

XXIV. GTFCh-Symposium - Poster

P01 *In vitro* neurotransmitter reuptake inhibition and forensic case series in Sweden of the synthetic cathinones 2-, 3-, and 4-Me-alpha-PiHP

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Aims: This study characterized the inhibitory potential of the recently emerged synthetic cathinones, 2-, 3-, and 4-Me-alpha-PiHP (structural isomers) at the dopamine, norepinephrine, and serotonin transporters (DAT, NET, SERT). Further, we report on their detections in Sweden based on seizure and toxicologic casework data by the National Forensic Centre (NFC) and the National Board of Forensic Medicine (Rättsmedicinalverket; RMV). **Methods:** Cells expressing either human DAT, NET, or SERT were incubated following a semi-automated experimental protocol (14 concentration points). Dose response curves and mean inhibitory concentrations (IC₅₀) were determined. The resulting IC₅₀ values were used to calculate DAT/SERT inhibition ratios to estimate the abuse liability. **Results and Discussion:** The three structural isomers showed similar inhibitory potentials, with clear selectivity for DAT over NET and SERT. The IC₅₀s at DAT were extremely low with 0.5 nM for 4-Me-alpha-PiHP (~250x IC_{50cocaine}), 1.7 nM for 3-Me-alpha-PiHP (~80x IC_{50cocaine}), and 7.6 nM for 2-Me-alpha-PiHP (~18x IC_{50cocaine}). Elevated DAT/SERT ratios imply a high abuse liability for all three synthetic cathinones. NFC reported the emergence of 4-Me-alpha-PiHP in 2020, with a total of 36 seizures (2020 - 2022). 2-Me-alpha-PiHP emerged in 2022 (8 seizures) and was seized additional 36 times in 2023. Similarly, 4-Me-alpha-PiHP was detected 24 times in the years 2020 – 2021 in forensic toxicological casework samples, followed by 5 detections of 2-Me-alpha-PiHP in 2023. Recently, 3-Me-alpha-PiHP has been detected in 3 tox cases in 2024. These synthetic cathinones have been detected in driving under the influence, violent crimes, and autopsy cases, often in combination with further drugs of abuse, including other synthetic cathinones. **Conclusion:** Current market observations imply that 2-, 3-, and 4-methyl-alpha-PiHP currently are a regional phenomenon in Sweden. The close monitoring of these potent stimulants with high abuse liability remains important nonetheless, as they might emerge further into Europe.

P02 Cross-reactivity study on old and new benzodiazepines in the CEDIA™ benzodiazepine assay

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Aims: The cross-reactivity (CR) of many new designer benzodiazepines and their metabolites is unknown for most benzodiazepine urine immunoassays. Furthermore, these substances often are not included in chromatographic methods. This may lead to false negative confirmation analysis. We investigated the CR of all benzodiazepines included in our confirmatory benzodiazepine UPLC-MS/MS method in the CEDIA™ benzodiazepine assay. **Methods:** The study was performed on an Olympus AU 680 (Beckmann Coulter) with the CEDIA™ benzodiazepine assay (ThermoFisher) and online hydrolysis. Saline was spiked with certified reference substances at a concentration of 2000 ng/mL, serial dilutions (1:2 v/v) were made down to 62.5 ng/mL. CR was calculated at 125 and 250 ng/mL. The following benzodiazepines were tested: 1-Hydroxy-Alprazolam, 1-Hydroxy-Alprazolam-glucuronide, 1-Hydroxy-Midazolam, 1-Hydroxy-Midazolam-glucuronide, 1-Hydroxy-Triazolam, 2-Hydroxy-Ethyl-Flurazepam, 3-Hydroxy-Bromazepam, 3-Hydroxy-Flubromazepam, 3-Hydroxy-Phenazepam, 3-Hydroxy-Przepam, 4-Chlorodiazepam, 7-Aminoclonazepam, 7-Aminoflunitrazepam, 7-Aminonimetazepam, 7-Aminonitrazepam, Adinazolam, Alprazolam, Bentazepam, Bromazepam, Bromazolam, Brotizolam, Chlordiazepoxid, Clobazam, Clonazepam, Clonazolam, Cloniprazepam, Delorazepam, Demoxepam, Desalkylflurazepam, Deschloroetizolam, Desmethylflunitrazepam, Diazepam, Diclazepam, Difludiazepam, Estazolam, Etizolam, Flualprazolam, Flubromazepam, Flubromazolam, Flubrotizolam, Fluclotizolam, Flunitrazepam, Flunitrazolam, Flurazepam, Flutazolam, Halazepam, Ketazolam, Lorazepam, Lorazepam-glucuronide, Lormetazepam, Meclonazepam, Metizolam, Midazolam, N-Desmethylclobazam, Nifoxipam, Nimetazepam, Nitrazepam, Nitrazolam, Norchlordiazepoxid, Nordiazepam, Nortetrazepam, Oxazepam, Oxazepam-glucuronide, Phenazepam, Phenazolam, Pivoxazepam, Przepam, Pyrazolam, Rilamazepam, Temazepam, Temazepam-glucuronide, Tetrazepam, Thionordiazepam, Tofisopam, Triazolam. **Results:** Of 75 benzodiazepines tested, only Pivoxazepam, Flutazolam and Tofisopam revealed no CR in the CEDIA™. CR at 125 ng/mL ranged from 6 % to 416 % and at 250 ng/mL from 14 % to ≥ 800 %. Forty benzodiazepines showed increasing CR with increasing concentrations, 24 showed decreasing and 9 linear CR in the CEDIA™. **Discussion and Conclusion:** 72 (96 %) of the 75 tested benzodiazepines revealed CR > 6 % in the CEDIA™ benzodiazepine assay and therefore could be expected for positive confirmation analysis of immunoassay positive urine samples. A limitation of our study is, that several designer benzodiazepines and their metabolites could not be tested, as they were not available as certified reference material.

