Supplementary info to

Impact of transformation, photodegradation and interaction with glutaraldehyde on the acute toxicity of the biocide DBNPA in cooling tower water

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Figure S1

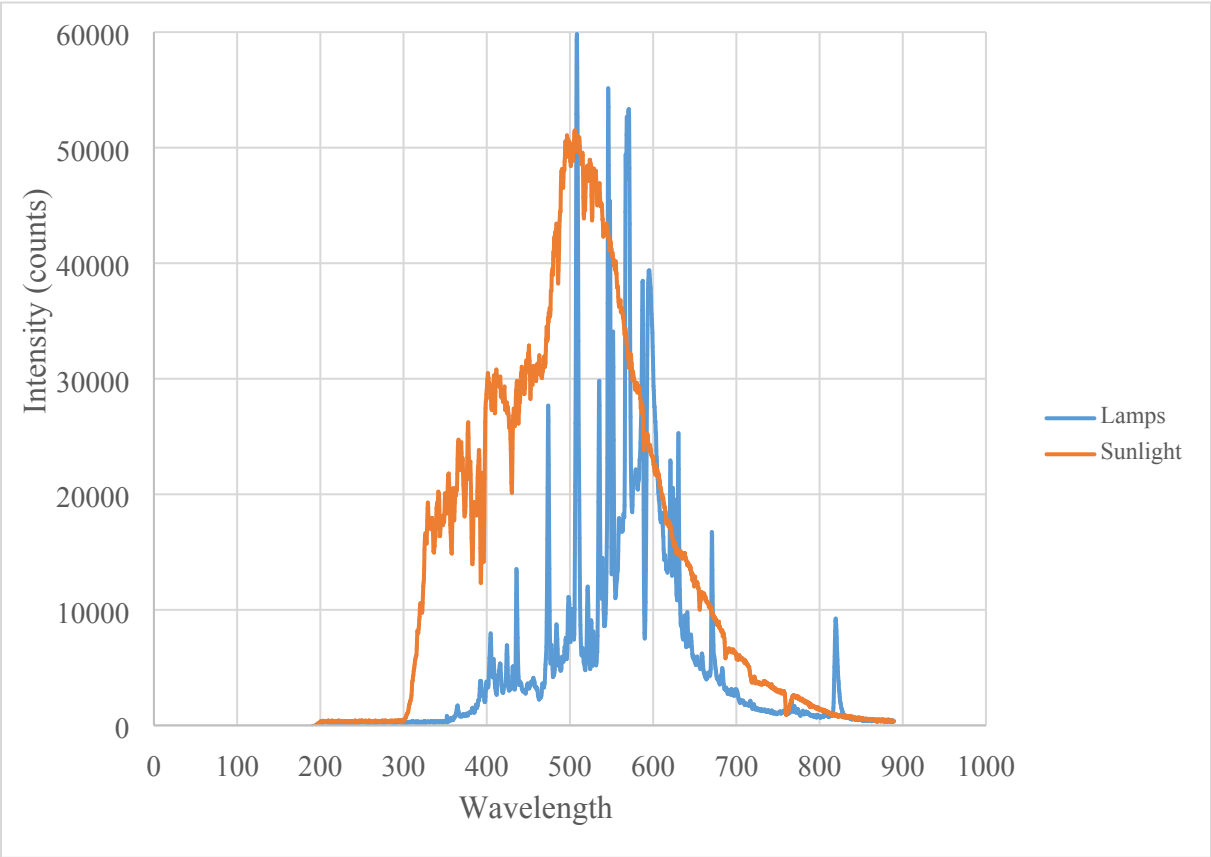


Figure S1. Wavelength intensity distribution for the lamps mimicking sunlight (blue) and actual sunlight (orange). The actual intensities per wavelength are not comparable, since a filter was used to prevent peak overload

Figure S2

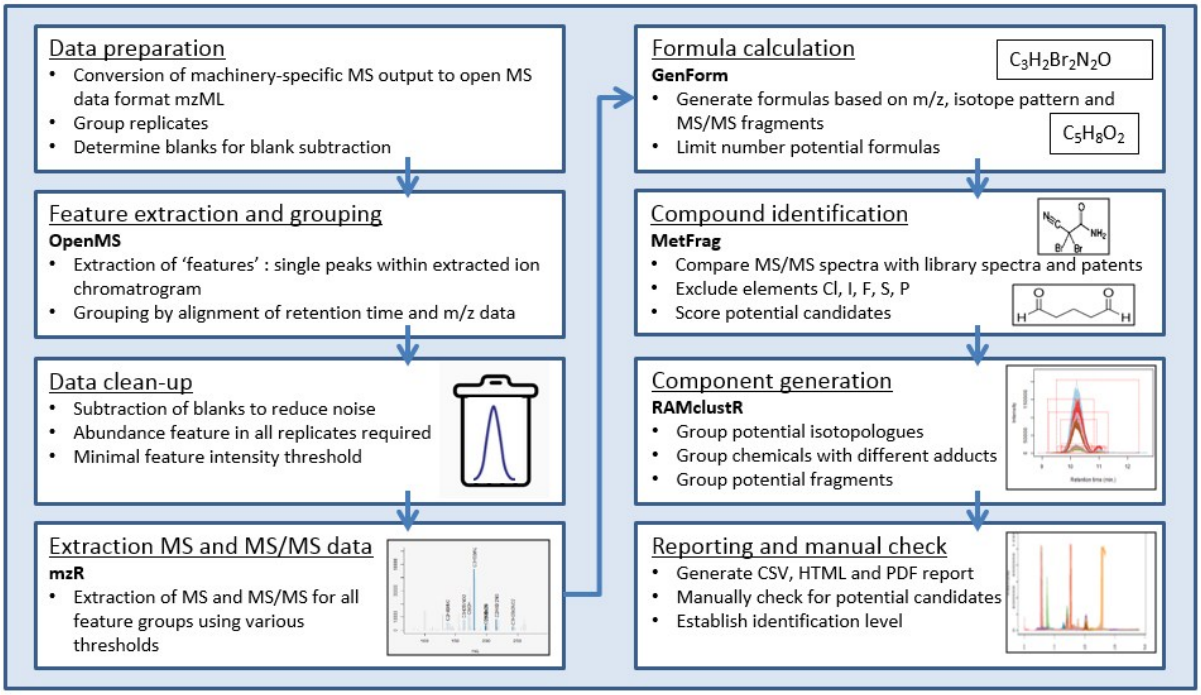


Figure S2. Workflow for non-target data analysis. Workflow-steps are underlined and accompanying software packages are displayed in bold (Wagner et al., 2019a).

