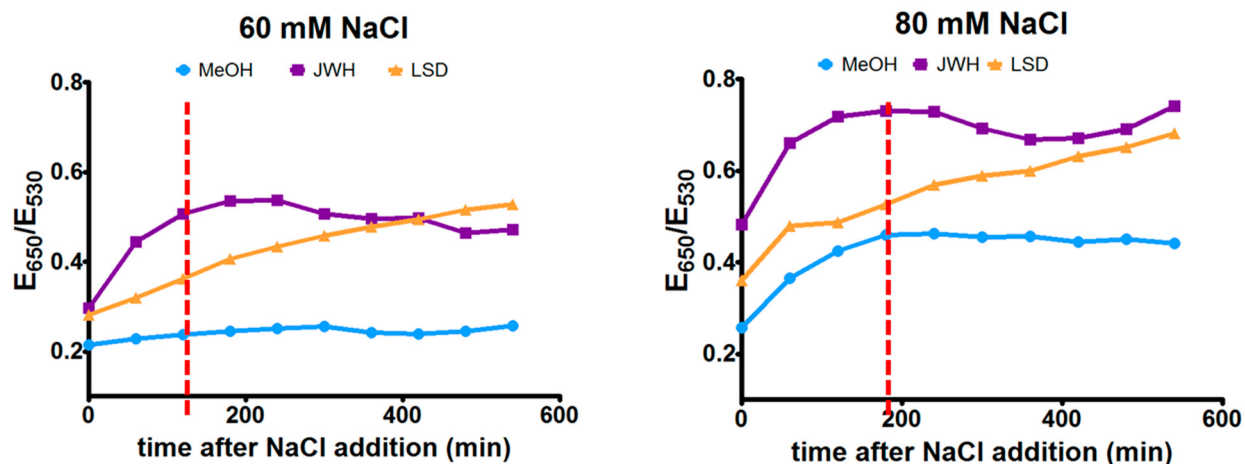
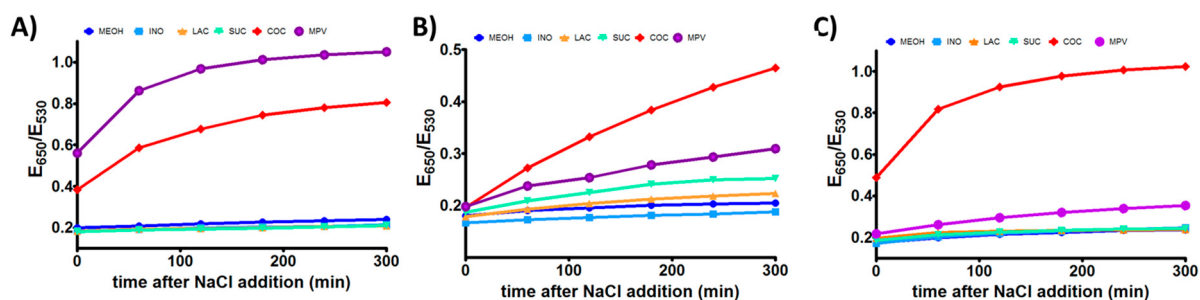
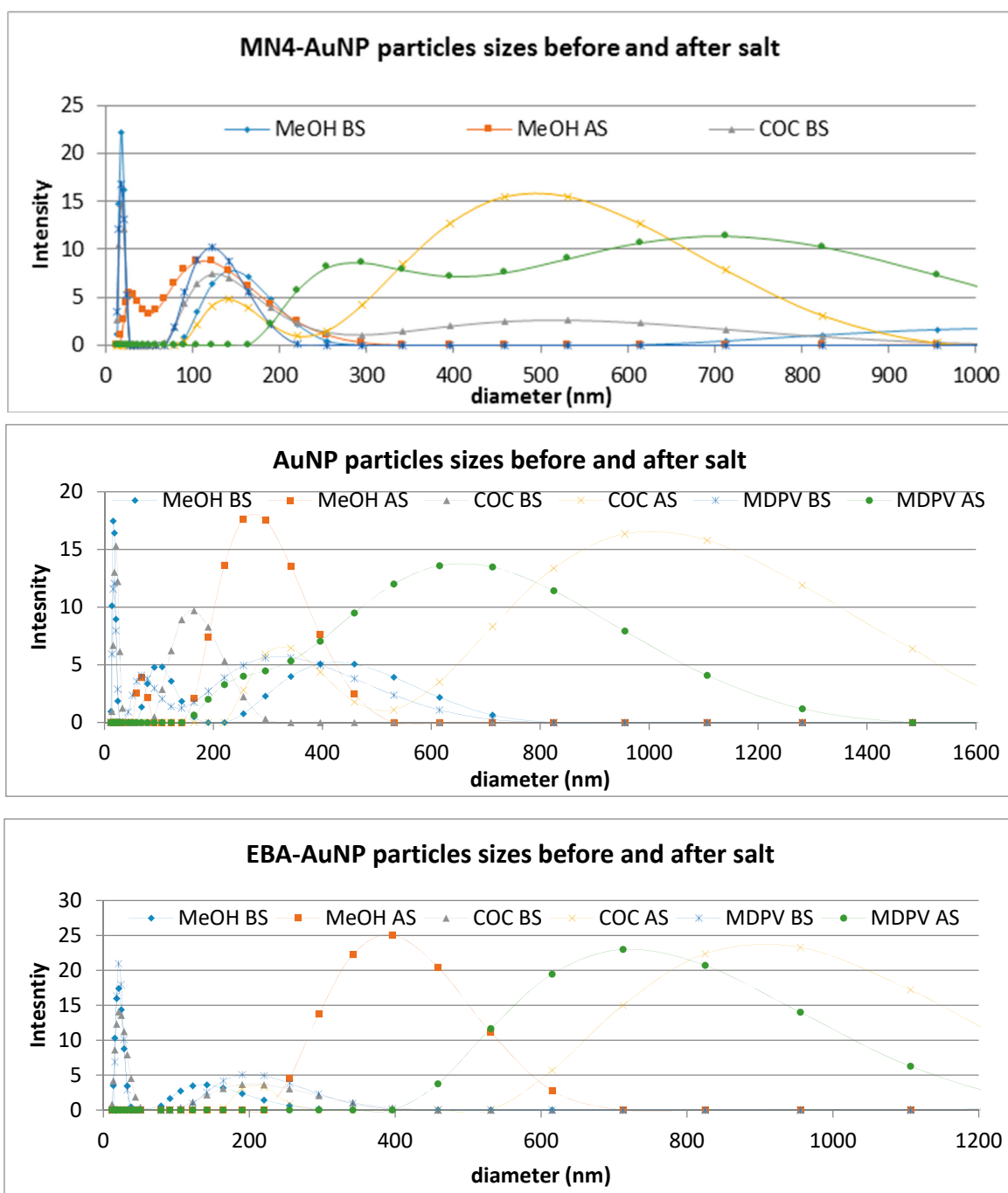




