor and RCC were not included in this review. In Monteiro, M. *et al* 2016[5], hypoxanthine and isoleucine are potentially confounded. In Monteiro, M. *et al* 2017[7], the biomarkers were selected from the 21 initial VOCs, using two independent small samples sets. In Niziol *et al* 2018[8], the biomarkers in urine were also present in tissue. In Wang *et al* 2019[10], bladder cancer patients are confounders; the biomarkers identified were validated in an external cohort consisting of 30 RCC and 44 controls; and in addition, only early stage RCC patients (T1 and T2 stages) were considered. C* = clear cell renal cell carcinoma. C = renal cell carcinoma, N=healthy controls, C** = 53 bladder cancer patients and 64 RCC. HILIC-LC-MS: Hydrophilic Interaction Chromatography Liquid Chromatography Mass Spectrometry, GC-TOF-MS: Gas Chromatography Time-Of-Flight Mass Spectrometry, RP-UHPLC-MS: Reverse Phase Ultra High-Performance Liquid Chromatography Mass Spectrometry, LC-HRMS: Liquid Chromatography-High Resolution Mass Spectrometry, VOC: Volatile organic compounds.

**Table S16:** Metabolomic features with *q*-values < 0.05 and > 1-fold change in the model cohort**.** (Fold change (FC) was calculated as the base 2 logarithm of the average intensity ratios between RCC and controls samples). Positive FC values indicate increased abundance in RCC, while negative values indicate higher abundance in control samples. *q*-values were computed by taking the FDR correction (Benjamini-Hochberg) after an independent *t*-test. 433 features from LC-MS are included in the table, and 2 features from NMR (*scyllo*-Inositol and aminohippurate) are in Table S4.

| ID | Metabolite ID | Mode | RT [min] | FC | FDR p-value |
|---|---|---|---|---|---|
| 1 | 1 | positive | 2.317 | -1.4885 | 0.045959 |
| 9 | 5-acetylamino-6-amino-3-methyluracil | positive | 1.901 | -1.22771 | 0.008743 |
| 95 | 3-(1H-1,2,4-Triazol-3-yl)alanine | positive | 1.898 | -1.23304 | 0.005531 |
| 147 | 147 | positive | 2.58 | -1.20166 | 0.007443 |
| 163 | venlafaxine | positive | 2.654 | -1.27957 | 0.018961 |
| 170 | 170 | positive | 2.313 | -1.56798 | 0.017014 |
| 173 | 173 | positive | 2.605 | -1.23927 | 0.004285 |
| 245 | 245 | positive | 4 | -2.33614 | 0.00089 |
| 250 | 250 | positive | 2.581 | -2.11065 | 0.009913 |
| 260 | leupeptin | positive | 3.989 | -2.30823 | 0.000917 |
| 278 | 278 | positive | 3.057 | -1.65576 | 0.002262 |
| 293 | 293 | positive | 2.617 | -1.64467 | 0.020423 |
| 312 | 312 | positive | 4.523 | 1.113604 | 0.022578 |
| 314 | 314 | positive | 2.592 | -2.15533 | 0.012163 |
| 332 | N-(3-amino-4-methyl-5-nitrophenyl)acetamide | positive | 1.591 | -2.21385 | 0.035889 |
| 347 | 6-methylquinoline | positive | 2.322 | -1.64636 | 0.008933 |
| 363 | 363 | positive | 1.903 | -1.38184 | 0.003166 |
| 429 | 429 | positive | 4.525 | 1.279205 | 0.018937 |
| 435 | paraxanthine | positive | 1.12 | -1.17746 | 0.006372 |
| 474 | 474 | positive | 1.917 | -1.29632 | 0.009562 |
| 479 | phenylacetaldehyde | positive | 2.404 | -1.95368 | 0.011539 |
| 523 | 523 | positive | 2.183 | -1.06828 | 0.017172 |
| 562 | 6-methylquinoline | positive | 2.594 | -1.95682 | 0.010866 |
| 595 | 595 | positive | 2.67 | -2.75395 | 0.0418 |
| 610 | 610 | positive | 2.952 | -1.95838 | 0.005316 |
| 640 | 1_5-anhydro-mannitol | positive | 1.895 | -1.22667 | 0.00129 |
| 643 | 643 | positive | 2.869 | -2.0903 | 0.03379 |

| | | | | | |
|---|---|---|---|---|---|
| 666 | 666 | positive | 2.961 | -1.67651 | 0.009093 |
| 672 | moxaverine | positive | 2.66 | -1.15696 | 0.039852 |
| 688 | 688 | positive | 4.009 | 1.099602 | 0.023352 |
| 712 | 3-dehydrocarnitine | positive | 1.449 | -1.19664 | 0.02724 |
| 720 | 2-phenylacetamide | positive | 2.562 | -1.06888 | 0.000386 |
| 726 | 726 | positive | 6.009 | 1.043296 | 0.04006 |
| 790 | 2-acetolactate | positive | 1.914 | -1.03801 | 0.001699 |