Text S1 – The full R-script for data-processing of the non-target screening data for positive ionization

```
## Script automatically generated on Wed May 15 12:16:32 2019

library(patRoon)


# ------------------------# initialization# ------------------------


workPath <- "D:/maXis/Projects/Thomas/Non-target - 15-5/pos"
setwd(workPath)

# Load analysis table
anaInfo <- read.csv("analyses.csv", stringsAsFactors = FALSE, colClasses =
"character")

# Subset for proper calibration
anaInfo_manCal <- anaInfo[1:24,]
anaInfo_autoCal <- anaInfo[25:72,]


# Set to FALSE to skip data pre-treatment
doDataPretreatment <- TRUE
if (doDataPretreatment)
{
    setDAMethod(anaInfo_autoCal, "D:/maXis/Projects/Thomas/Non-target - 15-
5/pos/20190515-pos.m")
    recalibrarateDAFiles(anaInfo_autoCal)
    convertMSFiles(anaInfo = anaInfo,
                   to = "mzML", algorithm = "bruker", centroid = TRUE)
}


# ------------------------# features# ------------------------


# Find all features.
# NOTE: see manual for many more options
fList <- findFeatures(anaInfo, "openms")

# Group and align features between analysis
fGroups <- groupFeatures(fList, "openms")

# Basic rule based filtering
fGroups <- filter(fGroups, preAbsMinIntensity = 100, absMinIntensity = 5000,
                  relMinReplicateAbundance = 1, maxReplicateIntRSD = 0.75,
                  blankThreshold = 5, removeBlanks = TRUE,
                  retentionRange = c(120, Inf), mzRange = NULL)


# ------------------------# annotation# ------------------------


# Retrieve MS peak lists
avgPListParams <- getDefAvgPListParams(clusterMzWindow = 0.002)
plists <- generateMSPeakLists(fGroups, "mzr", maxMSRtWindow = 5, precursorMzWindow
= 4,
                              avgFeatParams = avgPListParams, avgFGroupParams =
avgPListParams)
# uncomment and configure for extra filtering of MS peak lists
# plists <- filter(plists, absMSIntThr = NULL, absMSMSIntThr = NULL, relMSIntThr =
NULL,
```

```
#                   relMSMSIntThr = NULL, topMSPeaks = NULL, topMSMSPeaks = NULL,
#                   deIsotopeMS = FALSE, deIsotopeMSMS = FALSE)

# Calculate formula candidates
formulas <- generateFormulas(fGroups, "genform", plists, relMzDev = 5,
                             adduct = "[M+H]+", elements = "CHNOPBr",
                             calculateFeatures = TRUE, featThreshold = 0.75)

# Find compound structure candidates
compounds <- generateCompounds(fGroups, plists, "metfrag", method = "CL",
dbRelMzDev = 5,
                               fragRelMzDev = 5, fragAbsMzDev = 0.002,
                               adduct = "[M+H]+", database = "pubchem",
maxCandidatesToStop = 2500,
                               extraOpts = list(FilterExcludedElements = c("I",
"Si", "B")))
compounds <- addFormulaScoring(compounds, formulas, TRUE)

# Perform automatic generation of components
components <- generateComponents(fGroups, "ramclustr", ionization = "positive")


# -------------------------# reporting# -------------------------


reportCSV(fGroups, path = "report", reportFeatures = FALSE, formulas = formulas,
          compounds = compounds, compoundsNormalizeScores = "max",
          components = components)

reportPDF(fGroups, path = "report", reportFGroups = TRUE, formulas = formulas,
reportFormulaSpectra = TRUE,
          compounds = compounds, compoundsNormalizeScores = "max",
          components = components, MSPeakLists = plists)

reportMD(fGroups, path = "report", reportPlots = c("chord", "venn", "upset",
"eics", "formulas"), formulas = formulas,
         compounds = compounds, compoundsNormalizeScores = "max",
         components = components, MSPeakLists = plists,
         selfContained = FALSE, openReport = TRUE)
```

Figure S3

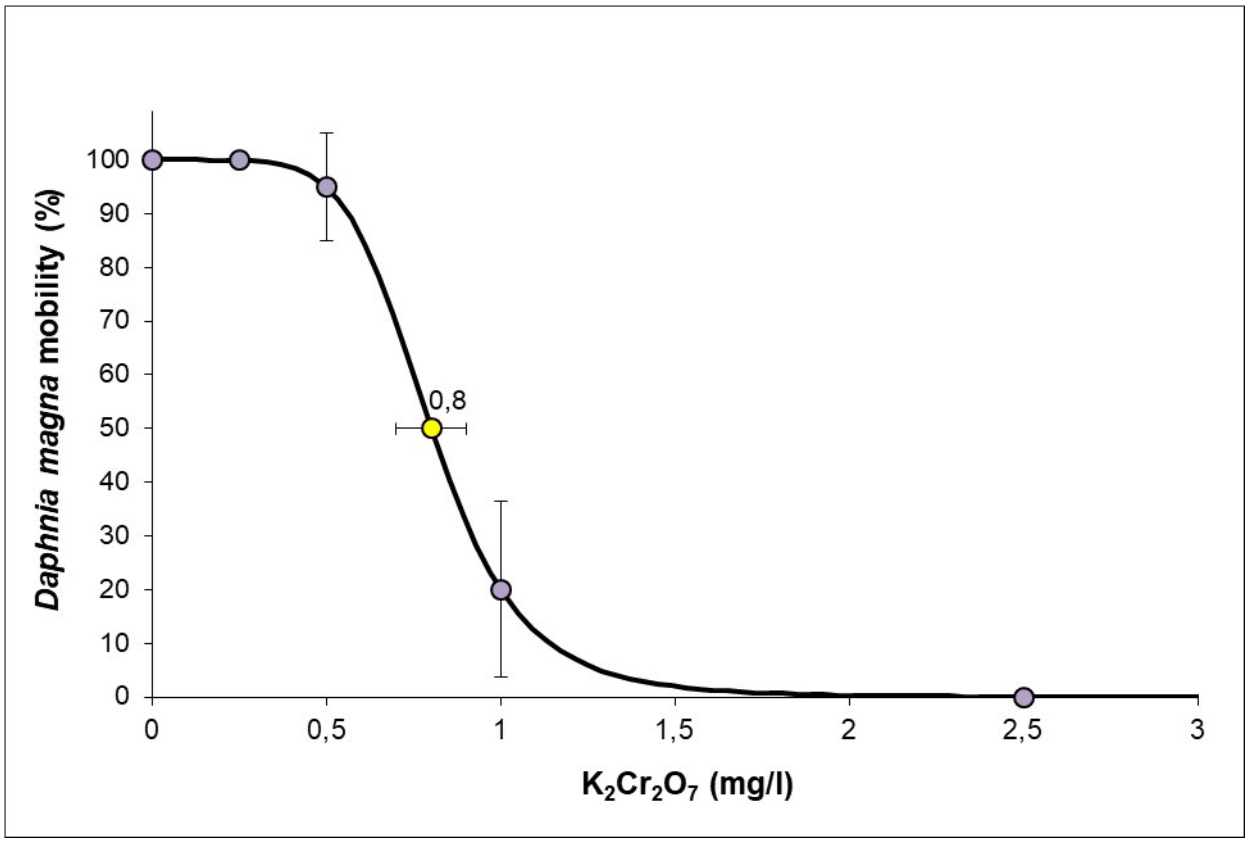


Figure S3. The mobility of Daphnia magna in the presence of different concentrations of $K_2Cr_2O_7$ (purple circles), corresponding model fit (black lines) and LC_{50} (+ 95% confidence interval) (yellow circle).