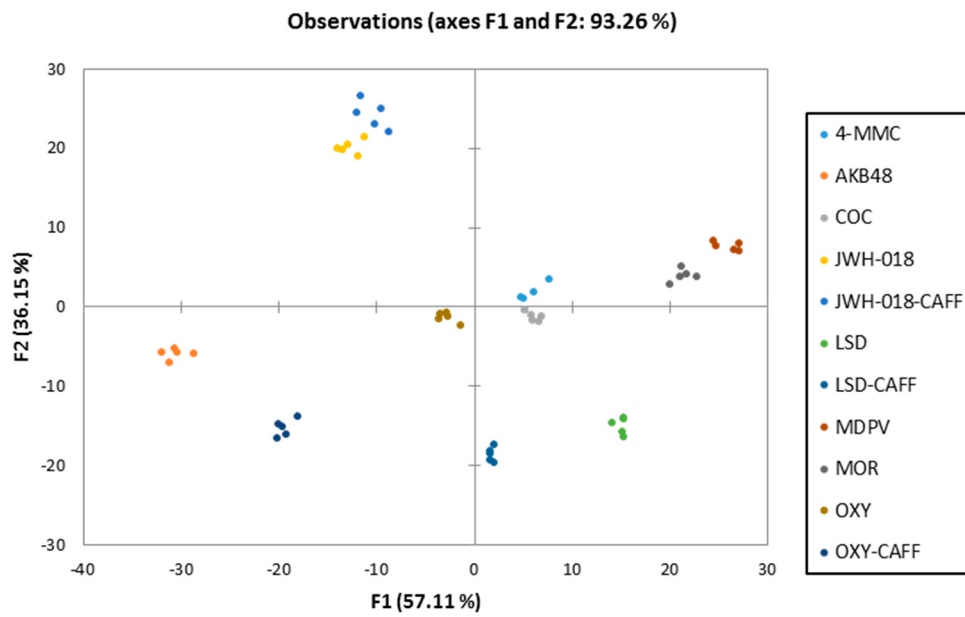
**Figure S1.** Assay Optimization. Initial trial to identify the optimal salt concentration and incubation time with salt to obtain different assay responses to the analytes of interest using the MN4-AuNPs. The red lines indicate the proposed optimal test time for each salt concentration tested.