| 798 | indole;1-benzazole | positive | 2.653 | -1.46462 | 0.000526 |
| 800 | 800 | positive | 4.125 | 3.41514 | 0.033921 |
| 819 | 4-dehydropantoate | positive | 1.896 | -1.19163 | 0.00116 |
| 825 | 3-methyldioxyindole | positive | 1.942 | -1.20393 | 0.002882 |
| 880 | 880 | positive | 4.528 | 1.182288 | 0.032715 |
| 900 | 900 | positive | 4.532 | 1.172157 | 0.02614 |
| 926 | 926 | positive | 2.25 | -1.29961 | 0.015545 |
| 954 | 954 | positive | 4.019 | -2.6166 | 0.000917 |
| 958 | 958 | positive | 0.579 | -2.49872 | 0.032106 |
| 960 | 960 | positive | 0.58 | -2.4511 | 0.034206 |
| 995 | 995 | positive | 0.882 | -1.5801 | 0.035476 |
| 1035 | zeatin | positive | 3.628 | 1.022119 | 0.039696 |
| 1047 | 1047 | positive | 2.631 | -2.17057 | 0.012641 |
| 1066 | 1066 | positive | 2.591 | -2.28759 | 0.005362 |
| 1098 | 4-imidazolone-5-propanoate | positive | 3.729 | -1.0197 | 0.027361 |
| 1153 | 1153 | positive | 4.533 | 1.222509 | 0.017014 |
| 1163 | 1163 | positive | 2.45 | -1.39804 | 0.00089 |
| 1214 | 1214 | positive | 1.242 | -1.57143 | 0.020423 |
| 1261 | 1261 | positive | 1.855 | 1.046534 | 0.04275 |
| 1262 | 1262 | positive | 1.9 | -1.49775 | 0.003903 |
| 1307 | 1307 | positive | 1.147 | -5.46823 | 0.001452 |
| 1356 | delta-guanidinovalericacid | positive | 3.929 | -1.93217 | 0.043481 |
| 1365 | 1365 | positive | 4.524 | 1.098536 | 0.009093 |
| 1391 | N- glucosylarylamine | positive | 3.97 | 1.803794 | 0.04275 |
| 1481 | lys-Ile | positive | 6.29 | 1.447371 | 0.001342 |
| 1550 | 1550 | positive | 2.611 | -2.04338 | 0.012124 |
| 1566 | 1566 | positive | 3.909 | -1.29952 | 0.035716 |
| 1579 | 1579 | positive | 5.937 | 1.358104 | 0.041545 |
| 1587 | 1587 | positive | 4.509 | 1.376935 | 0.024 |
| 1662 | 1662 | positive | 2.502 | 2.944322 | 0.047485 |
| 1673 | 1673 | positive | 0.804 | -3.74992 | 0.001296 |
| 1676 | 1676 | positive | 2.648 | -1.80767 | 0.01703 |
| 1689 | 1689 | positive | 4.715 | 4.292918 | 0.017731 |
| 1691 | 1691 | positive | 3.418 | -1.91163 | 0.040973 |

| | | | | | |
|---|---|---|---|---|---|
| 1701 | 4-(2-aminoethyl)benzenesulfonylfluoride | positive | 1.807 | -1.93725 | 0.000526 |
| 1723 | 1723 | positive | 2.574 | -1.7161 | 0.00597 |
| 1741 | PEG monolaurate n5 | positive | 3.738 | -2.74589 | 0.001453 |
| 1760 | 4-(2-aminoethyl)benzenesulfonylfluoride | positive | 1.704 | -2.14754 | 0.000826 |
| 1771 | 1771 | positive | 3.001 | 1.262088 | 0.030398 |
| 1776 | N-acetylleucylleucine | positive | 2.587 | -1.89519 | 0.021927 |
| 1820 | 1820 | positive | 1.953 | -2.10726 | 0.004044 |
| 1839 | 2_3-diaminopropionicacid | positive | 2.6 | -2.15507 | 0.008703 |
| 1858 | 1858 | positive | 3.883 | -3.72213 | 0.006498 |
| 1861 | 1861 | positive | 4.81 | -2.79138 | 0.019817 |
| 1870 | 1870 | positive | 3.824 | -3.63177 | 0.010482 |
| 1880 | 1880 | positive | 4.528 | 1.332245 | 0.017731 |
| 1931 | anisole | positive | 2.618 | -1.12591 | 0.009093 |
| 1942 | 1942 | positive | 3.603 | -1.29462 | 0.049502 |
| 1961 | 1961 | positive | 2.434 | -1.87936 | 0.000515 |
| 1969 | 1969 | positive | 2.583 | -1.36431 | 0.009913 |
| 1991 | 1991 | positive | 3.247 | -3.24599 | 0.015568 |
| 2005 | 2005 | positive | 4.876 | -2.9057 | 0.016077 |
| 2056 | 2056 | positive | 2.649 | -1.55177 | 0.008562 |
| 2082 | 2082 | positive | 2.594 | -1.30732 | 0.008588 |
| 2102 | dibutylamine, N-butylisobutylamine, diisobutylamine (isomer) | positive | 3.449 | 1.567367 | 7.87E-05 |
| 2138 | 2138 | positive | 1.12 | -1.15134 | 0.014564 |
| 2158 | 2158 | positive | 4.528 | 1.256293 | 0.021194 |
| 2196 | 2196 | positive | 0.814 | -2.89012 | 0.00089 |
| 2207 | 2207 | positive | 1.097 | -3.82236 | 0.003166 |
| 2218 | 2218 | positive | 2.432 | -1.70991 | 0.000586 |
| 2230 | 2230 | positive | 2.59 | -2.19533 | 0.006379 |
| 2239 | 2239 | positive | 1.101 | -3.85416 | 0.002882 |
| 2240 | 2240 | positive | 2.886 | 1.631405 | 0.00451 |