Table S1.

Table S1. Chemical parameters of test solutions of DBNPA used for the determination of the EC_{50} for Daphnia magna in cooling tower water and ADaM medium

DBNPA concentration (mg/L)	**Cooling tower water**			**ADaM medium**		
	pH	**Temp (°C)**	**DO (mg/L)**	**pH**	**Temp (°C)**	**DO (mg/L)**
0	6.6	20.4	8.94	6.2	20.5	8.94
0.25	6.9	20.4	8.96	6.2	20.5	8.89
0.5	6.7	20.4	8.89	6.2	20.5	8.91
1	6.7	20.5	8.91	6.2	20.5	8.92
2.5	6.9	20.5	8.87	6.2	20.5	8.83
5	6.9	20.5	8.89	6.3	20.5	8.81

Table S2

Table S2. Chemical parameters of test solutions from photodegradation experiments used for Daphnia magna toxicity test.

	pH day 1	pH day 3	pH day 5	T (°C) day 1	T (°C) day 3	T (°C) day 5	DO (mg/L) day 1	DO (mg/L) day 3	DO (mg/L) day 5
CTW-L	7.9	7.9	8.0	19.8	20.5	20.3	8.6	8.6	9.1
CTW-D	7.9	8.0	8.0	19.8	20.2	20.3	8.4	8.6	9.1
DBNPA-L	7.9	8.0	8.0	19.7	20.3	19.9	8.4	8.5	9.0
DBNPA-D	8.0	8.0	8.0	19.9	20.4	20.2	8.7	8.7	8.7
Gluta-L	8.0	8.0	8.0	20.1	20.1	20.0	8.4	8.7	8.7
Gluta-D	8.0	8.1	8.1	20.1	20.3	20.2	8.0	8.7	8.6
G_D-L	7.9	8.0	8.0	20.3	19.9	20.4	8.5	8.7	8.8
G_D-D	7.9	8.1	8.0	20.4	19.9	20.5	8.3	8.7	8.7

Text S2 – Detailed info about TP1-MBNPA

Name: 2-bromo-2-cyanoacetamide (MBNPA)

Origin: Direct transformation product of DBNPA

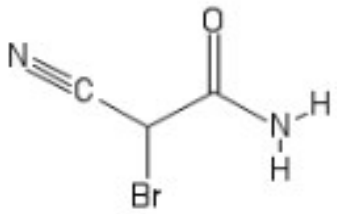
Formula: $C_3H_3BrN_2O$

m/z: 162.95001 (+); 160.9338 (-)

Smiles: C(#N)C(C(=O)N)Br

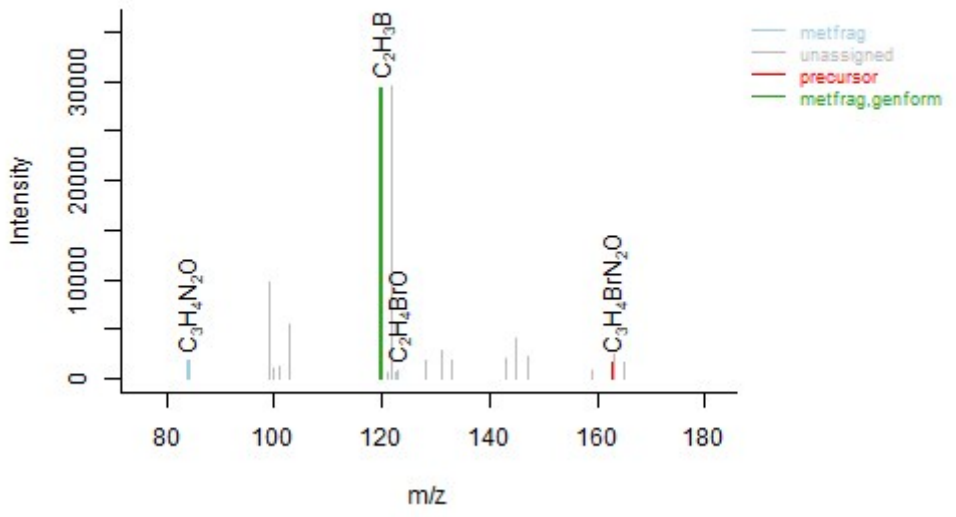
Confidence level: 1

Molecular structure:



Reasoning: Confirmation with reference standard

MS/MS:



Text S3 – Detailed info about TP2-dibromopropanediamide

Name: 2,2-dibromopropanediamide

Origin: Direct transformation product of DBNPA

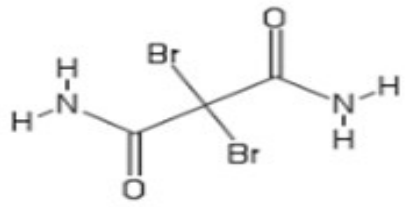
Formula: $C_3H_4Br_2N_2O_2$

m/z: 258.8714 (+)

Smiles: C(=O)(C(C(=O)N)(Br)Br)N

Confidence level: 1

Molecular structure:



Reasoning: Confirmation with reference standard

MS/MS:

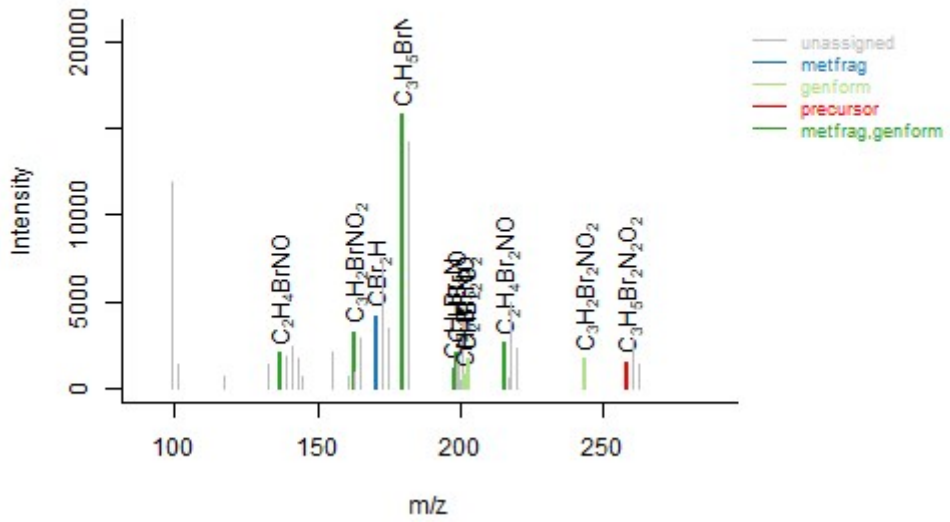


Figure S4.