P03 Qualitative and semi-quantitative evaluation of an untargeted, LC-HRMS screening method in 500 authentic, whole-blood forensic toxicology routine cases

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Aims: In forensic toxicology (FT), comprehensive screening for drugs (of abuse) is crucial but challenging. Thus, an established LC-low resolution (LR)MS urine screening should be replaced by a LC-HRMS blood screening, allowing selective and sensitive detection of over 2500

substances, including challenging analytes like THC, its metabolites and NPS. Additionally, this LC-HRMS screening will allow semi-quantitative determination of routinely detected drugs (of abuse). **Methods:** After protein precipitation, the LC-HRMS TargetScreener[®] vendor platform (from Bruker, combining Impact II[®] and TASQ[®] software) was used to measure 500 authentic whole blood samples in ESI⁺ and data-independent acquisition (DIA) mode, using a RP C18 column. Results were qualitatively compared to the LC-LRMS ToxTyper[®] platform. Semi-quantification (using eight calibration points) was validated according to ANSI and GTFCh guidelines, covering 81 common drugs (of abuse). **Results and Discussion:** The LC-HRMS screening accurately identified 368 out of 460 positive cases (80 %) and all 40 negative cases (100 %). After further quantification, the non-identified substances showed blood concentrations below the methods LOQ/LOD. Cases with known THC and/or THC-COOH concentrations above 3.0 ng/mL and 8.0 ng/mL, respectively, were reliably detected, as were six cases in which NPS were suspected according to the HighResNPS database (4-CBC, 2x 4-Cl-PPP, 2x 4F-PV8, N-Methyl-1-phenethylamin and Medazepam). Validation of semi-quantification was successful (accuracy CV $\leq \pm 30$ % and precision CV ≤ 30 %) for 67 drugs (of abuse), with low analyte concentrations proving difficult. **Conclusion:** The new LC-HRMS blood screening shows comparable, qualitative results to the current LC-LRMS urine screening. Additionally, the LC-HRMS is able to screen for substances like THC, its metabolites and NPS. Semi-quantification of 67 drugs (of abuse) in blood was successfully implemented, facilitating decisions regarding further (targeted) analytical steps. Finally, this untargeted, DIA LC-HRMS screening not only allows further (simultaneous) routine and research applications, but also retrospective screening for newly emerging substances.

P04 Non-invasive blood alcohol monitoring with NIR spectroscopy

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Aims: A significant demand for reliable blood alcohol concentration (BAC) testing exist from health, legal and personal perspectives. This study explores the feasibility of non-invasive blood alcohol monitoring (NIBAM) using diffusive reflectance near-infrared (DR-NIR) spectroscopy. The objective was to develop a reliable and non-invasive alternative to traditional blood and breath alcohol tests, focusing on its application in clinical and automotive contexts. **Methods:** A high-throughput Fourier-transform infrared (FTIR) spectrometer with a custom fiber probe was designed. The setup was validated using *in-vitro* skin proxies doped with ethanol. A non-clinical trial involving 8 subjects was conducted, measuring blood alcohol concentration (BAC) and breath alcohol concentration (BrAC) alongside NIR spectroscopy. Chemometric models were trained to correlate NIR data with BAC and classify intoxication levels based on legal limits. **Results and Discussion:** *In-vitro* validation achieved an $R^2 = 0.95$ and a RMSE_{CV} of 17.3 mg/dL (0.22‰) for ethanol detection. The *in-vivo* study demonstrated a strong correlation between true and predicted BAC values with an RMSE_{CV} of 16 mg/dL (0.2‰). Classification models accurately distinguished BAC levels above and below 0.5‰ with high specificity. Performance varied between subjects, highlighting the influence of individual and day-to-day variations, which will require further optimization. **Conclusion:** The performed *in-vivo* study confirms the feasibility of NIBAM using DR-NIR spectroscopy, achieving reliable results. The approach offers significant potential for safer, non-invasive alcohol monitoring in clinical, personal, and automotive applications. Future efforts will focus on enhancing chemometric models and miniaturizing spectrometer hardware for broader accessibility.