| 2241 | 2241 | positive | 4.838 | -1.06192 | 0.015545 |
| 2242 | 2242 | positive | 2.587 | -2.31188 | 0.006372 |
| 2259 | 2259 | positive | 3.706 | -2.85038 | 0.032369 |
| 2267 | beta-Ionone | positive | 2.371 | -2.11197 | 0.011891 |
| 2306 | 2306 | positive | 2.623 | -1.92523 | 0.009819 |
| 2320 | 2320 | positive | 0.866 | -2.38259 | 0.000912 |
| 2321 | 2321 | positive | 1.138 | -3.78215 | 0.001432 |
| 2348 | 2348 | positive | 3.89 | -1.05941 | 0.02614 |
| 2353 | leucinamide | positive | 1.118 | -3.10452 | 0.003468 |
| 2359 | 2359 | positive | 6.637 | 1.794645 | 0.023123 |
| 2385 | 2385 | positive | 4.513 | 1.09136 | 0.01182 |

| | | | | | |
|---|---|---|---|---|---|
| 2403 | 2403 | positive | 4.53 | 1.352297 | 0.012999 |
| 2418 | 2418 | positive | 4.477 | -3.95489 | 0.027524 |
| 2426 | 2426 | positive | 2.851 | 1.572021 | 0.035289 |
| 2448 | 2448 | positive | 4.712 | 6.772872 | 0.024075 |
| 2455 | 2455 | positive | 1.14 | -1.32081 | 0.00129 |
| 2462 | 2462 | positive | 4.536 | 1.262401 | 0.005734 |
| 2476 | 2476 | positive | 4.534 | 1.481945 | 0.027785 |
| 2496 | 2496 | positive | 2.581 | -2.27665 | 0.004492 |
| 2499 | 2499 | positive | 4.508 | 1.286938 | 0.027563 |
| 2524 | 2524 | positive | 1.299 | 1.044756 | 0.048626 |
| 2540 | 2540 | positive | 4.732 | 7.106785 | 0.020956 |
| 2568 | 2568 | positive | 3.711 | -1.24199 | 0.013193 |
| 2571 | 2571 | positive | 3.122 | -1.9284 | 0.035889 |
| 2577 | pipecolicacid | positive | 1.457 | -1.52805 | 0.013548 |
| 2601 | 2601 | positive | 2.658 | -2.07536 | 0.009562 |
| 2621 | 2621 | positive | 3.989 | -2.63727 | 0.001453 |
| 2625 | 2625 | positive | 1.869 | -1.023 | 0.022102 |
| 2652 | 2652 | positive | 4.544 | 1.009831 | 0.04275 |
| 2653 | 2653 | positive | 2.602 | -1.53429 | 0.016708 |
| 2668 | cyclo(leucylprolyl) | positive | 1.067 | -1.04975 | 0.048489 |
| 2702 | 2702 | positive | 4.518 | 1.167157 | 0.018248 |
| 2709 | N-acetyl- glucosaminate | positive | 3.857 | 1.090917 | 0.037911 |
| 2731 | 2731 | positive | 4.714 | 4.920091 | 0.007029 |
| 2732 | 2732 | positive | 4.653 | 4.191832 | 0.017527 |
| 2749 | 2_3-dimethylmalate | positive | 2.894 | 1.338155 | 0.043239 |
| 2803 | 2803 | positive | 4.539 | 1.220539 | 0.005355 |
| 2804 | 2804 | positive | 1 | -3.42199 | 0.009073 |
| 2809 | 2809 | positive | 3.775 | -1.14183 | 0.016836 |
| 2815 | 2815 | positive | 4.557 | 1.935817 | 0.047151 |
| 2821 | 2821 | positive | 4.504 | 1.247197 | 0.033979 |
| 2829 | 2829 | positive | 0.823 | -1.68661 | 0.001711 |
| 2840 | 2840 | positive | 3.317 | -2.53883 | 0.005362 |
| 2850 | 2850 | positive | 3.157 | -2.40116 | 0.001896 |
| 2852 | 2852 | positive | 4.529 | 1.310407 | 0.021409 |
| 2853 | 2853 | positive | 0.923 | -3.21639 | 0.003894 |
| 2860 | 2860 | positive | 4.531 | 1.181536 | 0.030398 |
| 2905 | 2905 | positive | 2.579 | -1.84755 | 0.013293 |
| 2914 | 2914 | positive | 5.316 | -3.13431 | 0.00451 |
| 2924 | 2924 | positive | 4.727 | 5.847712 | 0.023309 |
| 2926 | 2926 | positive | 4.809 | -2.25885 | 0.018961 |
| 2932 | 2932 | positive | 2.649 | -1.82929 | 0.018961 |

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|---|---|---|---|---|---|
| 2971 | 2971 | positive | 4.531 | 1.228874 | 0.013964 |
| 2973 | 2973 | positive | 2.385 | -1.31894 | 0.004285 |
| 2986 | 2986 | positive | 0.904 | -2.80027 | 0.023728 |
| 2992 | 2992 | positive | 3.323 | -4.44632 | 0.014442 |
| 3025 | 3025 | positive | 4.7 | 3.344933 | 0.032349 |
| 3033 | 3033 | positive | 4.52 | 1.085292 | 0.013193 |
| 3035 | 3035 | positive | 4.53 | 1.226327 | 0.022578 |
| 3043 | 3043 | positive | 4.532 | 1.480025 | 0.016585 |
| 3074 | 3074 | positive | 4.539 | 1.62393 | 0.017014 |
| 3082 | 3082 | positive | 1.951 | -1.10581 | 0.004436 |
| 3116 | 3-hydroxyaminophenol | positive | 1.107 | 1.515954 | 0.040104 |
| 3127 | 3127 | positive | 1.232 | -1.43787 | 0.02724 |
| 3141 | 3141 | positive | 1.133 | -2.42642 | 0.001216 |