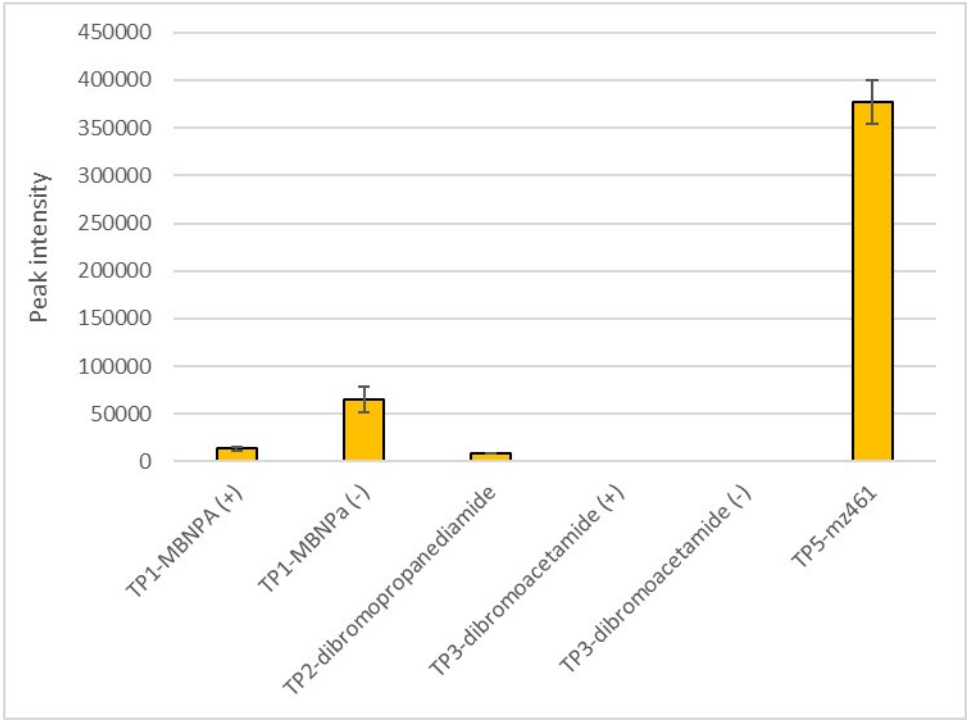


Figure S4. Peak intensities of DBNPA transformation products at t = 1 in the dark.

Figure S5

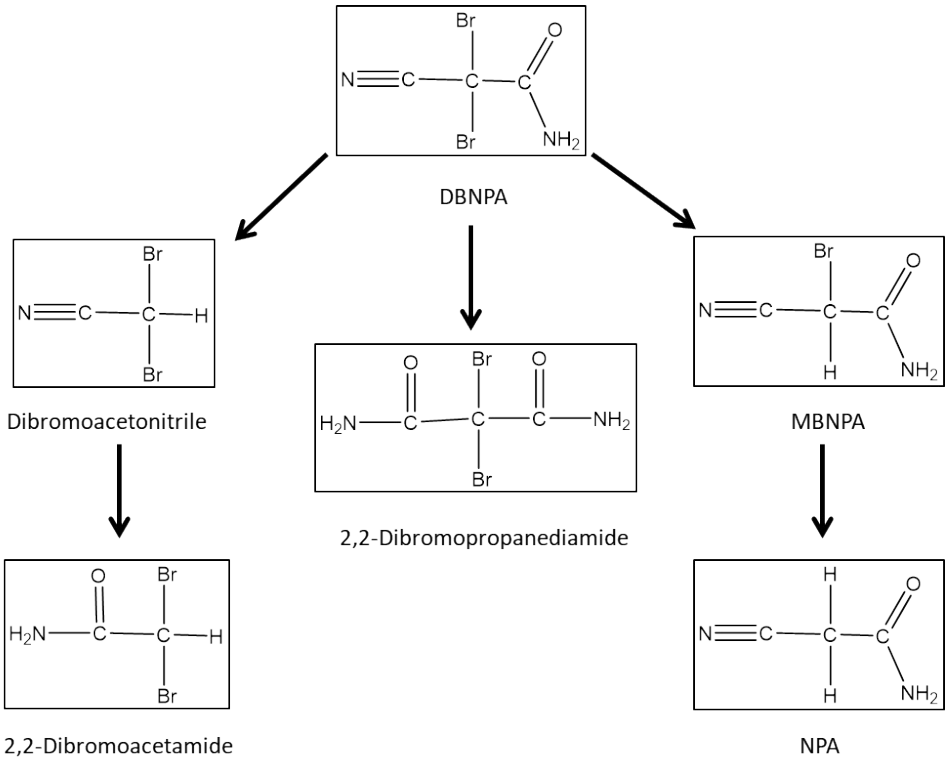


Figure S5. Degradation pathways of DBNPA in the dark.

Text S4 – Detailed info about TP3-dibromoacetamide

Name: 2,2-dibromoacetamide

Origin: Direct transformation product of DBNPA

Formula: $C_2H_3Br_2NO$

m/z: 215.86482 (+); 213.85049 (-)

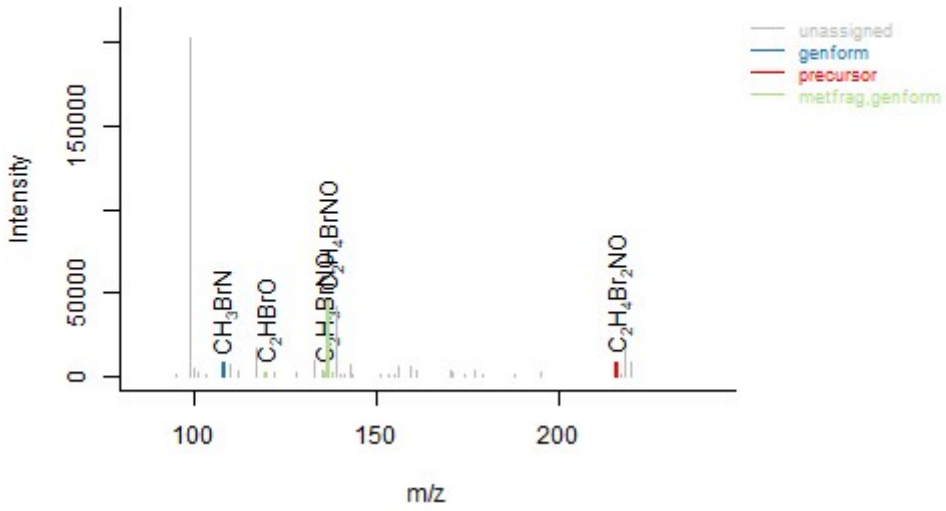
Smiles: C(=O)(C(C(=O)N)(Br)Br)N

Confidence level: 1

Molecular structure:

Reasoning: Confirmation with reference standard

MS/MS:



Text S5. Detailed info about TP4-mz461

Name: -

Origin: Direct transformation product of DBNPA

Formula: $C_{19}H_{37}N_7O_6$

m/z: 460.5483 (+)