P05 LSD in urine: New potential metabolite and biomarkers

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Aims: LSD represents a major analytical challenge in clinical/forensic toxicology, given its low concentrations and rapid metabolism. Systematic (untargeted) metabolism studies in human matrices are scarce. Previous untargeted analysis of urine samples (0-8 h post administration) from a placebo-controlled, crossover LSD (200 µg) administration study in humans revealed several interesting, not yet identified, features indicative of LSD consumption. This project aims to further characterize these features, in particular, a potential unknown LSD metabolite with a mass-to-charge ratio of 400.2322. **Methods:** Six LSD-positive routine cases were protein precipitated using three different solvents (methanol, methanol:acetone 9:1 v/v, acetonitrile). In addition, further urine samples from the placebo-controlled study, collected during three time periods (0-8 h, 8-16 h, and 16-24 h), were analyzed using liquid chromatography high-resolution mass spectrometry (RP, ESI+, DDA, Sciex 6600). The data was processed and evaluated with a targeted approach using MultiQuant[®], PeakView[®], and R (version 4.4.1). **Results and Discussion:** The feature with m/z 400.2322 was detected in all routine cases, independent of the extraction solvent, and was present in 87 % of LSD-positive study samples, but not detected in any placebo samples. Significant time-dependent changes ($p < 0.05$, Wilcoxon, Holm correction for multiple testing) were observed similar to those of the known LSD-metabolites 2-oxo-3-hydroxy-LSD and hydroxy-LSD glucuronide. The highest normalized areas were observed in time period three (8-16 h). Eleven additional features (out of 46 detected) were detected in the routine cases and showed time-dependent changes in the study samples. Of those, three were tentatively identified based on their MS and MS/MS information, as pyroglutamic acid, dihydrocortisol, and corticosterone sulfate. **Conclusion:** Feature 400.2322 m/z , as well as the other tentatively identified features, appear to be reliable and promising new LSD ingestion markers, facilitating routine analysis in forensic toxicology. More efforts (e.g., different ionization techniques) are needed to elucidate and confirm their chemical structure.

P06 The mystery of post-mortem gamma-hydroxybutyrate formation - method development and validation for the detection of endogenous GHB and its metabolites

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Aims: Interpretation of gamma-hydroxybutyrate (GHB) results in post-mortem samples is a challenge: A possible increase due to post-mortem development in the body as well as *in vitro* must be taken into account. Whether endogenous substances that are metabolically related to GHB may have an influence in post-mortem generation has not been investigated sufficiently. The aim was to investigate GHB, GBL and eight other endogenous substances related to GHB in one method. **Methods:** A method was developed and validated to detect GHB, γ -butyrolactone, succinic semialdehyde, γ -aminobutyric acid, putrescine, α -hydroxybutyrate (AHB), β -hydroxybutyrate (BHB), L-glutamic acid, succinic acid, and GHB-glucuronide. Extraction was carried out using precipitation with acetonitrile and methanol (85:15 v/v) and additionally SPE (STRATA-X, Phenomenex). The extracts were measured using LC-MS/MS within a 33-minute

run on a Waters Atlantis Premier BEH C18 AX 2.5 μ m with a gradient (mobile phase A: water with 0.1 % FA; mobile phase B: acetonitrile with 0.1 % FA). **Results and Discussion:** For calibration and quality controls human whole blood was used. The endogenous concentrations were subtracted using a matrix blank. Limits of detection (LOD) and limits of quantification (LOQ) of 0.5 mg/L for all analytes were determined. The calibration ranges started at 0.5 mg/L and ranged up to 75 mg/L, depending on the analyte. Linear regression could be used for most substances, except BHB, GHB, putrescine and succinic acid. Accuracy and precision were < 10 % at all concentrations and for all analytes. **Conclusion:** An LC-MS/MS method was developed that can be used to detect GHB and nine related substances which could potentially contribute to the increase in GHB concentrations in post-mortem samples. In order to test the applicability of this method and to analyse whether the nine endogenous substances selected here actually have an influence on the GHB concentration, the method will be used to analyse real post-mortem samples.