| 3148 | 3148 | positive | 0.812 | -3.09632 | 0.000826 |
| 3154 | 3154 | positive | 3.884 | -2.69949 | 0.000828 |
| 3159 | 3159 | positive | 1.095 | -1.68751 | 0.011539 |
| 3160 | 3160 | positive | 0.927 | -1.16062 | 0.008933 |
| 3169 | 2-hydroxyphenethylamine | positive | 4.094 | 2.210126 | 0.042089 |
| 3171 | 3171 | positive | 1.131 | -3.39246 | 0.011164 |
| 3175 | 3175 | positive | 2.581 | -1.89816 | 0.003896 |
| 3200 | 3200 | positive | 3.487 | 1.035815 | 0.038541 |
| 3208 | 3208 | positive | 1.098 | -5.19983 | 0.00451 |
| 3234 | 3234 | positive | 1.147 | -2.41971 | 0.016388 |
| 3260 | 3260 | positive | 3.056 | 1.146707 | 0.012962 |
| 3262 | 3262 | positive | 4.751 | 3.143639 | 0.048424 |
| 3283 | 3283 | positive | 2.138 | -1.42525 | 0.006117 |
| 3297 | 3297 | positive | 4.706 | 6.646371 | 0.016585 |
| 3301 | 3301 | positive | 4.531 | 1.001697 | 0.016003 |
| 3309 | 3309 | positive | 0.934 | -2.04523 | 0.003896 |
| 3353 | 3353 | positive | 4.509 | 1.111479 | 0.043452 |
| 3362 | 3362 | positive | 4.073 | -5.08514 | 0.001646 |
| 3370 | 3370 | positive | 4.703 | 6.087944 | 0.014648 |
| 3371 | 3371 | positive | 0.922 | -3.51338 | 0.003894 |
| 3385 | 3385 | positive | 3.329 | -2.60407 | 0.008088 |
| 3390 | 3390 | positive | 2.633 | -1.751 | 0.006372 |
| 3409 | 3409 | positive | 1.857 | -1.26767 | 0.014577 |
| 3415 | 3415 | positive | 4.534 | 1.165491 | 0.009093 |
| 3427 | 3427 | positive | 2.631 | -2.44854 | 0.013293 |
| 3441 | 2-hydroxyphenethylamine | positive | 3.894 | 2.55209 | 0.033619 |
| 3446 | deoxycytidine | positive | 3.531 | 1.085392 | 0.0313 |
| 3449 | 3449 | positive | 1.861 | -1.22268 | 0.008986 |

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|---|---|---|---|---|---|
| 3492 | cathinone | positive | 4.181 | 1.918288 | 0.022548 |
| 3514 | 3514 | positive | 4.533 | 1.39721 | 0.016455 |
| 3517 | pyridafenthion | positive | 0.812 | -3.76591 | 0.00132 |
| 3526 | 3526 | positive | 1.914 | -1.09411 | 0.002947 |
| 3528 | 3528 | positive | 2.258 | -1.87687 | 0.015545 |
| 3545 | 3545 | positive | 0.826 | -1.63608 | 0.00144 |
| 3546 | 3546 | positive | 0.827 | -1.63797 | 0.00144 |
| 3552 | 3552 | positive | 4.531 | 1.223489 | 0.013193 |
| 3558 | 3558 | positive | 4.54 | 1.239042 | 0.022102 |
| 3564 | 3564 | positive | 0.848 | 2.507408 | 0.019028 |
| 3582 | 3582 | positive | 1.214 | -1.5372 | 0.011539 |
| 3586 | 3586 | positive | 4.159 | -2.72666 | 0.014442 |
| 3596 | beta-ionone | positive | 2.632 | -1.75123 | 0.003896 |
| 3613 | 3613 | positive | 3.957 | -2.40024 | 0.000586 |
| 3624 | 3624 | positive | 1.069 | -1.34416 | 0.021839 |
| 3626 | 3626 | positive | 4.523 | 1.198337 | 0.01772 |
| 3632 | 3632 | positive | 4.555 | 1.298889 | 0.013193 |
| 3657 | 3657 | positive | 1.293 | -1.85846 | 0.009093 |
| 3675 | 3675 | positive | 1.184 | -1.08046 | 0.000262 |
| 3701 | 3701 | positive | 0.819 | -1.69784 | 0.00105 |
| 3757 | 3757 | positive | 4.003 | -2.38649 | 0.000326 |
| 3763 | 3763 | positive | 4.009 | -2.27464 | 0.001528 |
| 3764 | 3764 | positive | 3.954 | -2.33779 | 0.00089 |
| 3777 | 3777 | positive | 3.639 | -2.09303 | 0.00144 |
| 3791 | 3791 | positive | 3.69 | -2.04221 | 0.002947 |
| 3797 | 3797 | positive | 3.938 | -2.28156 | 0.001221 |
| 3799 | 3799 | positive | 3.621 | -2.11755 | 0.001216 |
| 3804 | hippuric acid | positive | 2.595 | -2.02465 | 0.000526 |
| 3820 | 3820 | positive | 3.979 | -2.26378 | 0.000579 |
| 3823 | 3823 | positive | 3.924 | -2.42884 | 0.000734 |
| 3829 | 3829 | positive | 3.894 | -2.35241 | 0.001496 |
| 3842 | 3842 | positive | 3.673 | -2.06698 | 0.001737 |
| 3856 | 3856 | positive | 4.016 | -2.4126 | 0.000734 |
| 3863 | 3863 | positive | 3.373 | -2.05737 | 0.043366 |
| 3871 | 3871 | positive | 1.021 | -2.67385 | 0.002122 |
| 3872 | 3872 | positive | 4.049 | -2.5791 | 0.00019 |
| 3873 | 3873 | positive | 3.698 | -2.19666 | 0.004044 |