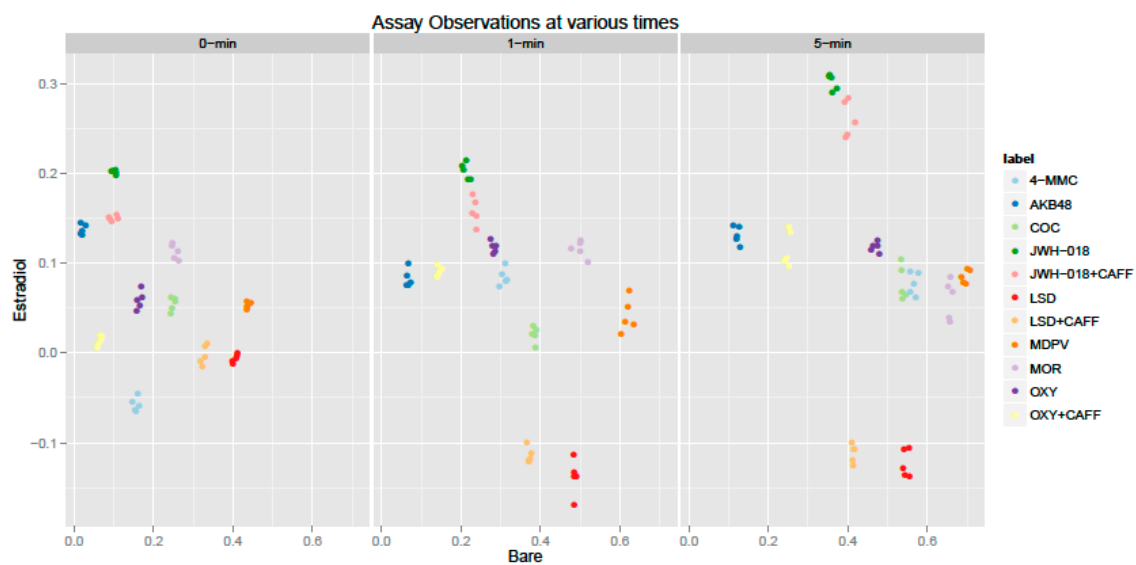
**Figure S2.** Comparison of Assay Response to Controlled Substances and Fillers. Analytes were dissolved in a 1:1 mixture of methanol:buffer to a concentration of 0.5 mg/mL. Apt-AuNPs were mixed with the analytes and incubated for 30 seconds before NaCl addition. Apt-AuNPs extinction was measured immediately after salt addition: (A) c-AuNPs, (B) MN4-AuNPs and (C) EBA-AuNPs.