P07 Detection of synthetic cannabinoids in dental hard tissue after single exposure: a promising approach for postmortem toxicology

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Aims: Postmortem toxicological analysis often faces challenges due to tissue decomposition and degradation, which can hinder the collection of conventional matrices like blood or urine. Therefore, developing methods for alternative postmortem matrices is essential. Dental hard tissue shows promising potential in forensic toxicology. Previous studies have explored the detection of various drugs in postmortem dental hard tissue of authentic intoxication cases. However, these studies were not suitable to examine whether a single inhalation of a drug is detectable in teeth. Therefore, the pig's tooth that had inhaled once either 5F-Cumyl-P7AICA or 5F-MDMB-P7AICA in a dose of 200 μ g/kg prior to euthanasia was analyzed in the present study. **Methods:** The porcine tooth was extracted from the jaws, and soft tissues (gum and pulp) were removed. The dental hard tissue was pulverized, sequentially ultrasonicated with methanol and acetonitrile and analyzed by LC-MS/MS. Blank porcine dental samples were used as negative controls and matrix for calibration. **Results and Discussion:** Approximately 1.9 pg/mg of 5F-Cumyl-P7AICA was detected in a tooth of the respective pig. In a tooth of the other pig the hydrolysis product of 5F-MDMB-P7AICA was detected. 5F-MDMB-P7AICA itself was not detected, likely due to the instability of ester-containing synthetic cannabinoids. **Conclusion:** This study shows that a single inhalation of synthetic cannabinoids of the 7-azaindole type is detectable in postmortem teeth. These findings highlight the potential of dental tissue as an alternative matrix, particularly for postmortem toxicological analysis.

P08 Detection of exogenous substances in postmortem bones

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Aims: Blood, urine and body tissues are usually sampled for postmortem toxicological analyses. In cases of extreme decomposition, these matrices may no longer be available and bones

may be the only remaining source for examination. Only a few studies have focused on analysis of bone samples. Therefore, the aim was to develop a suitable sample preparation method for bones. **Methods:** Ethics committee approval was obtained prior to sampling bones from deceased bodies during autopsy. Samples from the petrous bone, rib and lumbar vertebrae were collected. Soft tissue was removed with scalpels, followed by washing with water for decontamination. After drying at 30 °C and 40 °C, bones were cut into 0.5 cm pieces. The segments were milled 4 times in steel jars with steel beads in a ball mill, while cooled in liquid nitrogen. From the pulverized sample, 500 mg was weighed and potentially incorporated substances were extracted with methanol using ultrasonication for 4 hours, then filtered through a syringe filter. The extract was evaporated to dryness and reconstituted in acetonitrile/methanol/water (3:3:2 v/v/v). Samples were analyzed by liquid chromatography quadrupole time-of-flight mass spectrometry and compounds identified by comparing spectra to a comprehensive database. **Results and Discussion:** The processed bone samples were initially too moist and insufficiently pulverized, which required improvements for reproducibility. Due to the moist and fatty nature of fresh bones, the decontamination step was adjusted to remove bone marrow, and segments were washed sequentially with water, 15 % (v/v) sodium chloride, 2 % (v/v) sodium dodecyl sulfate and ethanol/ether (3:1 v/v). Drying at 40 °C improved grinding efficiency and reduced the process to three 1-minute steps. These adjustments resulted in a fine bone powder suitable for analysis. To date, numerous analytes, such as antidepressants, opioids and anticonvulsants, were detected in bone samples. **Conclusion:** Bones may serve as a valuable alternative matrix for detecting intoxicants when other matrices are unavailable. In order to allow an evaluation of the concentrations in bones, the obtained values should now be compared with blood concentrations.

P09 The silent killer: Forensic detection of butane inhalation in a decomposed body

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Aims: The detection of toxic gases in decomposing bodies presents a significant challenge in forensic toxicology. We present the case of a 20-year-old woman (65 kg, 166 cm) found deceased in a state of progressive putrefaction four days after her last contact. Twelve bottles of deodorant were found at the scene, and the deceased had a known history of butane gas inhalation addiction. **Methods:** HS-GC/MS was applied to identify butane and additional components of the deodorant. Volatile compounds (ethanol, n-butane, isobutane, propane, 2-butanone, 2-butanol, and n-butanol) were quantified in femoral blood, heart, brain, and lung tissue using HS-GC/FID. Processed urine samples were analyzed for centrally acting substances using GC/MS. **Results and Discussion:** N-butane, isobutane, ethanol, propane, butyl esters, and pentyl esters were identified in the deodorant bottle. N-butane, isobutane, ethanol, propane, 2-butanone, 2-butanol, and n-butanol were detected in the autopsy samples. The highest n-butane concentrations were measured in femoral blood, followed by heart, brain, and lung tissues. Notably, concentrations were higher on the right side of the body. 2-Butanone and 2-butanol concentrations showed less variation between samples, with values of 177 µg/mL and 311 µg/mL in femoral blood, respectively, which are higher than those reported in previous literature. Ethanol was detected at 0.7 ‰ in femoral blood, but its origin is uncertain due to its presence in the deodorant. It may also occur during putrefaction processes along with n-butanol, which was 0.18 ‰. **Conclusion:** Post-mortem detection of gaseous substances, particularly in decomposing bodies, remains challenging. Our findings demonstrate acute butane inhalation through the detection of n-butane and its metabolites (2-butanone, 2-butanol) in multiple autopsy specimens.