| 3876 | 3876 | positive | 3.998 | -2.39598 | 0.000515 |
| 3893 | 3893 | positive | 3.727 | -2.47178 | 0.004492 |
| 3905 | 3905 | positive | 3.87 | -2.37325 | 0.000826 |
| 3906 | 3906 | positive | 3.848 | -2.54952 | 0.003254 |
| 3907 | 3907 | positive | 3.779 | -2.70752 | 0.004285 |

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|---|---|---|---|---|---|
| 3909 | 3909 | positive | 3.579 | -2.32192 | 0.00089 |
| 3923 | 3923 | positive | 4.016 | -3.24391 | 0.00089 |
| 3925 | 3925 | positive | 3.751 | -2.62574 | 0.004245 |
| 3939 | 3939 | positive | 3.304 | -2.57864 | 0.028679 |
| 3944 | 3944 | positive | 3.035 | 1.066588 | 0.048739 |
| 3960 | 3960 | positive | 3.996 | -2.13732 | 0.001054 |
| 3963 | 3963 | positive | 2.184 | 1.099552 | 0.008986 |
| 3968 | 3968 | positive | 4.566 | -2.64268 | 0.01657 |
| 3976 | 3976 | positive | 4.73 | 6.193516 | 0.022548 |
| 3987 | 3987 | positive | 3.971 | -2.30352 | 0.000679 |
| 3991 | leucinamide | positive | 1.074 | -3.08451 | 0.002279 |
| 3992 | 3992 | positive | 3.954 | -2.7693 | 0.001256 |
| 4001 | 4001 | positive | 1.099 | -4.10801 | 0.025007 |
| 4025 | 4025 | positive | 3.439 | 1.279099 | 0.008317 |
| 4042 | 4042 | positive | 3.883 | -2.52495 | 0.000734 |
| 4075 | 4075 | positive | 3.416 | -2.03456 | 0.008933 |
| 4080 | 4080 | positive | 0.821 | -3.4898 | 0.000515 |
| 4133 | 4133 | positive | 4.036 | -2.72562 | 0.004666 |
| 4162 | 4162 | positive | 3.455 | -2.11785 | 0.005543 |
| 4179 | 4179 | positive | 3.602 | -1.00487 | 0.021297 |
| 4180 | 4180 | positive | 3.622 | -1.00845 | 0.018248 |
| 4189 | 4189 | positive | 1.281 | -1.58871 | 0.0418 |
| 4195 | 4195 | positive | 1.114 | -1.26086 | 0.008469 |
| 4210 | 4210 | positive | 3.555 | -2.31772 | 0.001701 |
| 4218 | 4218 | positive | 3.48 | -2.25802 | 0.00129 |
| 4250 | 4250 | positive | 4.057 | -2.558 | 0.00132 |
| 4258 | 5-hydroxy-tryptophan | positive | 3.033 | 1.202781 | 0.04448 |
| 4265 | 4265 | positive | 4.102 | -2.60288 | 0.00129 |
| 4267 | 4267 | positive | 3.758 | -1.26957 | 0.04047 |
| 4278 | 4278 | positive | 3.072 | 1.28831 | 0.011416 |
| 4279 | 4279 | positive | 6.949 | 2.098916 | 0.046946 |
| 4281 | 4281 | positive | 4.05 | -2.76374 | 0.00089 |
| 4283 | 4283 | positive | 4.687 | 2.857637 | 0.017731 |
| 4288 | 4288 | positive | 4.612 | 1.304232 | 0.030454 |
| 4303 | 4303 | positive | 2.623 | -1.02546 | 0.00597 |
| 4318 | 4318 | positive | 4.037 | -2.72294 | 0.00073 |
| 4323 | 4323 | positive | 4.122 | -2.34523 | 0.001873 |
| 4328 | 4328 | positive | 3.96 | -2.35681 | 0.002367 |
| 4340 | 4340 | positive | 5.076 | -1.9714 | 0.02593 |
| 4352 | leucinamide | positive | 0.919 | -2.16241 | 0.00132 |
| 4355 | 4355 | positive | 4.057 | -2.73285 | 0.00129 |

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|---|---|---|---|---|---|
| 4367 | pregabalin | positive | 1.06 | -2.97568 | 0.001496 |
| 4370 | 4370 | positive | 4.113 | -2.29843 | 0.002367 |
| 4381 | 4381 | positive | 4.646 | 1.122786 | 0.030398 |
| 4382 | 4382 | positive | 4.541 | -3.18497 | 0.004544 |
| 4384 | 4384 | positive | 4.073 | -2.25311 | 0.001565 |
| 4392 | 4392 | positive | 3.941 | -2.10559 | 0.000734 |
| 4401 | 4401 | positive | 4.039 | -2.69474 | 0.000481 |
| 4408 | 1-aAmino-1-deoxy-scyllo-inositol | positive | 3.867 | -1.12923 | 0.011113 |
| 4413 | 4413 | positive | 4.052 | -2.37203 | 0.001699 |
| 4428 | 4428 | positive | 4.057 | -2.22436 | 0.002596 |
| 4444 | 4444 | positive | 3.998 | -2.43206 | 0.000912 |
| 4447 | 4447 | positive | 4.061 | -2.50909 | 0.000579 |
| 4490 | 4490 | positive | 3.94 | -2.3298 | 0.001721 |
| 4553 | 4553 | positive | 3.85 | -2.1915 | 0.002694 |
| 4587 | 4587 | positive | 4.035 | -2.55891 | 0.002251 |
| 4616 | 4616 | positive | 0.847 | 1.303658 | 0.008088 |
| 4632 | 1,7-dimethyluric acid | negative | 2.337 | -1.11272 | 0.013263 |
| 4659 | hippuric acid | negative | 2.622 | -1.12485 | 0.003896 |