Smiles: -

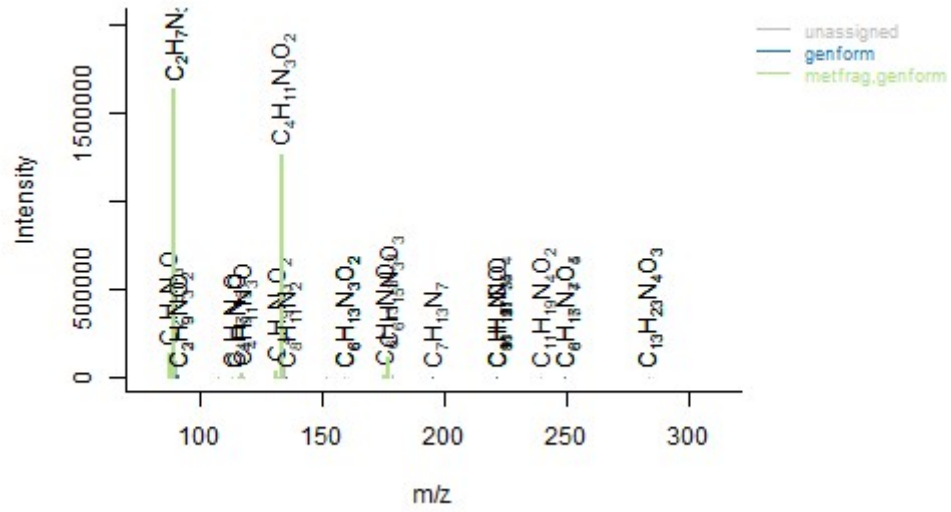
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S6. Detailed info about TP5-mz151

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_2N_7O_2$

m/z: 153.0043 (-)

Smiles: -

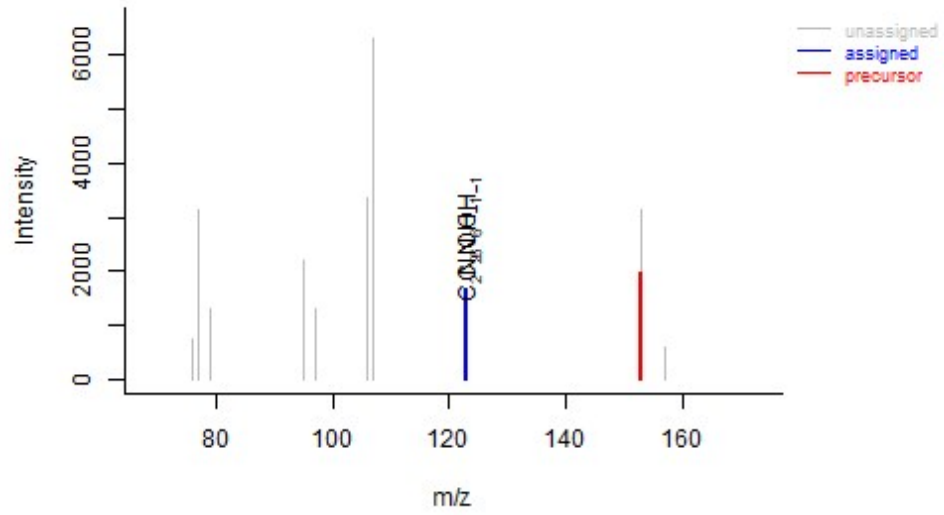
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S7. Detailed info about TP6-mz163

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_2HBrN_2O_2$

m/z: 162.9137 (-)

Smiles: -

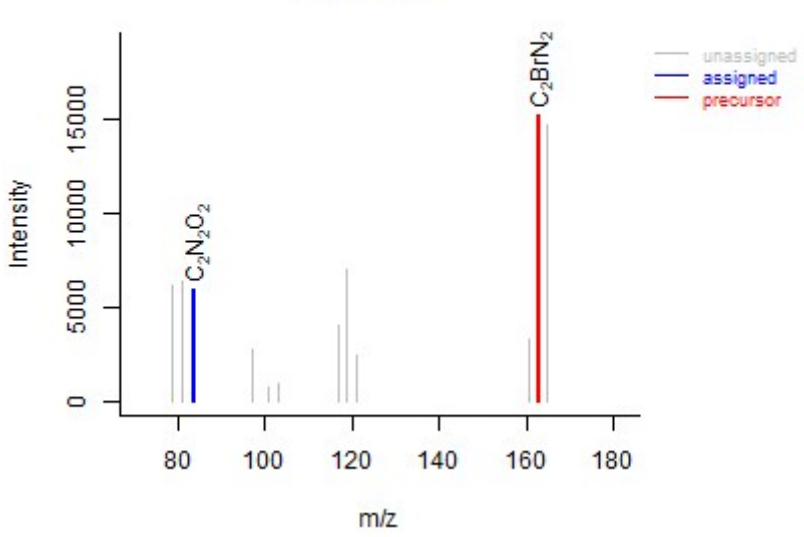
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S8. Detailed info about TP7-mz186

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_8H_5N_5O$

m/z: 186.0409 (-)

Smiles: -

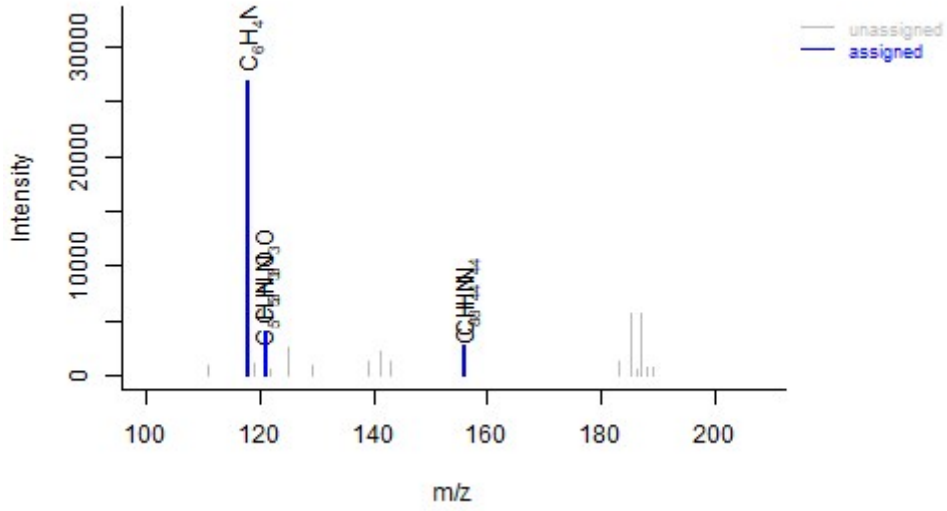
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S9. Detailed info about TP8-mz148