P10 Headspace analyses for gaseous molecules in Western Switzerland

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Aims: Gas analyses are seldom requested and usually not part of forensic screening procedures. Nevertheless, gaseous molecules like helium, argon, nitrous oxide, carbon dioxide, methane, and others, can be the cause of intoxication or death or can help to evaluate the state of body alteration. To be able to help with such suspicions, qualitative and quantitative analytical approaches must be available. **Methods:** Qualitative and quantitative methods for the detection of gaseous molecules consisting of a combination of gas chromatography (GC) with mass spectrometric (MS) or thermal conductivity detection (TCD) have been developed and optimised during the last years. Chromatographic separation was realised on an Agilent 6890N GC equipped with a molecular sieve 5 Å PLOT (10 m × 0.32 mm i.d.) and a Porabond Q (50 m × 0.53 mm i.d.) capillary column. The detection was performed using manual commutation by a three-way valve linked to MS and TCD at the end of the capillary column. Injection was performed manually with a gas-tight syringe. **Results and Discussion:** 87 screenings for gaseous compounds and 76 determinations for six different gases (argon, helium, methane, propane, butane, and nitrous oxide) have been carried out between 2020 and 2024. Screenings were performed for body alteration investigation or diving accidents. Quantitative analyses were requested mostly for helium (32) and nitrous oxide (27). Detection of gases absent in ambient air were successfully in post-mortem cases, only, with few exceptions. Lung and brain tissues as well as lung and gastric air have shown to be the preferred matrices, knowing that they are hard to obtain in non-fatal poisonings. **Conclusion:** Gas analysis enables the detection and quantification of different molecules suspected in cases of suffocation due to hypoxia. To be beneficial, prompt sampling and direct preservation in gas-tight sample containers are essential.

P11 THC concentrations in blood samples before and after legalisation of cannabis in Germany

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Aims: In April 2024, legalization of cannabis in Germany came into effect. This was linked to the endeavor increasing the legal limit for THC in DUID cases. In August 2024, the limit for THC in serum was raised from 1.0 ng/mL to 3.5 ng/mL. The aim of this study was to evaluate possible changes of acute cannabis use by motor vehicle drivers. **Methods:** At the FTC Munich, more than 50,000 blood samples were analyzed for THC and its major metabolites between January 2021 and September 2024. A retrospective review of these cases was conducted to statistically evaluate the concentrations of THC and its metabolites during the months before and after legalization will be discussed. For statistical evaluation, only DUID cases (n = 47,020) were included. **Results and Discussion:** In 68.0 % of the samples, only cannabis was detected. The median THC concentration was 3.2 ng/mL across all cases, increasing from 3.1 ng/mL before to 3.7 ng/mL after legalization. The median THC-COOH concentration was 39.8 ng/mL across all cases, increasing from 38.9 ng/mL before legalization to 47.0 ng/mL after legalization. THC concentrations in single-cannabis use were higher (median 3.2 ng/mL before legalization, 3.8 ng/mL after legalization) than in multi-drug use (median 3.0 ng/mL before legalization, 3.5 ng/mL after legalization). The incidence of sanctions at a threshold of 3.5 ng THC/mL

serum increased from 59.3 % before to 63.5 % after legalization. **Conclusion:** The results of this study suggest that cannabis consumption had increased following the partial legalization of cannabis, as evidenced by the rise in THC and THC-COOH concentrations in blood samples of DUID cases. This increase is likely to have direct implications for road safety, as elevated THC levels can potentially increase the risk of impairment while driving.