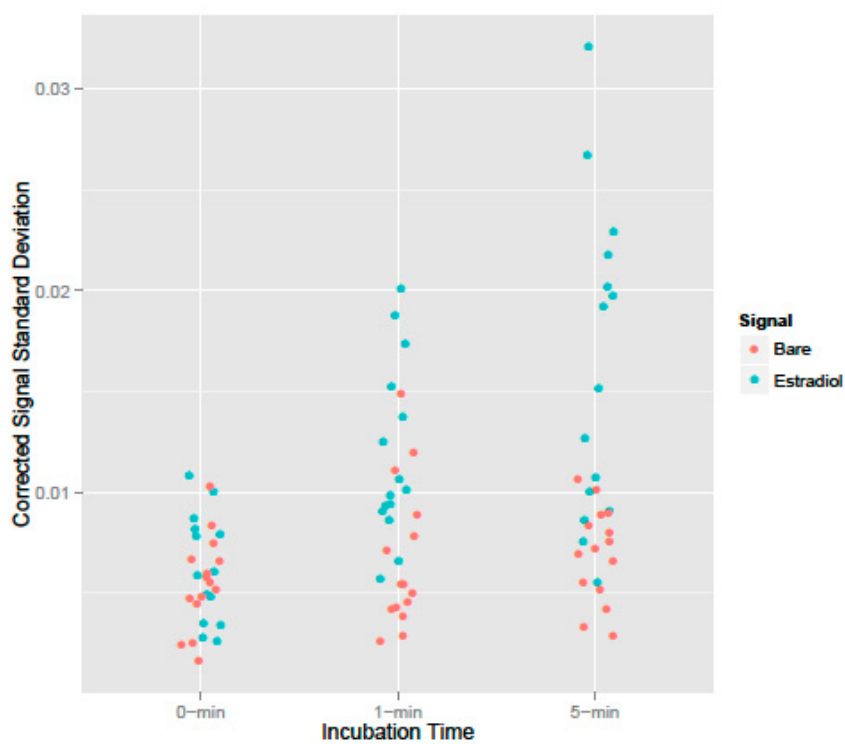
**Figure S3.** Dynamic light scattering data showing particle sizes before and after addition of salt. Each sensor shows the trend that the addition of salt causes the particles to destabilize and aggregate. The target also has an influence on stability, for methanol typically has the smallest size, showing the least reaction with the AuNP.



**Figure S4.** Differential analysis classification of non-corrected sensor data, data was recorded after three minutes incubation time with salt.



(a)



(b)

**Figure S5.** (a) Assay Scatterplot Matrix. C-AuNPs (Bare) and EBA-AuNPs (Estradiol) Signals are plotted against each other showing the separation between various chemical classes at either 0, 1, or 5 minute incubation times. (b) Variance comparison between c-AuNPs (Bare) and EBA-AuNPs (Estradiol) at 0, 1, or 5 minute incubation. Note increased Estradiol measurement variance at longer incubation time. Variance is computed from 5 replicates for each chemical class (e.g., OXY, OXY+CAFF, etc.) and incubation time.