| 4670 | cinnamoylglycine | negative | 2.651 | -1.55137 | 0.012441 |
| 4672 | cinnamoylglycine | negative | 2.616 | -1.814 | 0.017993 |
| 4673 | 7-methylxanthine | negative | 1.221 | -1.13939 | 0.009031 |
| 4685 | hippuric acid | negative | 4.039 | 1.414079 | 0.032791 |
| 4704 | 2-furoylglycine | negative | 2.58 | 1.951921 | 0.04693 |
| 4706 | 2-furoylglycine | negative | 2.512 | 2.68069 | 0.030454 |
| 4719 | 1-methyluric acid | negative | 2.545 | -1.157 | 0.011539 |
| 4739 | 5-hydroxyindole | negative | 2.626 | -1.0839 | 0.004436 |
| 4740 | 5-hydroxyindole | negative | 2.581 | -1.15681 | 0.005204 |
| 4766 | theophylline | negative | 1.432 | -2.1935 | 0.001975 |
| 4775 | 4775 | negative | 0.869 | -2.28845 | 0.027288 |
| 4791 | 4791 | negative | 4.511 | 1.244399 | 0.016348 |
| 4801 | 4801 | negative | 4.554 | 1.085615 | 0.016222 |
| 4844 | 4844 | negative | 3.257 | 2.311035 | 0.036851 |
| 4870 | 3-[-5-oxo-7-oxabicyclo[4.1.0]hept-2-yl]-alanine | negative | 3.24 | -1.41362 | 0.015568 |
| 4953 | 7,8-dihydro-8-oxoguanine | negative | 1.198 | -1.23311 | 0.008986 |
| 4958 | 4958 | negative | 3.679 | -1.25361 | 0.048489 |
| 5010 | 5010 | negative | 4.637 | 2.953354 | 0.003254 |
| 5029 | 5029 | negative | 4.152 | -2.73447 | 0.008511 |
| 5038 | 5038 | negative | 2.632 | 2.080506 | 0.037266 |
| 5065 | 5065 | negative | 0.855 | -1.47704 | 0.003254 |
| 5077 | 5077 | negative | 1.965 | 1.045916 | 0.012965 |
| 5083 | 5083 | negative | 2.564 | 1.75927 | 0.04448 |

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|---|---|---|---|---|---|
| 5106 | 5106 | negative | 3.534 | 1.315406 | 0.025624 |
| 5110 | 1-O-[5-(3,4-dihydroxyphenyl)-4-hydroxypentanoyl]-beta-glucopyranuronic acid | negative | 3.5 | -1.96301 | 0.0151 |
| 5129 | 5129 | negative | 2.64 | -1.94809 | 0.00129 |
| 5133 | 5133 | negative | 3.707 | -2.21718 | 0.032369 |
| 5137 | 5137 | negative | 2.517 | -1.15218 | 0.023309 |
| 5153 | 5153 | negative | 1.421 | -1.48097 | 0.007752 |
| 5216 | 5216 | negative | 0.863 | -1.85939 | 0.005858 |
| 5239 | 5239 | negative | 3.529 | -1.09635 | 0.043366 |
| 5241 | butopyronoxyl | negative | 0.908 | 1.206619 | 0.038863 |
| 5248 | 5248 | negative | 4.531 | 1.057774 | 0.012999 |
| 5285 | 5285 | negative | 4.644 | 3.023183 | 0.009913 |
| 5310 | 5310 | negative | 0.87 | 2.988694 | 0.017548 |
| 5341 | 5341 | negative | 0.706 | -1.09358 | 0.03992 |
| 5352 | 5352 | negative | 2.355 | -1.00687 | 0.04099 |
| 5358 | 5358 | negative | 3.897 | 1.636177 | 0.021839 |
| 5379 | 5379 | negative | 0.863 | -1.86478 | 0.009093 |
| 5381 | 5381 | negative | 3.395 | 1.698478 | 0.004044 |
| 5383 | 5383 | negative | 4.067 | -2.62497 | 0.000385 |
| 5393 | 5393 | negative | 0.862 | -1.87627 | 0.006038 |
| 5423 | 5423 | negative | 3.745 | -1.83453 | 0.030529 |
| 5463 | N-acetyl-methionine | negative | 3.758 | -1.85466 | 0.033538 |
| 5470 | sulfurol acetate | negative | 3.902 | -2.52855 | 0.034206 |
| 5482 | 5482 | negative | 0.858 | -1.17721 | 0.034734 |
| 5507 | 5507 | negative | 3.132 | -1.06713 | 0.01418 |
| 5514 | 5514 | negative | 2.517 | -1.48437 | 0.004245 |
| 5553 | 5553 | negative | 2.562 | -1.25185 | 0.03108 |
| 5576 | 5576 | negative | 0.728 | -1.56108 | 0.008088 |
| 5604 | 5604 | negative | 3.891 | -3.78571 | 0.003896 |
| 5612 | 5612 | negative | 3.805 | -1.02889 | 0.043481 |
| 5647 | 5647 | negative | 3.993 | -2.70499 | 0.000679 |
| 5648 | 5648 | negative | 3.966 | -2.75565 | 0.006117 |
| 5683 | 5683 | negative | 4.089 | -3.72301 | 0.001247 |
| 5698 | 5698 | negative | 3.381 | 1.402511 | 0.011539 |
| 5712 | 5712 | negative | 4.376 | -1.96165 | 0.029132 |
| 5724 | 5724 | negative | 4.531 | 1.170521 | 0.015527 |
| 5728 | 5728 | negative | 2.931 | -1.11438 | 0.039852 |
| 5737 | gly-Lys | negative | 3.995 | 1.486072 | 0.017371 |
| 5770 | 5770 | negative | 0.855 | -1.45399 | 0.00451 |
| 5796 | pidotimod | negative | 2.951 | 1.72657 | 0.040795 |