Name: -

Origin: Photodegradation product of DBNPA

Formula: 148.0146 (-)

m/z: $C_6H_3N_3O_2$

Smiles: -

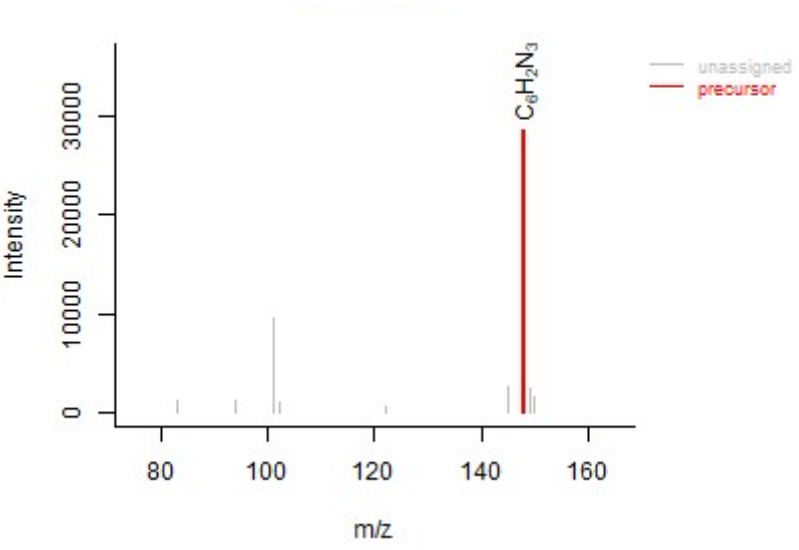
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S10. Detailed info about TP9-mz169

Name: -

Origin: Photodegradation product of DBNPA

Formula: 169.0139 (-)

m/z: $C_7H_6O_5$

Smiles: -

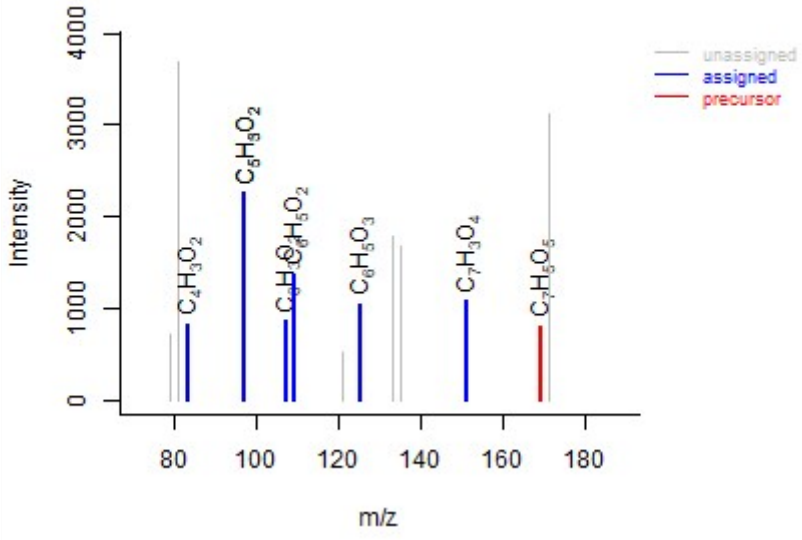
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S11. Detailed info about TP10-mz173

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_{10}H_6O_3$

m/z: 173.0239 (-)

Smiles: -

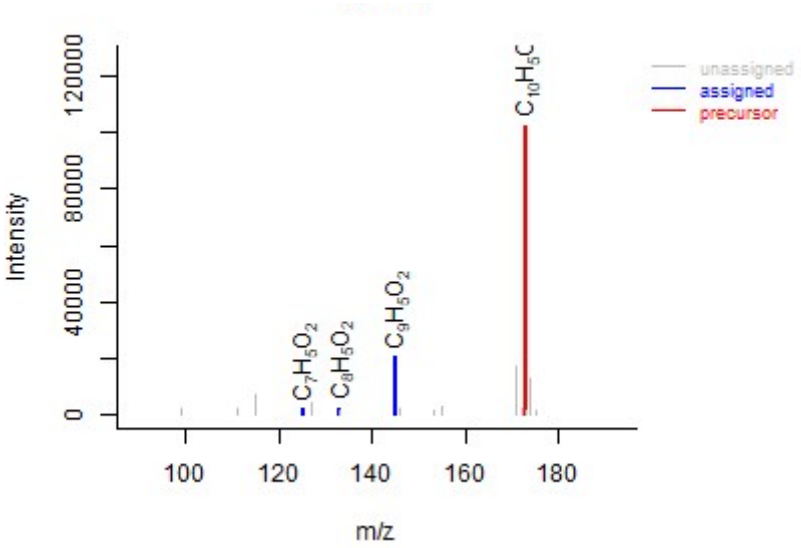
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S12. Detailed info about TP11-mz206 and TP12mz206

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_8H_8N_5O_2$

m/z: 206.0666 (+)

Smiles: -

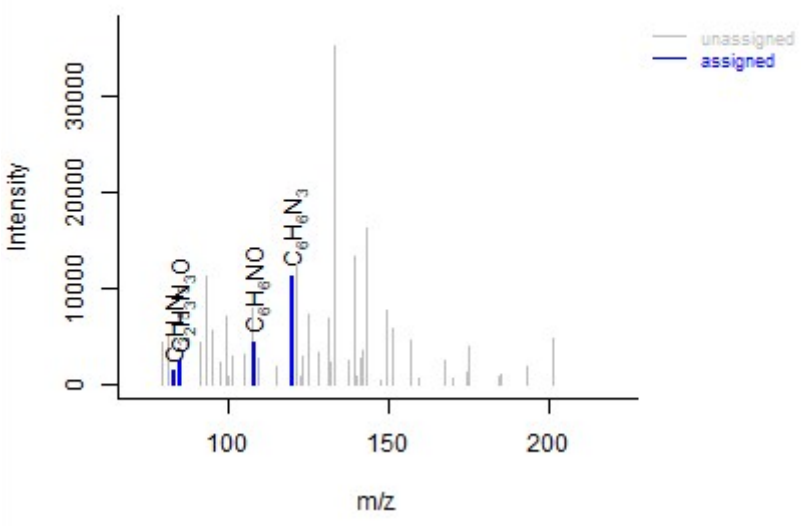
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S13. Detailed info about TP13-mz344

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_{12}H_{21}N_7O_5$

m/z: 344.1688 (+)

Smiles: -

Confidence level: 5

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:

No MS/MS was available

Text S14. Detailed info about TP14-mz170

Name: -

Origin: Photodegradation product of DBNPA

Formula: $C_6H_7N_3O_3$

m/z: 170.0559 (+) ; 168.0411 (-)