P12 Driving under hexahydrocannabinol – Results of the examination of blood samples in Lower Saxony from 2021 to 2024

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Aims: Hexahydrocannabinol (HHC) plays a role as a semi-synthetic cannabinoid in road traffic with regard to its potential impact on driving ability. Currently, there is limited data available for the classification of HHC concentrations. The aim of the study is to classify HHC findings based on percentiles in terms of their concentration. **Methods:** GC-MS (Göttingen): 1 mL serum, SPE with Chromabond-C18 columns, elution with acetonitrile. Evaporation under N₂ and re-constitution in ethyl acetate. Derivatization with MSTFA, injection into the GC-MS system (Agilent Technologies 7890A GC combined with a 5975C MS; column: Macherey-Nagel Optima 5MS Accent). LC-MS-MS (Hannover): 30 µL serum, protein precipitation with 300 µL eluent, filtration, injection into the LC-MS system (Shimadzu HPLC coupled with API 5500 QTRAP tandem mass spectrometer). **Results and Discussion:** Approximately 2000 cannabinoid-positive blood samples submitted by the police between 2021 and 2024 were analyzed for HHC and HHC-carboxylic acid (HHC-COOH). Positive findings for HHC and HHC-carboxylic acid were observed in 244 samples. Based on the calculated percentiles, concentration ranges for HHC and HHC-carboxylic acid were established: low: < 0.58 ng/mL HHC / < 8.9 ng/mL HHC-COOH (25th percentile), moderate: 0.58-4.3 ng/mL HHC / 8.9-54 ng/mL HHC-COOH, high: > 4.3 ng/mL HHC / > 54 ng/mL HHC-COOH (75th-100th percentile). **Conclusion:** When addressing the question of whether individuals are impaired in terms of their fitness to drive, factors such as driving irregularities, physical abnormalities, and loss of motor and cognitive control are assessed alongside measurement values from toxicological examinations. The concentration ranges established in the study can help classify HHC findings. For a conclusive evaluation of concentrations, other factors must, of course, also be considered (e.g., time elapsed between consumption and blood sampling, tolerance etc.). To further refine this initial assessment, additional studies and investigations are still necessary.

P13 EMCDDA framework for naming cathinones

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Aims: Cathinones are the second largest group of new psychoactive substances (NPS). The short names attributed to the 170 cathinones monitored by the EUDA (formerly EMCDDA) are only loosely associated with structural features. Over time different naming approaches have been applied, leading to cathinones being known by several names. Although related to the parent compound cathinone, one of the psychoactive principals in khat, attributing consistent,

informative, and user-friendly common names to these substances is challenging. **Methods:** Current naming approaches were reviewed and common structural features of cathinones identified, for which abbreviations were derived from organic chemistry nomenclature and current names. **Results and Discussion:** An EMCDDA naming framework based on the main motifs ‘cathinone’ and ‘phenone’ was developed by incorporating earlier naming approaches. The framework name of each cathinone is composed of a parent element, which, combined with information on the keto alkyl chain or the amine substitution, yields the principal name. Additional substitutions are prepended to the principal name. Other parent elements besides the two main motifs (e. g. naphthalen-2-yl) are included in the naming framework. This way, a consistent and accurate naming syntax that represents structural features in the resulting semi-systematic names has been established. The framework also provides exceptions for several cathinones scheduled under UN and EU legislation and structural analogs (e. g. mephedrone/4-MMC/4-methylmethcathinone). **Conclusion:** The EMCDDA framework on naming cathinones provides practical guidance through examples and explanations of the rationale on how consistent semi-systematic names can be derived. Owing to the structural diversity of NPS, the forensic community, researchers, and policy makers widely recognize the need for harmonized naming to achieve consensus in the denomination of NPS in legislative texts and scientific dialogue.

P14 Stabilities of selected amphetamine synthesis markers from clandestinely disposed production waste in soil environment

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Aims: Clandestine amphetamine production via the Leuckart route is associated with large amounts of emerging toxic and corrosive production waste. Deliberate disposal of production waste in the environment (“dumpsites”) is of environmental concern due to the physicochemical properties and the number of compounds being present in the waste. Aim of this work is to reveal the fate of five characteristic synthesis markers when production waste is disposed in soil environment. **Methods:** The stabilities of five synthesis markers characteristic for amphetamine production via the Leuckart route were investigated in three standard soils under biotic and abiotic conditions for up to 60 d using GC- and LC-MS methods. Soil samples from a real case dumpsite next to a dismantled laboratory were also investigated using these techniques. **Results and Discussion:** Rapid degradation of the key reaction educt benzyl methyl ketone reported in previous research was confirmed and the route-specific by-product 4-methyl-5-phenylpyrimidine was degraded up to 82 %. Amphetamine, the characteristic intermediate *N*-formylamphetamine and the common by-product *N,N*-di-(β -phenylisopropyl)amine (isomers) showed maximum degradation of 31 % for the period of 60 d. Analysis of seized soil samples from a real case dumpsite next to a dismantled amphetamine laboratory revealed the presence of amphetamine synthesis markers in depths up to 80 cm due to the disposal of production waste. Target compounds from the laboratory-based stability study were detected plus several other characteristic synthesis markers, partly persistence-proven ones, including biotransformation products of the reaction educt benzyl methyl ketone. **Conclusion:** The results obtained allow authorities to determine environmental crimes, i.e., disposal of production waste, based on the stabilities of synthesis markers. Characterization of dumpsite samples allowed to infer on the origin of the disposed production waste, highlighted the potential of groundwater contamination with synthesis by-products plus biotransformation products and demonstrated the forensically relevant information that can be obtained in such a case.

P15 Unveiling Berlin's drug market: The surging dominance and complexity of cathinones

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Aims: This study focuses on the increasing dominance of cathinones in Berlin's recreational drug market, as revealed by the analysis of 2900 samples. It explores the structural diversity of cathinones and the challenges posed to harm reduction and laboratory methodologies. **Methods:** Drug samples were collected and analyzed as part of Berlin's drug-checking project. In addition to HPLC and GC-MS analyses, high-resolution mass spectrometry were employed to identify cathinones. Special attention was given to tracking new derivatives. **Results and Discussion:** Cathinones accounted for an unexpected 19.4 % of the analyzed samples. These synthetic stimulants exhibited significant structural variability, complicating their identification. This posed unique challenges, as the rapid emergence of new derivatives necessitated continuous attention to detail and up-to-date libraries to ensure accurate detection and characterization. An alarming 74.4 % of all cathinone samples were falsely declared containing e.g. 2-/3-MMC or 3-/4-CMC instead of 4-MMC, but less frequently also NEP, alpha-PHiP or other cathinones. HPLC-DAD was proven as the ideal system for differentiating between the dominant derivatives MMC and CMC and their regioisomers. The rapid influx of new cathinones requires frequent attention to detail in detection protocols and up-to-date libraries, underscoring the challenges laboratories face in keeping pace with market dynamics. **Conclusion:** The rise of cathinones underscores the necessity of agile and robust laboratory infrastructures capable of responding to an ever-evolving drug landscape. The continual appearance of new and modified substances demands ongoing investment in analytical technology and expertise. These findings highlight the critical role of drug-checking projects not only in harm reduction but also as sentinels for emerging trends, enabling timely public health interventions.

P16 Drugchecking Berlin – Highlights, lowlights and trends from 2900 samples

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Aims: This study summarizes key findings from Berlin's drug-checking project, which analyzed 2900 samples submitted between June 2023 and for the time being until now. It highlights the chemical trends, the challenges of identifying emerging substances, and the implications for public health and harm reduction. **Methods:** Drug-checking services were offered at harm reduction centers in Berlin starting in June 2023. Samples were analyzed mainly using HPLC as well as GC-MS to determine active ingredients, adulterants, and dosages. Particular emphasis was placed on detecting new psychoactive substances (NPS) and other emerging compounds. **Results and Discussion:** The project revealed that MDMA (23.4 %), cocaine (17.1 %), and amphetamines (11.5 %) were prevalent, but a notable 19.4 % of samples contained cathinones – an emerging class of synthetic stimulants characterized by significant structural diversity and a wide range of psychoactive effects. Notably, 50.6 % of all submitted samples were conspicuous with 19.2 % being falsely declared, i. e. containing substances entirely different from what users believed they had purchased. Furthermore, 25.5 % of the samples were contaminated with pharmacologically active adulterants. Almost all ecstasy pills were overdosed with MDMA. Fluctuations in purity were also observed, particularly for cocaine, which showed a sharp rise in adulterants starting in February 2024. These findings highlight the dual challenges

of identifying unknown compounds and addressing risks posed by contaminated samples. **Conclusion:** Berlin's drug-checking project illustrates the critical need for adaptable laboratory techniques to address an evolving drug market. By identifying trends and emerging risks, the project not only enhances individual safety but also serves as an early warning system for public health interventions.

P17 The EU project "NETZWERK ADEBAR"

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Aims: The NETZWERK ADEBAR project is a German cooperation project funded by the EU between several institutions (Federal Criminal Police Office, several State Criminal Police Offices, customs, universities) and presents the continuation of predecessor projects ADEBAR and ADEBAR *plus*. The main goal is the continuous collection of analytical data and structural elucidation of recently emerging substances on the drug market to supplement national, European, and world-wide databases. Furthermore, several sub-projects are carried out by partners as the assessment of pharmacological data, the prospective synthesis of new designer drug variants, the collection of reference material and the expansion of the internationally accessible, web-based database platform NPS DataHub (<https://nps-datahub.com/>). **Methods:** Analysis and characterization of samples included GC-MS, LC-MS, GC-sIR, FTIR, NIR, Raman and NMR. **Results and Discussion:** The data obtained is filed into electronic reports, converted to universal exchange formats and published in the databases NPS DataHub, DrugNews Forum and European Database on New Drugs (EDND). Over the last seven years, more than 1100 samples have been sent in, 612 data sets have been published, 439 reports have been filed for the EDND among which were 120 first identifications in Europe and an additional 160 first identifications in Germany. Through this process, an overview of the currently prevalent substance classes is gained which directly supports the legislation (NpSG) and the expansion of the NpSG tool (hosted within the NPS DataHub) which serves to review whether a structure is covered by the NpSG. **Conclusion:** With the structural elucidation and complementation of databases within the ADEBAR project as well as the reporting of data to institutions like the European Union Drugs Agency, we provide an essential contribution to the efforts against NPS and towards the necessary legislative developments. Not only is the identification of new and unknown substances supported, but also the future identification of new compounds is greatly facilitated with the provision of the data to all forensic laboratories.

P18 Cannabinoid analysis using GC-FID, HPLC-DAD/-QQQ/-Orbitrap: Overcoming analytical challenges with different analysis techniques

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Aims: Cannabis analysis has become increasingly relevant in recent years. During forensic investigations, the analysis of seized plant material as well as human samples is of importance. These analyses require reliable methods with varying sensitivities ranging from percentage by weight (% w/w) for major cannabinoids in plant material over parts per million (ppm w/w) for minor cannabinoids in plants to nanograms per milliliter (ng/mL) for cannabinoid quantitation

in biological samples. Additionally, plant material as well as biological samples have matrix-specific analytical challenges. Consequently, sensitive methods and sample preparation are crucial for reliable results. Besides natural cannabinoids, cannabis products spiked with synthetic cannabinoids add complexity, as these synthetic cannabinoids are present at low ppm (w/w) concentrations in addition to the naturally occurring cannabinoids. Furthermore, their analysis is challenging since a variety of substances with different structural characteristics emerged on the market over the last years. **Methods:** Various methods for analyzing cannabis plant material, cannabinoids in biological samples, and synthetic cannabinoid-infused cannabis products were developed and tested. All analyses employed chromatographic techniques (GC or HPLC) coupled with FID, DAD, or mass spectrometers (QQQ or Orbitrap) and different sample preparation techniques. **Results and Discussion:** For analysis of cannabis or hashish, sample preparation as well as chromatographic parameters are of integral importance. Different extraction mechanisms and chromatographic techniques and parameters were compared. Analysis of major and minor cannabinoids and metabolites in biological samples was achieved using LC-QQQ analysis. Sample preparation as well as detection mechanism is important to achieve adequate sensitivities. Detection of synthetic cannabinoids in plant materials using HPLC-Orbitrap analysis was achieved for 60 analytes with LODs of mainly < 5 ng/mL. **Conclusion:** We highlight challenges and solutions in (trace) analysis of cannabinoids across different matrices using analytical techniques, focusing on sample preparation, chromatographic separation, detection, and data evaluation.

P19 Validation of method for quantification of antimycotics utilizing DART-MS

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Aims: This work aimed to validate a rapid and quantitative method for measuring antifungal agents utilizing Direct Analysis in Real Time-Mass Spectrometry (DART-MS). Quick and accurate testing is crucial for effective antifungal therapy; therefore, the focus was on ensuring the method's robustness while reducing analysis time compared to LC-MS. It details the development and validation of a chromatography-free approach to quantifying six common antifungal agents in serum: 5-fluorocytosine, hydroxy-itraconazole, isavuconazole, itraconazole, ketoconazole, and posaconazole. **Methods:** For validation, five replicate calibration series were prepared in serum utilizing a RECIPE kit using a labeled internal standard for each compound. Protein precipitation was performed by vortexing 50µL of serum with 100µL of precipitant followed by centrifugation. 3.5µL aliquots of supernatant were transferred onto a QuickStrip HTS-96 screen and dried under heated nitrogen. For analysis, the screen was loaded onto a DART TQ-Plus (Bruker Daltonics) mass spectrometer for analysis. Inter and Intra-day precision was determined using matrix matched quality controls at three levels. This DART-MS method successfully measures 96 samples, with a throughput of 26 seconds per sample. **Results and Discussion:** The DART-MS method demonstrated excellent linearity ($R^2 > 0.99$) across the clinically relevant concentration ranges. Intra- and inter-assay precision were within acceptable limits (< 15 % CV), and accuracy ranged between 85 % and 115 % for all analytes. Results for spiked serum samples (n=105) passed correlation with LC-MS. The LOD and LOQ values were sufficiently low to detect therapeutic and subtherapeutic drug concentrations. Matrix effect experiments met criteria, confirming the method's reliability in serum and plasma. **Conclusion:** The validated DART-MS method is a robust, accurate, and efficient tool for quantification of antifungal agents. The method's high throughput and simple sample preparation provide an advantageous alternative to traditional liquid chromatography-based methods.