**Table S1.** Input data for discriminant analysis of assay response training set (30 s).

|              | <b>Bare</b> | <b>Estradiol</b> | <b>MN4</b> |
|--------------|-------------|------------------|------------|
| COC          | 0.242747    | 0.043034         | 0.032422   |
| COC          | 0.244034    | 0.062167         | 0.018335   |
| COC          | 0.253313    | 0.057461         | 0.02521    |
| COC          | 0.252413    | 0.05968          | 0.032008   |
| COC          | 0.246154    | 0.050114         | 0.023525   |
| JWH-018      | 0.092773    | 0.201931         | 0.178824   |
| JWH-018      | 0.103827    | 0.204169         | 0.179728   |
| JWH-018      | 0.095444    | 0.202929         | 0.16916    |
| JWH-018      | 0.104125    | 0.200342         | 0.170512   |
| JWH-018      | 0.104125    | 0.196993         | 0.171582   |
| LSD          | 0.398833    | -0.00912         | -0.00807   |
| LSD          | 0.409389    | -0.00352         | 0.002378   |
| LSD          | 0.407837    | -0.00623         | 0.002414   |
| LSD          | 0.399593    | -0.01259         | 0.005095   |
| LSD          | 0.411216    | 0.000197         | 0.004428   |
| MOR          | 0.251978    | 0.105866         | 0.026715   |
| MOR          | 0.260599    | 0.113454         | 0.034492   |
| MOR          | 0.246414    | 0.118502         | 0.028738   |
| MOR          | 0.247573    | 0.12216          | 0.029224   |
| MOR          | 0.262727    | 0.102793         | 0.031923   |
| MDPV         | 0.434748    | 0.048838         | 0.058796   |
| MDPV         | 0.435382    | 0.05123          | 0.040697   |
| MDPV         | 0.439548    | 0.053891         | 0.046526   |
| MDPV         | 0.43521     | 0.057297         | 0.047991   |
| MDPV         | 0.445161    | 0.056135         | 0.049713   |
| OXY          | 0.171144    | 0.061445         | 0.00789    |
| OXY          | 0.164933    | 0.052371         | 0.002808   |
| OXY          | 0.158335    | 0.058637         | 0.010613   |
| OXY          | 0.168007    | 0.07338          | -0.00257   |
| OXY          | 0.157347    | 0.046786         | -0.00257   |
| JWH-018-CAFF | 0.093932    | 0.146455         | 0.162545   |
| JWH-018-CAFF | 0.109971    | 0.148952         | 0.133489   |
| JWH-018-CAFF | 0.089943    | 0.149995         | 0.125958   |
| JWH-018-CAFF | 0.087418    | 0.150911         | 0.147592   |
| JWH-018-CAFF | 0.107195    | 0.153682         | 0.121859   |
| LSD-CAFF     | 0.33574     | 0.010434         | -0.00777   |
| LSD-CAFF     | 0.329774    | 0.006722         | 0.004414   |
| LSD-CAFF     | 0.322931    | -0.01503         | -0.00314   |
| LSD-CAFF     | 0.330476    | -0.00432         | -0.0023    |
| LSD-CAFF     | 0.318852    | -0.0098          | 0.001099   |
| OXY-CAFF     | 0.065197    | 0.019133         | -0.04701   |
| OXY-CAFF     | 0.05945     | 0.00945          | -0.06856   |
| OXY-CAFF     | 0.066207    | 0.012819         | -0.07173   |
| OXY-CAFF     | 0.058462    | 0.005348         | -0.0687    |
| OXY-CAFF     | 0.069613    | 0.018486         | -0.06903   |
| AKB48        | 0.028672    | 0.142253         | 0.041106   |
| AKB48        | 0.019799    | 0.130954         | 0.03008    |
| AKB48        | 0.019084    | 0.135689         | 0.037095   |
| AKB48        | 0.016252    | 0.132409         | 0.028259   |
| AKB48        | 0.015758    | 0.144836         | 0.039653   |
| 4-MMC        | 0.152492    | -0.06345         | 0.043322   |

|       |          |          |          |
|-------|----------|----------|----------|
| 4-MMC | 0.159755 | -0.04522 | 0.053286 |
| 4-MMC | 0.163302 | -0.05933 | 0.055216 |
| 4-MMC | 0.146143 | -0.05433 | 0.042341 |
| 4-MMC | 0.155352 | -0.06471 | 0.032487 |

**Table S2.** Output data for discriminant analysis of assay response training set shown in Table S1.

| Observation | Prior        | Posterior    | F1      | F2      | F3     |
|-------------|--------------|--------------|---------|---------|--------|
| Obs1        | COC          | COC          | 7.608   | -0.830  | -0.719 |
| Obs2        | COC          | COC          | 6.368   | -0.028  | 2.424  |
| Obs3        | COC          | COC          | 8.160   | 0.472   | 1.423  |
| Obs4        | COC          | COC          | 7.765   | 1.122   | 0.907  |
| Obs5        | COC          | COC          | 7.651   | -0.661  | 0.861  |
| Obs6        | JWH-018      | JWH-018      | -30.800 | 17.212  | -3.765 |
| Obs7        | JWH-018      | JWH-018      | -29.242 | 18.003  | -3.521 |
| Obs8        | JWH-018      | JWH-018      | -30.363 | 16.734  | -2.649 |
| Obs9        | JWH-018      | JWH-018      | -28.786 | 17.003  | -2.904 |
| Obs10       | JWH-018      | JWH-018      | -28.519 | 16.773  | -3.308 |
| Obs11       | LSD          | LSD          | 37.078  | -1.175  | 0.870  |
| Obs12       | LSD          | LSD          | 38.182  | 0.588   | 0.421  |
| Obs13       | LSD          | LSD          | 38.160  | 0.270   | 0.160  |
| Obs14       | LSD          | LSD          | 37.354  | -0.510  | -0.779 |
| Obs15       | LSD          | LSD          | 38.143  | 1.163   | 0.559  |
| Obs16       | MOR          | MOR          | 3.921   | 4.957   | 5.495  |
| Obs17       | MOR          | MOR          | 4.581   | 6.619   | 5.470  |
| Obs18       | MOR          | MOR          | 1.972   | 6.000   | 6.323  |
| Obs19       | MOR          | MOR          | 1.847   | 6.425   | 6.609  |
| Obs20       | MOR          | MOR          | 5.827   | 5.557   | 4.827  |
| Obs21       | MDPV         | MDPV         | 37.297  | 10.645  | -0.468 |
| Obs22       | MDPV         | MDPV         | 37.381  | 9.592   | 1.611  |
| Obs23       | MDPV         | MDPV         | 37.762  | 10.452  | 1.298  |
| Obs24       | MDPV         | MDPV         | 36.777  | 10.665  | 1.391  |
| Obs25       | MDPV         | MDPV         | 38.434  | 11.151  | 1.239  |
| Obs26       | OXY          | OXY          | -5.022  | -4.280  | 2.504  |
| Obs27       | OXY          | OXY          | -5.203  | -5.771  | 2.152  |
| Obs28       | OXY          | OXY          | -6.847  | -4.945  | 1.814  |
| Obs29       | OXY          | OXY          | -6.403  | -4.085  | 4.586  |
| Obs30       | OXY          | OXY          | -5.889  | -7.027  | 2.118  |
| Obs31       | JWH-018-CAFF | JWH-018-CAFF | -25.855 | 11.007  | -6.939 |
| Obs32       | JWH-018-CAFF | JWH-018-CAFF | -23.229 | 9.901   | -3.526 |
| Obs33       | JWH-018-CAFF | JWH-018-CAFF | -26.414 | 8.512   | -2.916 |
| Obs34       | JWH-018-CAFF | JWH-018-CAFF | -27.107 | 10.033  | -5.094 |
| Obs35       | JWH-018-CAFF | JWH-018-CAFF | -23.944 | 9.368   | -1.951 |
| Obs36       | LSD-CAFF     | LSD-CAFF     | 25.453  | -2.331  | 1.746  |
| Obs37       | LSD-CAFF     | LSD-CAFF     | 24.693  | -2.076  | 0.092  |
| Obs38       | LSD-CAFF     | LSD-CAFF     | 25.487  | -4.938  | -1.126 |
| Obs39       | LSD-CAFF     | LSD-CAFF     | 25.786  | -3.539  | -0.176 |
| Obs40       | LSD-CAFF     | LSD-CAFF     | 24.364  | -4.345  | -1.155 |
| Obs41       | OXY-CAFF     | OXY-CAFF     | -17.758 | -17.102 | 3.087  |
| Obs42       | OXY-CAFF     | OXY-CAFF     | -17.651 | -19.811 | 4.381  |
| Obs43       | OXY-CAFF     | OXY-CAFF     | -16.827 | -19.412 | 5.089  |
| Obs44       | OXY-CAFF     | OXY-CAFF     | -17.466 | -20.245 | 4.024  |
| Obs45       | OXY-CAFF     | OXY-CAFF     | -16.784 | -18.538 | 5.352  |
| Obs46       | AKB48        | AKB48        | -34.635 | -1.190  | 4.350  |

|       |       |       |         |         |         |
|-------|-------|-------|---------|---------|---------|
| Obs47 | AKB48 | AKB48 | -34.995 | -3.437  | 4.380   |
| Obs48 | AKB48 | AKB48 | -35.571 | -2.532  | 4.064   |
| Obs49 | AKB48 | AKB48 | -35.659 | -3.602  | 4.650   |
| Obs50 | AKB48 | AKB48 | -36.882 | -1.666  | 4.561   |
| Obs51 | 4-MMC | 4-MMC | 2.018   | -14.065 | -12.328 |
| Obs52 | 4-MMC | 4-MMC | 1.559   | -11.335 | -11.663 |
| Obs53 | 4-MMC | 4-MMC | 3.271   | -12.323 | -13.052 |
| Obs54 | 4-MMC | 4-MMC | 0.266   | -13.599 | -11.509 |
| Obs55 | 4-MMC | 4-MMC | 2.685   | -14.826 | -11.287 |

**Table S3.** Leave-one-out cross validation results obtained with analysis performed on the dataset shown in Table S1.

| From \ To    | 4-M MC | AK B48 | COC | JWH-018 | JWH-018-CAFF | LSD | LSD-CAFF | MD PV | MOR | OXY | OXY-CAFF | Total | Correct % |
|--------------|--------|--------|-----|---------|--------------|-----|----------|-------|-----|-----|----------|-------|-----------|
| 4-MMC        | 5      | 0      | 0   | 0       | 0            | 0   | 0        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| AKB48        | 0      | 5      | 0   | 0       | 0            | 0   | 0        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| COC          | 0      | 0      | 5   | 0       | 0            | 0   | 0        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| JWH-018      | 0      | 0      | 0   | 5       | 0            | 0   | 0        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| JWH-018-CAFF | 0      | 0      | 0   | 0       | 5            | 0   | 0        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| LSD          | 0      | 0      | 0   | 0       | 0            | 5   | 0        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| LSD-CAFF     | 0      | 0      | 0   | 0       | 0            | 0   | 5        | 0     | 0   | 0   | 0        | 5     | 100.00%   |
| MDPV         | 0      | 0      | 0   | 0       | 0            | 0   | 0        | 5     | 0   | 0   | 0        | 5     | 100.00%   |
| MOR          | 0      | 0      | 0   | 0       | 0            | 0   | 0        | 0     | 5   | 0   | 0        | 5     | 100.00%   |
| OXY          | 0      | 0      | 0   | 0       | 0            | 0   | 0        | 0     | 0   | 5   | 0        | 5     | 100.00%   |
| OXY-CAFF     | 0      | 0      | 0   | 0       | 0            | 0   | 0        | 0     | 0   | 0   | 5        | 5     | 100.00%   |
| Total        | 5      | 5      | 5   | 5       | 5            | 5   | 5        | 5     | 5   | 5   | 5        | 55    | 100.00%   |