Introduction
Synthetic cannabinoids (SC) pose a great challenge to practitioners in the forensic field. Over the last years, SC available on the market have undergone significant structural changes making immunochemical testing unsuitable. Accordingly, MS methods have become the gold standard for analysis of SC in biological specimens but require frequent adaption to include newly emerged SC.

Since most SC are metabolized extensively prior to renal excretion, metabolite identification is invaluable for urine analysis. After conjugate cleavage, the main phase I metabolites are suitable targets. Consequently, the metabolism of new SC needs to be known prior to updating analytical methods. In cases where no authentic human sample material with confirmed uptake of the parent compound can serve as a plausibility check and may help in estimating the time of the last drug uptake.

II. Data Evaluation
1.) Manual Data Evaluation:
- Evaluation of Extracted Ion Chromatograms (EIC)
- Evaluation of b/cCID data based on characteristic fragments
- Mass defect filtering
With manual data evaluation 25 possible metabolites could be identified.

2.) Automatic Data Evaluation:
- MassMetaSite software was used to analyze the LC-MS/MS datasets of the incubation, revealing 10 metabolites with at least two fragment ions each.

3.) Identification of the main in vitro phase I metabolites
- By manual data evaluation 5 additional metabolites at low intensities could be identified. However, the main in vitro metabolites described in the literature[1,2] were identified by the software.

III. Generation / Update of the Screening Method
The name, formula, retention time and the accurate mass of the precursor and fragment ions of potential phase I metabolites are added to a .csv file. Subsequently, a TASQ method for screening authentic samples is generated from this .csv file.

V. Additional Screening of Positive Authentic Samples
Positive urine samples are screened after conjugate cleavage in analogy to the in vivo sample to identify the predominant in vivo phase I metabolites and to screen for further metabolites. Additionally, samples are screened for phase II metabolites after urine precipitation allowing to update methods omitting conjugate cleavage in the sample preparation process.

IV. Analysis of Routine Case Work Samples
Reports are generated by TASQ software for each sample listing every hit. The hits are rated based on their mass and retention time error, m/z sigma value and the presence of qualifier ions (MRM-Score).

VI. Additional Screening Results
- 1.) Without Cleavage of Glucuronides
- 2.) After Cleavage of Glucuronides
Despite varying relative abundances of the detected metabolites, the in vitro and in vivo data showed good agreement with respect to the MDMB-CHMICA metabolites chosen as target candidates. Consequently, a new metabolite was identified in the in vivo samples so a total of 42 metabolites of MDMB-CHMICA (phase I and II) could be added to the database.

Conclusions
Using MassMetaSite software and the described workflow proved to be a suitable, less laborious and time consuming procedure compared to manual data evaluation. The here described approach can be helpful for updating screening methods with metabolite information. This is necessary whenever dealing with analytes that are extensively metabolized such as SC. In other cases identification of metabolites along with the parent compound can serve as a plausibility check and may help in estimating the time of the last drug uptake.

Acknowledgement
The project was funded by the Prevention of and Fight against Crime program of the European Commission [JUST/2013/ISEC/DRUGS/AG/6/9421], and the Deutsche Forschungsgemeinschaft (INST 38/92-3 FUGG).