



## Data Collection Worksheet

**Please Note:** The Data Collection Worksheet (DCW) is a tool to aid integration of a PhenX protocol into a study. The PhenX DCW is not designed to be a data collection instrument. Investigators will need to decide the best way to collect data for the PhenX protocol in their study. Variables captured in the DCW, along with variable names and unique PhenX variable identifiers, are included in the PhenX Data Dictionary (DD) files.

## Biochemical Validation of Cannabis Consumption in Plasma and Urine by Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS)

The following is a summary of the protocol from Klawitter et al. (2017). Medical Cannabis Use, Polysubstance Use, and Recovery (MCPR) Working Group (WG) encourages interested investigators to identify laboratories that offer this assay and to ask about any specific requirements for biospecimen collection and transport. Investigators are encouraged to review the manuscript for more details.

### Reagents

See Klawitter et al. (2017) for a description of the 11 cannabis analytes, internal standards, buffers, and where they were purchased.

### Calibration Curves

For both urine and plasma, calibration standards for all 11 analytes were prepared by diluting 20  $\mu$ L of 10X stock solutions into 180  $\mu$ L of matrix resulting in following concentrations:

- 0.39 ng/mL
- 0.78 ng/mL
- 1.56 ng/mL
- 3.13 ng/mL
- 6.25 ng/mL
- 12.5 ng/mL
- 25 ng/mL
- 50 ng/mL
- 100 ng/mL

- 200 ng/mL
- 400 ng/mL

The authors note that the calibration curves for THC-C-gluc needs to be 5x higher to account for its higher concentration in urine

### Sample Extraction

Aliquots are transferred to 1.5 mL low-binding polypropylene vials, vortexed, centrifuged and added to high-performance liquid chromatography (HPLC) autosampler vials.

### Analysis

Investigators are encouraged to review Klawitter et al. (2017) for the details concerning the detection of the 11 cannabis analytes according to HPLC-tandem mass spectrometry. The WG notes that over time, additional analytes may become recognized and available.

The MCPR WG notes that there are other HPLC tandem mass spectrometry procedures and instruments that are appropriate for the measurement of cannabis analytes in urine and plasma. Once an assay is chosen for a particular study, the WG recommends that no changes in the protocol be made over the course of the study. To aid comparability, the WG recommends that the investigator record the make and manufacturer of equipment used and the repeatability and coefficients of variation for the assay. If using a commercial provider, then that provider should be used for the duration of the study, whenever possible.

### Results

1.  $\Delta^9$ -tetrahydrocannabinol (THC): \_\_\_\_\_ ng/mL
2. 11-hydroxy- $\Delta^9$ -tetrahydrocannabinol (11OH-THC): \_\_\_\_\_ ng/mL
3. 11-nor- $\Delta^9$ -tetrahydrocannabinol-9-carboxylic acid glucuronide (THC-C-gluc): \_\_\_\_\_ ng/mL
4. cannabinol (CBN): \_\_\_\_\_ ng/mL
5. cannabidiol (CBD): \_\_\_\_\_ ng/mL
6. Cannabidiol (CBD): \_\_\_\_\_ ng/mL
7. cannabichromene (CBC): \_\_\_\_\_ ng/mL
8. cannabigerol (CBG): \_\_\_\_\_ ng/mL
9.  $\Delta^9$ -tetrahydrocannabivarin (THCV): \_\_\_\_\_ ng/mL
10. 11-nor- $\Delta^9$ -tetrahydrocannabinol-9-carboxylic acid (THC-COOH): \_\_\_\_\_ ng/mL
11. 11-nor-9-carboxy- $\Delta^9$ -tetrahydrocannabivarin (THCV-COOH): \_\_\_\_\_ ng/mL

Protocol source: <https://www.phenxtoolkit.org/protocols/view/511401>