| 5799 | 5799 | negative | 4 | -2.4648 | 0.038921 |

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|---|---|---|---|---|---|
| 5818 | 5818 | negative | 0.948 | 1.355602 | 0.048525 |
| 5820 | 5820 | negative | 4.663 | 1.182431 | 0.020072 |
| 5849 | 5849 | negative | 0.708 | -1.10963 | 0.039503 |
| 5868 | 5868 | negative | 4.66 | 1.321084 | 0.010661 |
| 5887 | 5887 | negative | 4.532 | -1.65544 | 0.001001 |
| 5899 | 5899 | negative | 3.5 | 1.151716 | 0.032924 |
| 5911 | 5911 | negative | 3.966 | -2.51989 | 0.001092 |
| 5925 | 5925 | negative | 2.671 | -1.32396 | 0.006117 |
| 5931 | 5931 | negative | 2.571 | -1.02026 | 0.010661 |
| 5941 | 5941 | negative | 1.148 | 1.901496 | 0.032924 |
| 5942 | 5942 | negative | 2.631 | -1.18178 | 0.002324 |
| 5994 | 5994 | negative | 2.625 | -1.08762 | 0.00451 |
| 6001 | 6001 | negative | 2.782 | -1.94029 | 0.006743 |
| 6007 | 6007 | negative | 0.678 | -1.21015 | 0.041623 |
| 6014 | N-acetyl-tyrosine | negative | 2.632 | -1.5866 | 0.009093 |
| 6021 | 6021 | negative | 0.708 | -1.36188 | 0.003896 |
| 6057 | 6057 | negative | 0.947 | 1.642112 | 0.031478 |
| 6094 | 5-acetylamino-6-formylamino-3-methyluracil | negative | 0.727 | -1.59148 | 0.004603 |
| 6095 | chiro-inositol | negative | 1.118 | -1.37163 | 0.00129 |
| 6101 | 6101 | negative | 4.052 | -1.52997 | 0.045816 |
| 6111 | 6111 | negative | 2.577 | 2.205537 | 0.04782 |
| 6148 | 6148 | negative | 2.587 | 1.33016 | 0.028806 |
| 6161 | 6161 | negative | 0.711 | -1.44766 | 0.005882 |
| 6190 | 6190 | negative | 2.594 | -1.31465 | 0.027361 |
| 6212 | glaucarubin | negative | 3.361 | -2.88626 | 0.025629 |
| 6233 | 6233 | negative | 1.515 | -1.15693 | 0.015545 |
| 6236 | 6236 | negative | 0.625 | 1.384644 | 0.025156 |
| 6261 | 6261 | negative | 2.591 | -1.8809 | 0.000368 |
| 6262 | hippurate-mannitol derivative | negative | 2.667 | -1.80394 | 0.000515 |
| 6267 | 6267 | negative | 2.581 | 2.159467 | 0.032369 |
| 6276 | 6276 | negative | 2.636 | -1.65054 | 0.000481 |
| 6286 | 2_5_6-trihydroxy-5_6-dihydroquinoline | negative | 2.622 | -1.30303 | 0.005058 |
| 6314 | 6314 | negative | 2.625 | -1.08453 | 0.004492 |
| 6322 | 6322 | negative | 2.623 | -1.04139 | 0.00451 |
| 6325 | 6325 | negative | 2.64 | -1.97981 | 0.001564 |
| 6327 | 6327 | negative | 2.637 | -1.5487 | 0.028679 |
| 6337 | 6337 | negative | 0.862 | -1.69061 | 0.002457 |
| 6348 | 6348 | negative | 2.993 | -2.21045 | 0.017548 |
| 6349 | 6349 | negative | 0.93 | -1.14826 | 0.021927 |
| 6361 | 6361 | negative | 2.619 | -1.1059 | 0.001844 |
| 6375 | 6375 | negative | 2.775 | -1.81499 | 0.009031 |

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|---|---|---|---|---|---|
| 6385 | 6385 | negative | 2.655 | -1.00016 | 0.008703 |
| 6389 | 6389 | negative | 2.516 | -1.13448 | 0.011046 |
| 6390 | 6390 | negative | 2.627 | -1.00038 | 0.004603 |
| 6392 | 6392 | negative | 0.75 | -1.01094 | 0.012242 |
| 6396 | 6396 | negative | 0.884 | -1.32351 | 0.027563 |
| 6406 | 6406 | negative | 0.903 | -2.03474 | 0.04006 |
| 6425 | leupeptin | negative | 4.004 | -2.47724 | 0.00119 |
| 6447 | 6447 | negative | 2.421 | 3.406793 | 0.024455 |
| 6496 | 6496 | negative | 1.25 | -1.16132 | 0.008933 |
| 6504 | 6504 | negative | 2.582 | 2.311681 | 0.032369 |
| 6508 | 6508 | negative | 2.569 | -1.00789 | 0.004285 |
| 6534 | 6534 | negative | 3.989 | -2.43582 | 0.001737 |
| 6544 | 6544 | negative | 0.719 | -1.5791 | 0.004603 |
| 6545 | 6545 | negative | 4.005 | -2.41651 | 0.001342 |
| 6565 | 6565 | negative | 2.935 | -1.08416 | 0.041344 |
| 6569 | 4-[(-2-amino-1-hydroxyethyl]-2-hydroxyphenyl hydrogen sulfate | negative | 1.181 | 1.368161 | 0.047987 |
| 6578 | 2-mercaptobenzothiazole | negative | 0.832 | 2.229249 | 0.009064 |
| 6594 | N-acetyl-glucosaminate | negative | 3.871 | 1.156155 | 0.021542 |
| 6628 | 6628 | negative | 2.908 | -1.35524 | 0.022578 |
| 6637 | 6637 | negative | 4.866 | 1.31038 | 0.01503 |
| 6662 | 6662 | negative | 3.528 | -2.06367 | 0.011539 |
| 6668 | 6668 | negative | 2.612 | -1.00887 | 0.004285 |
| 6676 | 6676 | negative | 3.184 | -2.34598 | 0.002651 |
| 6683 | 6683 | negative | 3.025 | -2.14478 | 0.012441 |
| 6687 | 6687 | negative | 0.866 | -1.68528 | 0.003166 |
| 6731 | 6731 | negative | 0.859 | -1.79499 | 0.002105 |
| 6762 | 6762 | negative | 3.751 | -2.80798 | 0.003896 |
| 6802 | 6802 | negative | 1.504 | -1.71402 | 0.030398 |
| 6819 | 6819 | negative | 1.109 | -3.47044 | 0.008588 |
| 6882 | 6882 | negative | 3.692 | -3.0581 | 0.011543 |
| 6885 | 6885 | negative | 2.615 | -1.03401 | 0.002324 |
| 6939 | 6939 | negative | 2.586 | -1.33936 | 0.043481 |
| 6956 | 6956 | negative | 2.715 | 1.925851 | 0.017014 |
| 6972 | 6972 | negative | 1.112 | -3.3917 | 0.008088 |
| 6990 | 6990 | negative | 2.647 | -1.67258 | 0.011539 |
| 6996 | 6996 | negative | 2.535 | -1.04791 | 0.021194 |
| 7001 | 7001 | negative | 0.869 | -1.71768 | 0.003166 |
| 7087 | 7087 | negative | 2.399 | -1.19546 | 0.007612 |

## References


1. Bingol, K.; Li, D. W.; Zhang, B.; Bruschweiler, R., Comprehensive Metabolite Identification Strategy Using Multiple Two-Dimensional NMR Spectra of a Complex Mixture Implemented in the COLMARm Web Server. *Anal Chem* **2016,** *88* (24), 12411-12418.
2. Kind, T.; Tolstikov, V.; Fiehn, O.; Weiss, R. H., A comprehensive urinary metabolomic approach for identifying kidney cancerr. *Anal Biochem* **2007,** *363* (2), 185-95.
3. Kim, K.; Aronov, P.; Zakharkin, S. O.; Anderson, D.; Perroud, B.; Thompson, I. M.; Weiss, R. H., Urine metabolomics analysis for kidney cancer detection and biomarker discovery. *Mol Cell Proteomics* **2009,** *8* (3), 558-70.
4. Kim, K.; Taylor, S. L.; Ganti, S.; Guo, L.; Osier, M. V.; Weiss, R. H., Urine metabolomic analysis identifies potential biomarkers and pathogenic pathways in kidney cancer. *OMICS* **2011,** *15* (5), 293-303.
5. Monteiro, M. S.; Barros, A. S.; Pinto, J.; Carvalho, M.; Pires-Luis, A. S.; Henrique, R.; Jeronimo, C.; Bastos, M. L.; Gil, A. M.; Guedes de Pinho, P., Nuclear Magnetic Resonance metabolomics reveals an excretory metabolic signature of renal cell carcinoma. *Sci Rep* **2016,** *6*, 37275.
6. Rosa Ragone, F. S., Sara Piccinonna, Monica Rutigliano, Galleggiante Vanessa, Silvano Palazzo, Giuseppe Lucarelli, Pasquale Ditonno, Michele Battaglia, Francesco Paolo Fanizzi, Francesco Paolo Schena, Renal Cell Carcinoma: A Study Through NMR-Based Metabolomics Combined With Transcriptomics. *Diseases* **2016,** *4* (1), 7.
7. Monteiro, M.; Moreira, N.; Pinto, J.; Pires-Luis, A. S.; Henrique, R.; Jeronimo, C.; Bastos, M. L.; Gil, A. M.; Carvalho, M.; Guedes de Pinho, P., GC-MS metabolomics-based approach for the identification of a potential VOC-biomarker panel in the urine of renal cell carcinoma patients. *J Cell Mol Med* **2017,** *21* (9), 2092-2105.
8. Niziol, J.; Bonifay, V.; Ossolinski, K.; Ossolinski, T.; Ossolinska, A.; Sunner, J.; Beech, I.; Arendowski, A.; Ruman, T., Metabolomic study of human tissue and urine in clear cell renal carcinoma by LC-HRMS and PLS-DA. *Anal Bioanal Chem* **2018,** *410* (16), 3859-3869.
9. Liu, X.; Zhang, M.; Liu, X.; Sun, H.; Guo, Z.; Tang, X.; Wang, Z.; Li, J.; Li, H.; Sun, W.; Zhang, Y., Urine Metabolomics for Renal Cell Carcinoma (RCC) Prediction: Tryptophan Metabolism as an Important Pathway in RCC. *Front Oncol* **2019,** *9*, 663.
10. Wang, Z.; Liu, X.; Liu, X.; Sun, H.; Guo, Z.; Zheng, G.; Zhang, Y.; Sun, W., UPLC-MS based urine untargeted metabolomic analyses to differentiate bladder cancer from renal cell carcinoma. *BMC Cancer* **2019,** *19* (1), 1195.
11. Zhang, M.; Liu, X.; Liu, X.; Li, H.; Sun, W.; Zhang, Y., A pilot investigation of a urinary metabolic biomarker discovery in renal cell carcinoma. *Int Urol Nephrol* **2020,** *52* (3), 437-446.