Smiles: -

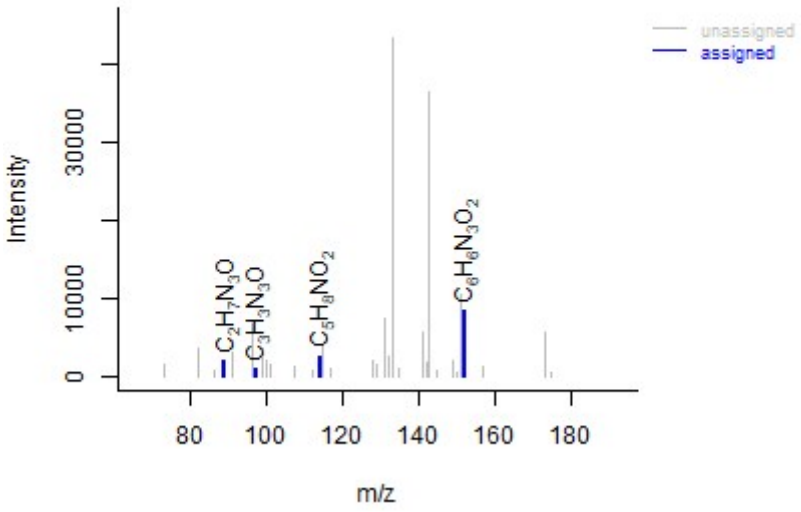
Confidence level: 4

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S15. **Detailed info about GIP1**

Name: -

Origin: Interaction product between DBNPA and glutaraldehyde

Formula: $C_6H_9NO_2$

m/z: 128.07079 (+)

Smiles: -

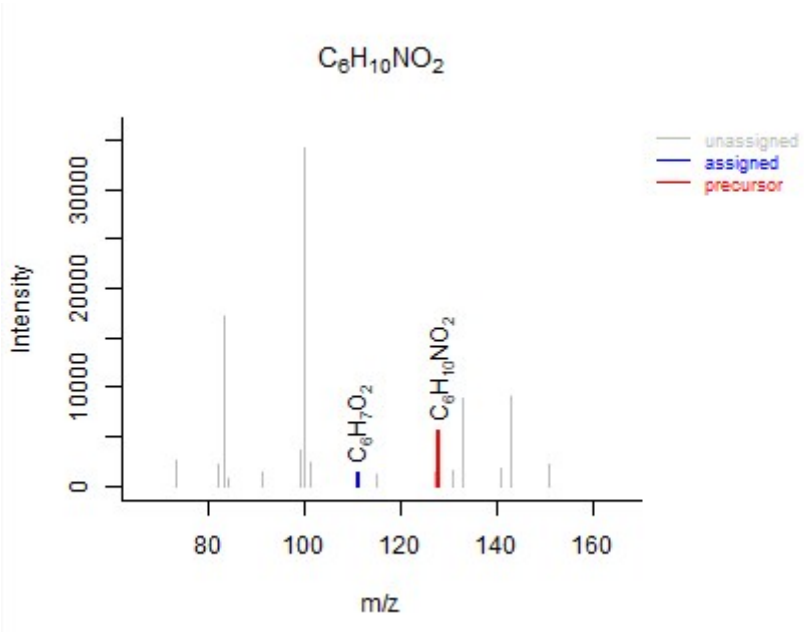
Confidence level: 5

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S16. **Detailed info about GIP2**

Name: -

Origin: Interaction product between DBNPA and glutaraldehyde

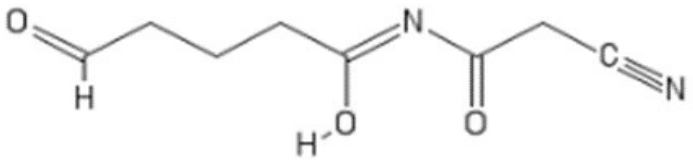
Formula: $C_8H_{10}N_2O_3$

m/z: 181.06119 (-), 183.07633 (+)

Smiles: -

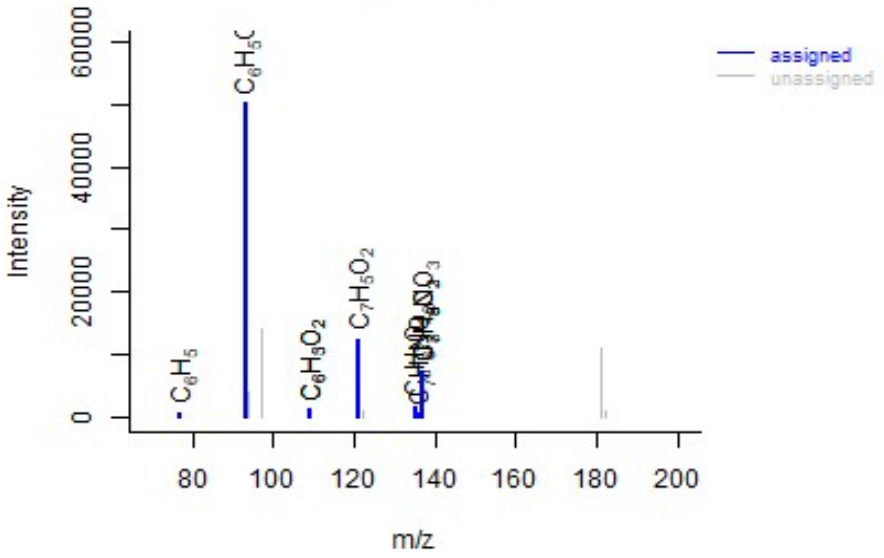
Confidence level: 3

Molecular structure:



Reasoning: Molecular formula; knowledge on parent compounds formula and structure; knowledge on parent compound's chemical behaviour. Wagner et al. 2019b

MS/MS:



Text S17. **Detailed info about GIP3**

Name: -

Origin: Interaction product between DBNPA and glutaraldehyde

Formula: $C_8H_{12}N_2O_3$

m/z: 183.07678 (-), 185.09193 (+)

Smiles: -

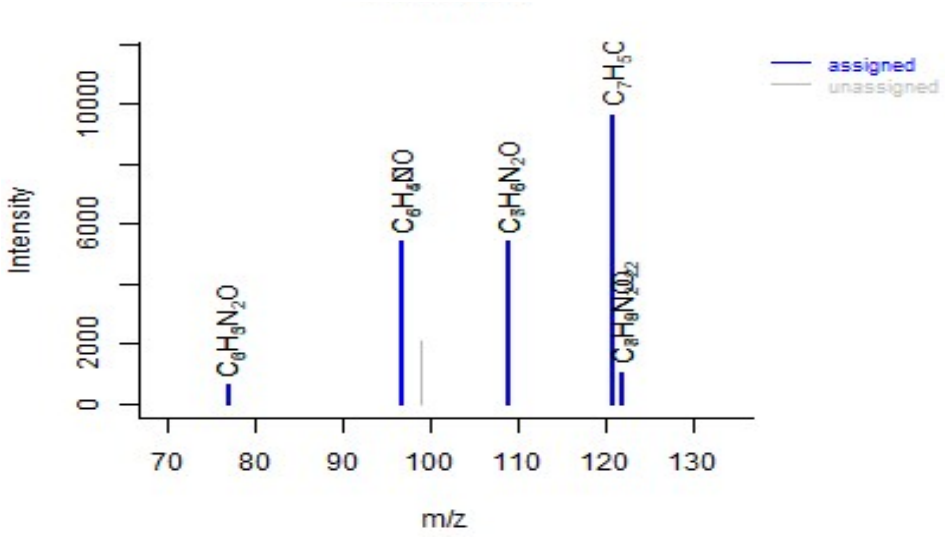
Confidence level: 5

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:



Text S18. **Detailed info about GIP4**

Name: -

Origin: Interaction product between DBNPA and glutaraldehyde

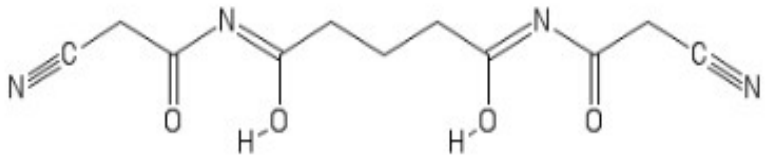
Formula: $C_{11}H_{12}N_4O_4$

m/z: 263.0777 (-), 265.0931 (+)

Smiles: -

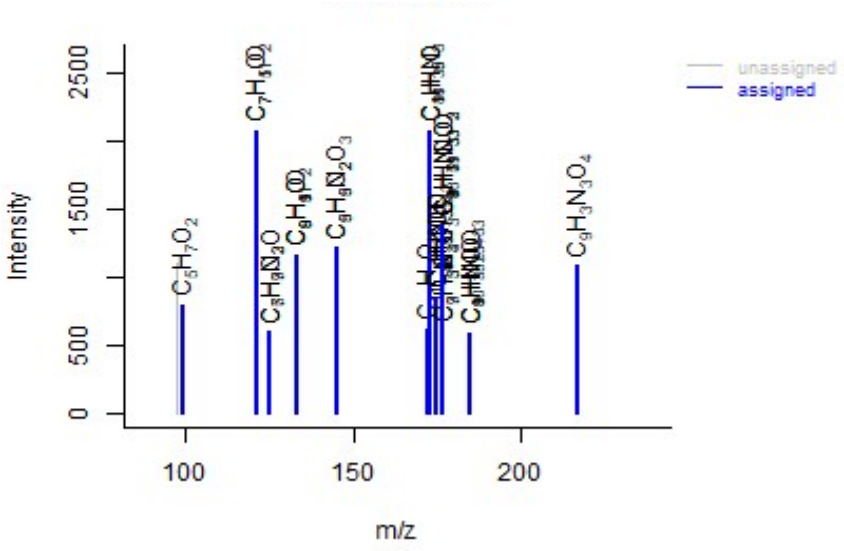
Confidence level: 3

Molecular structure:



Reasoning: Molecular formula; knowledge on parent compounds formula and structure; knowledge on parent compound's chemical behaviour

MS/MS:



Text S19. **Detailed info about GIP5**

Name: -

Origin: Interaction product between DBNPA and glutaraldehyde

Formula: $C_{17}H_{30}N_4O_6$

m/z: 385.20753 (-)

Smiles: -

Confidence level: 5

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS:

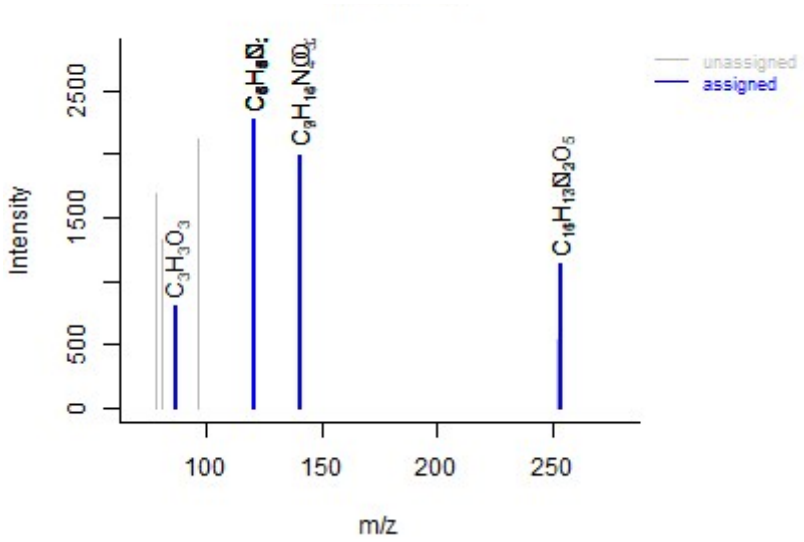


Figure S6 Peak intensity of GIP2

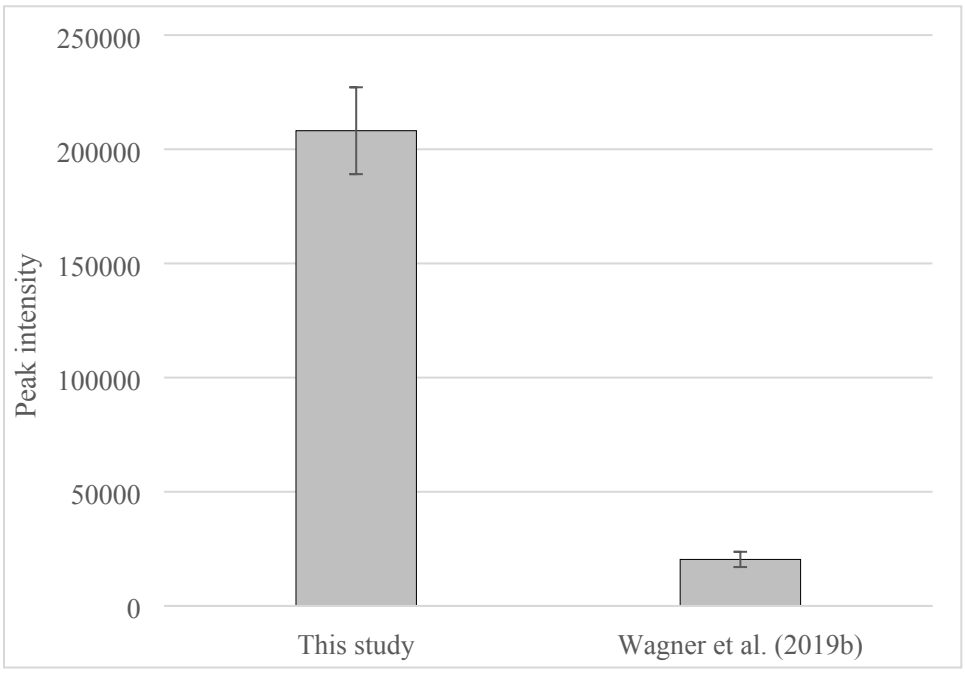


Figure S6. Peak intensity of GIP2 (fragment m/z 183.07633) in this study and in Wagner et al. (2019b)

Text S20. **Detailed info about GIP6**

Name: -

Origin: Interaction product between DBNPA and glutaraldehyde

Formula: $C_{11}H_{11}N_3O_5$

m/z: 264.06206 (-)

Smiles: -

Confidence level: 5

Molecular structure:

No molecular structure was proposed

Reasoning: -

MS/MS: