**Drug use by music festival attendees: A novel triangulation approach using self-reported data and test results of oral fluid and pooled urine samples**

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## ABSTRACT

**Background:** Self-reported data are commonly used when investigating illicit substance use. However, self-reports have well-known limitations such as limited recall and socially desirable responding. Mislabeling or adulteration of drugs on the illicit market may also cause incorrect reporting.

**Objectives:** We aimed to examine what could be gained in terms of illicit drug use findings among music festival attendees when including biological sample test results in the assessment.

**Methods:** We included 651 attendees at three music festivals in Norway from June to August 2016. Self-reported drug use was recorded using questionnaires, and samples of oral fluid were analyzed to detect use of illicit drugs. In addition, we analyzed samples of pooled urine from portable toilets at each festival.

**Results:** All methods identified cannabis, MDMA, and cocaine as the most commonly used drugs. Overall, 6.6% of respondents reported use of illicit substances during the previous 48 hours. Oral fluid testing identified a larger number of drug users as 12.6% tested positive for illicit drugs. In oral fluid testing, we identified ketamine and three new psychoactive substances (NPS) that had not been reported on the questionnaire. In pooled urine testing, we identified amphetamine and three additional NPS that were neither reported used nor found in oral fluid samples.

**Conclusions/Importance:** Drug testing of biological samples proved to be an important supplement to self-reports as a larger number of illicit substances could be detected.



**Keywords:** recreational drug use; illicit drugs; music festivals; self-reported drug use; oral fluid; pooled urine; drug testing

## Introduction

Illicit substance use is most commonly studied using self-reported data collected via questionnaires and/or interviews (Johnson & VanGeest, 2017; Sloboda, 2002). In addition to detailed information on drug use and consumption history, individual data on a range of potentially important variables can be collected for every respondent. However, self-reports have well-known limitations, such as under- or overreporting of actual drug use. Incorrect reporting may result from factors such as limited recall and socially desirable responding (Johnson & Richter, 2004; Johnson & Fendrich, 2005). Selection bias may be a problem, either because participants are non-randomly recruited or because some subgroups may have a lower probability of participation (Harrison & Hughes, 1997; Johnson, 2014). One particular problem for studies on the use of illegal drugs is that users may not know exactly what they have consumed (EMCDDA, 2016b; Tanner-Smith, 2006; Togni, Lanaro, Resende, & Costa, 2015; Vogels et al., 2009). The problem may apply in particular to inexperienced users, but even experienced users may not always know the true content of the substances used. This problem may have increased in recent years as a large number of so-called New Psychoactive Substances (NPS) have appeared on the drug market. NPS are defined as new narcotic or psychotropic drugs that were not included in the United Nations' Single Convention on Narcotic Drugs (UNODC, 1961) or the Psychotropic Substances Convention (UNODC, 1971). These mainly include synthetic stimulants, depressants, hallucinogens, and cannabinoids (EMCDDA, 2017; Nelson, Bryant, & Aks, 2014), but some plant-based drugs may also be classified as NPS (Schifano, Orsolini, Duccio Papanti, & Corkery, 2015). In cases where sales information or labels exist, these may be inaccurate or misleading due to intended mislabeling or adulteration of common drugs with NPS (Oliver et al., 2019; Palamar et al. 2017; Scherbaum, Schifano, & Bonnet, 2017; UNODC, 2016) or the chemical name may be difficult to remember. Hence, even when reporting to the best of their knowledge, users may still do so incorrectly.

An alternative to self-reports is drug testing of biological samples such as urine, oral fluid (saliva), sweat, hair, or blood (Fendrich, Johnson, & Becker, 2017; Fendrich, Johnson, Wislar, Hubbell, & Spiehler, 2004; Gjerde, Øiestad, & Christophersen, 2011; Salomone, Palamar, Gerace, Di Corcia, & Vincenti, 2017), which may be used to detect recent use of a wide range of substances. However, the refusal rate may be high if the sample collection is regarded as intrusive (Gjerde, Øiestad, & Christophersen, 2011). Fendrich and co-workers found in a population survey that about 10% refused to give a sample of oral fluid, whereas about 24%

refused to provide urine sample (Fendrich, Johnson, Wislar, & Hubbell, 2004). Some participants may fear that drug findings may be traced to the sample provider. Low participation rates may introduce a significant selection bias. Further, analysis of NPS presents a challenge compared with that of classical illicit drugs due to the large number of new substances and rapid changes in availability, as well as a complex pattern of metabolites in urine samples.

Wastewater-based epidemiology (WBE) has been recognized as a complementary tool for objectively monitoring the use of illicit drugs at population level (Bade et al., 2017; Brewer, Banta-Green, Ort, Robel, & Field, 2016; Burgard, Banta-Green, & Field, 2014; Thomas et al., 2012; Zuccato, Chiabrando, Castiglioni, Bagnati, & Fanelli, 2008). The methodology has recently also been explored for NPS (Bade et al., 2017; Gonzalez-Marino, Gracia-Lor, Rousis, et al., 2016); in the latter case, the above challenges also exist for WBE in relation to the low incidence of NPS use and therefore low concentrations in wastewater.

As an alternative to wastewater, analysis of pooled urine samples can be used to evaluate the consumption of both classical and new psychoactive drugs (Archer, Hudson, Wood, & Dargan, 2013; Mardal et al., 2017). Drug concentrations are obviously higher in pooled urine than in wastewater due to the much lower dilution factor, thereby increasing the possibility of detecting rarely used drugs, which is an important advantage. Few samples are needed, and a large number of different substances can be analyzed using the same sample, which represents a large number of individuals. As with wastewater testing, informed consent from individuals is not needed, and the sampling process is neither intrusive nor invasive. A disadvantage is that information about the participants is difficult to collect, including the number of people contributing to the pooled urine sample. Therefore, pooled urine testing does not contribute to estimating prevalence of illicit drug use. Nevertheless, pooled urine testing is a useful tool for determining the types of drugs consumed.

Advanced analytical methodologies are required to examine drugs in wastewater, pooled urine, oral fluid, or other biological samples, particularly for NPS (Hernandez et al., 2018). The most common approach is the monitoring of only specified substances. This allows quantification of very low drug concentrations in the samples, using techniques like liquid chromatography (LC) coupled to tandem mass spectrometry (MS/MS). Although this approach is highly useful and robust, it cannot be used to detect drugs that are not among the targeted compounds. Alternatively, the use of LC coupled to high-resolution mass

spectrometry (HRMS), linked to large mass spectral libraries, enables qualitative screening (i.e., detection and identification) of a large number of drugs, when quantification is not a primary objective. This is of particular relevance when many drugs are investigated, and/or when reference standards are not all available in the laboratory, which is a common situation when dealing with NPS.

Studies of nightlife settings and events such as music festivals have reported high rates of illicit substance use (Bijlsma, Serrano, Ferrer, Tormos, & Hernandez, 2014; Gripenberg-Abdon et al., 2012; Hesse & Tutenges, 2012; Hoegberg et al., 2018; Jenkinson, Bowring, Dietze, Hellard, & Lim, 2014; Johnson, Voas, Miller, & Holder, 2009; Lim, Hellard, Hocking, & Aitken, 2008; Miller, Byrnes, Branner, Voas, & Johnson, 2013; Miller et al., 2009; Miller et al., 2015; Mohr, Friscia, Yeakel, & Logan, 2018; Riley, James, Gregory, Dingle, & Cadger, 2001). Particularly high rates have been found at electronic dance music (EDM) events (Hesse & Tutenges, 2012; Johnson et al., 2009; Mohr et al., 2018; Riley et al., 2001). In addition to the use of classical drugs such as cannabis, amphetamines, cocaine, and MDMA (ecstasy), the use of NPS has been detected, although at lower levels than for classical drugs (Hoegberg et al., 2018; Riley et al., 2001; Palamar, Acosta, Sherman, Ompad, & Cleland, 2016; Palamar et al., 2017).

To the best of our knowledge, no previous studies have combined self-reported data and test results for drugs in oral fluid and pooled urine samples in settings such as music festivals. In this study, we aimed to examine what could be gained in terms of illicit drug use findings among music festival attendees when including biological sample test results in the assessment.

## Materials and methods

### *Setting*

Norway has a population of 5.2 million and the largest city has approximately 600,000 inhabitants. We selected three music festivals in Norway during the summer of 2016 for this study: a pop/rock music festival and an EDM festival, which both took place in a large city (>200,000 inhabitants), and a pop/rock music festival in a small town in a rural area. All three festivals had several thousand (8,500–20,000) visitors on each day of the festival.

### *Recruitment of participants*

At each festival site, a geographical recruitment area was defined. These were located in high-traffic areas, such as close to the entrances or exits or near toilet facilities. Data collection began between 7:00 and 9:00 p.m. and continued for about 3 to 4 hours, until about 200 participants had been recruited. All festivals had a large number of patrons passing through the selected area(s); it was therefore not possible to invite all patrons to participate or to use systematic random sampling. Consequently, this was a convenience sample. Participants were informed of the study and consented to taking part in the study. Data were collected using a questionnaire, and participants provided an oral fluid sample for drug testing. Participants received a voucher for food or soft drinks in lieu of reimbursement. Further details on participant recruitment and data collection have been previously published (Gjersing, Bretteville-Jensen, Furuhaugen, & Gjerde, 2019).

Participant recruitment and collection of data and oral fluid samples were approved by the Regional Committee for Medical and Health Research Ethics (approval no. 2016/337).

### *Self-report data*

A questionnaire for self-completion was used to record data on age, sex, education (less than 12 years; 12–13 years; bachelor's degree or higher), occupation (full-time job; part-time job; student; unemployed; sick leave), and self-reported use of cannabis, amphetamines, MDMA/ecstasy, cocaine, NPS, and MOP (which was a fictitious "dummy substance", to study the extent of overreporting) during the previous 48 hours, previous 12 months, and lifetime (yes/no for each drug class). Participants were asked to report which NPS they had used or tried during their lifetime, not during previous 48 hours or 12 months. We did not ask for use of specified NPS. If someone asked if a substance was included in the NPS category, and the research assistants were unsure, we asked them to tick "yes" and specify the type of substances used. The substances reported were assessed at a later stage.

### *Oral fluid samples*

Oral fluid samples were collected using the Intercept® Oral Fluid Collection Device (OraSure Technologies Inc., Bethlehem, PA, USA). Samples of oral fluid were analyzed to detect classical recreational drugs and a selection of NPS using ultra high-performance LC-MS/MS. The sample preparation and analytical methods have been described previously (Gjerde et al., 2016). Samples were analyzed by testing for either the active drug or inactive metabolites; this

was done for classical illicit drugs (amphetamines, MDMA, cocaine, cannabis, LSD, and heroin) as well as for 22 NPS, which were selected based on the opinion of experts and the types of NPS that participants reported using (see Supplementary Table S1). Sample extracts were reanalyzed to confirm tentative NPS findings using LC-HRMS with a quadrupole time-of-flight (q-TOF) mass spectrometer. Analytical data were matched with an in-house mass spectral library of approximately 1700 compounds.

***Pooled urine samples***

After each study day, the portable toilets on the festival grounds were emptied into a sewage disposal truck. Pooled urine samples were collected from the truck between 6:00 and 8:00 a.m. The samples were analyzed for the presence of a larger number of NPS than for the oral fluid samples due to differences in analytical methodologies; see Supplementary Table S1 for details. Qualitative analyses were performed for more than 190 NPS with HRMS using both quadrupole-time-of-flight and Orbitrap® (Thermo Fischer Scientific, Waltham, MA, USA) mass spectrometers, as described elsewhere (Bade et al., 2015; Gonzalez-Marino, Gracia-Lor, Bagnati, et al., 2016). We used mass spectral libraries or specific publications for identification of substances.

Quantitative analyses were performed with LC-MS/MS (Bade et al., 2017; Bijlsma, Beltran, Boix, Sancho, & Hernandez, 2014; Gonzalez-Marino, Gracia-Lor, Rousis, et al., 2016; Zuccato et al., 2016) for the same classical illicit drugs as listed above in oral fluid testing, except for LSD. Some selected NPS (mostly synthetic cathinones) were also quantified. The referenced quantitative methods were adapted (i.e., sample preparation and pre-concentration steps) and validated for the analysis of pooled urine, as these original methods were developed for the determination of illicit drugs and NPS in wastewater.

For cannabis, we only tested its main metabolite, 11-nor-9-carboxy-$\Delta^9$-tetrahydrocannabinol (THC-COOH) in pooled urine because the active substance tetrahydrocannabinol (THC) is mainly metabolized to THC-COOH and excreted via urine. This metabolite is generally used as a stable biomarker for cannabis in wastewater analysis (Bijlsma, Serrano, et al., 2014; Thomas et al., 2012).

***Statistical analysis***

We used Pearson's chi-squared test for categorical data to compare age distributions, education, and drug use among attendees at the three festivals. Wilson's binomial 95%

confidence intervals for proportions were calculated incorporating continuity correction (Newcombe, 1998).

## Results

### *Questionnaire responses*

Of the 651 study participants, 49% (n=320) were females. The proportion of participants younger than age 24 years was significantly higher at the EDM festival (74.1%) than the two pop/rock festivals (15.5% and 18.6%; $\chi^2$=201.3, p<0.001), and a larger proportion had not completed bachelor's degree or higher education (66.8% among EDM festival participants versus 20.4% and 19.5% among participants at the pop/rock festivals; $\chi^2$=136.2, p<0.001). Most attendees at the pop/rock festivals had full-time jobs, and four out of five held a bachelor's degree or higher. Characteristics of the three music festivals and the study cohorts are presented in Table 1.

[Insert Tables 1 and 2 near here]

In total, 6.6% of respondents reported use of illicit drugs during the previous 48 hours. Cannabis was the most commonly reported drug (5.1%), followed by MDMA (1.7%) and cocaine (1.1%). Cannabis was also most commonly reported drug used in the previous 12 months (21.8%), followed by MDMA (5.7%) and cocaine (5.4%). Details for each of the three festivals are presented in Tables 1 and 2. A larger proportion of the participants at the EDM festival reported use of MDMA during the previous 48 hours (3.9%) than attendees at the other festivals (0.7%; $\chi^2$=8.8, p=0.003).

Only 10 participants (1.5%) reported lifetime use of NPS, five (0.8%) during the previous 12 months and only one within the previous 48 hours. Respondents were only asked to specify the types of NPS used during their lifetime and not those used in the previous 48 hours; three participants reported having tried synthetic cannabinoids or "spice", two reported having used 2C-B (a psychedelic drug), one had used *Salvia divinorum*, and four did not specify which substance they had used.

### *Oral fluid test results*

Illicit drugs were found in 12.6% of oral fluid samples, which is a significantly larger proportion than self-reported use during the previous 48 hours ($\chi^2$=13.5, p<0.001). The most commonly detected drugs were THC, cocaine, and MDMA, the proportions and frequencies of which varied among the three festivals (Table 2).

Similar to the questionnaire responses, a larger proportion of participants at the EDM festival tested positive for MDMA ($\chi^2$=11.1, p=0.001) and cocaine ($\chi^2$=9.5, p=0.002) than attendees at the two pop/rock festivals. At the same time, a smaller proportion of participants tested positive for THC at the pop/rock festival in the small town than attendees at the two festivals in large cities ($\chi^2$=10.3, p=0.001). In analysis of oral fluid samples, we detected the use of ketamine by two persons at the EDM festival. Three NPS were detected: alpha-PVP and 2C-B had been used by a few participants at the large city pop/rock festival, and dimethyltryptamine by two participants at the small town pop/rock festival.

***Pooled urine test results***

In line with self-reports and results of oral fluid sample testing, analysis of pooled urine also revealed the highest proportion of MDMA use at the EDM festival. The highest concentrations of cocaine and its metabolite were found in the pooled urine sample from the pop/rock festival in the large city, with relatively high levels of MDMA detected as well. The sample from the small-town pop/rock festival showed the highest concentration of THC-COOH. Ketamine was detected only in the sample from the large city pop/rock festival. Overall, three NPS were detected: methcathinone, 4-chloro-alpha-PPP, and 2-phenethylamine; all three substances were found in the sample from the large city pop/rock festival, 2-phenethylamine also in samples from the two other festivals.

In some cases, a drug or its metabolite was found in pooled urine but it was not found in oral fluid samples nor its use reported on the questionnaires (amphetamines at the two pop/rock festivals and cocaine at the small town pop/rock festival; Table 2). Conversely, at the EDM festival, the use of cannabis was confirmed in self-reports and oral fluid testing but not in the pooled urine test results. A comparison of the three festivals based on self-reports and oral fluid testing was therefore slightly different than a comparison of the festivals using the pooled urine test results.

Finally, only 29 of the 82 persons (35.4%) who tested positive for illicit drugs in oral fluid, including NPS, reported having used the detected substance or NPS during the previous 48

hours. Among those who tested positive for cannabis, 51.3% reported such use during the previous 48 hours, whereas among those testing positive for cocaine or MDMA, only 25.5% reported such use ($\chi^2$=6.1, p=0.014).

## Discussion

To the best of our knowledge, this is the first study of illicit drug use at music festivals that combines self-reports with drug testing of both oral fluid and pooled urine samples. Although all methods identified the three same most commonly used drugs, the biological sample test results identified a larger number of illicit substances than the self-reports. Drug testing of biological samples therefore appears to be an important supplement to self-reports when investigating illicit substance use.

The biological sample test results and questionnaire responses indicated that the type of substances used differed among festivals. MDMA was more common among EDM festival attendees whereas cocaine was more common among participants at the pop/rock festival in the large city. At the small-town pop/rock festival, cannabis was the most commonly reported substance; few participants had used other drugs, as confirmed by analytical testing of oral fluid or pooled urine, and no one reported use of any other substance during the previous 48 hours.

Each of the three methods used — questionnaires, oral fluid sample testing, and pooled urine sample testing — have strengths and weaknesses. The use of a questionnaire enables the collection of sociodemographic data and information of self-reported drug use over a longer time period than can be detected with analysis of oral fluid or urine. We also used the questionnaire to collect data that are not presented in this article, such as the frequency and amount of drug use, other drug use habits, and some risk assessments.

Oral fluid drug testing is a more objective method to determine recent drug use than self-reporting. This methodology can be used to detect a large number of substances; we included 29 individual substances in our study; however, the number of oral fluid samples was relatively low. Each festival had thousands of attendees per day, so the selected study cohorts of about 200 people per festival constituted a small fraction of the total attendees at each event. Consequently, it was not possible to accurately estimate the prevalence rate of substance use in each festival, and it is possible that we did not detect all NPS used. However,

the latter was the main strength of the pooled urine samples; using pooled urine testing, we were able to identify substances not detected using the questionnaire or in oral fluid analysis.

It is difficult to estimate the prevalence rate of drug use based on pooled urine testing. It is also difficult to quantitatively compare the drug use levels at the different festivals because the number of participants contributing to the public toilet samples was unknown. In addition, the total drug dose per user might have been different at each festival.

Overall, all three methods had individual weaknesses, but when used in combination, these were able to strengthen the findings.

### *Discrepancies between self-reported data and results of oral fluid and urine testing*

The use of cocaine and MDMA during the previous 48 hours was clearly underreported. Underreporting was investigated in greater detail in a study including participants from six music festivals, including the three festivals in the present study (Gjerde, Gjersing, Furuhaugen, & Bretteville-Jensen, 2019). Underreporting has also been observed in previous studies (Gripenberg-Abdon et al., 2012; Harrison & Hughes, 1997; Johnson et al., 2009; Rendon, Livingston, Suzuki, Hill, & Walters, 2017); the magnitude may depend on age, sex, race, as well as type of drug (Harris, Griffin, McCaffrey, & Morral, 2008; Johnson, 2014; Rendon et al., 2017; Rosay, Najaka, & Herz, 2007). For example, there seems to be less hesitancy to report the use of cannabis than the use of amphetamine and cocaine in some settings (Gripenberg-Abdon et al., 2012; Johnson et al., 2009), possibly because the use of the latter drugs are more stigmatized. This seemed to be the case in our study as well.

Drug findings in oral fluid and pooled urine samples are not directly comparable. Drug detection in oral fluid samples mostly reflects drug use during the previous 10–50 hours, depending on the type of drug, whereas drug findings in urine samples may reflect drug use during the previous several days (Verstraete, 2004). Analysis of pooled urine samples revealed some drugs that were neither reported as having been used nor found in oral fluid samples; this is because the drugs found in pooled urine reflected drug intake by all users of the portable toilets during the entire festival day and not only the selection of participants who provided oral fluid samples and completed the questionnaire. The reason for not detecting the same NPS in pooled urine as those detected by oral fluid testing was probably that the drug concentrations in pooled urine were too low, either because of few users or because of drug

metabolism before excreted in urine. Many NPS are extensively metabolized, therefore mainly metabolites of those drugs can be found in urine with very low concentrations of the parent drugs (Favretto et al., 2013). Further undetectable substances may therefore have been consumed.

The discrepancy for cannabis at the EDM festival suggests that pooled urine testing is less sensitive than oral fluid testing in detection of cannabis use; this has also been previously reported to be a challenge in wastewater drug testing (Causanilles et al., 2017).

Furthermore, drugs might have been intentionally or unintentionally dumped into the public toilets, causing elevated drug concentrations that do not reflect actual drug use. The latter might have occurred for cocaine, as the observed ratio between cocaine and benzoylecgonine concentrations (3.4 and 1.8 for the two pop/rock festivals, respectively) was much higher than the commonly observed concentration ratios in wastewater (0.42±0.28), which reflects the excretion rate of human metabolism (EMCDDA, 2016a).

***Combining the three methods***

The findings when using the three methods were somewhat different; no single method gave a complete picture of drug use in the studied cohorts. Combining the three types of data, each with distinctive pros and cons, gave the most comprehensive picture of drug use. The three methods had advantages and limitations, with some overlapping information regarding qualitative data.

Self-reports and oral fluid samples provided specific information on the prevalence of individual drug use, including some NPS. The proportion who reported drug use during previous 48 hours was lower than the prevalence of drug findings in oral fluid samples due to under-reporting. However, self-reported data was needed to obtain information about drug use during the previous month, year and during lifetime as well as other information.

Pooled urine analysis is not well suited to study drug prevalence, but may instead show generic use in the studied cohorts. The wide-scope screening methodologies used and the large number of festival attendees contributing to the urine samples allowed for the potential detection of a very large number of drugs (190 NPS plus all traditional drugs); therefore, we were able to detect drugs whose use had not been reported.

The results indicated that neither amphetamine, methamphetamine, ketamine, cathinones, phenethylamines nor other NPS were used by a significant proportion of the participants. From other sources we know that the prevalence of NPS use is low in Norway (EMCDDA, 2018).

## Conclusions

The combination of three methods used in this study provided the most complete picture of illicit drug use. Although all methods identified the same three most commonly used drugs, the biological sample test results identified a larger number of illicit substances than the self-reports. Analysis of pooled urine samples did not add information on the prevalence of drug use, but identified some drugs that had been used by a small number of participants. Drugs used by few individuals may, however, not always be detected in pooled urine due to low concentrations. The drug testing of biological samples proved to be an important supplement to self-reporting. Future studies examining the type of substances used in a specific setting have much to gain by the addition of these methods. More comprehensive drug use data may indicate which measures are needed to reduce drug related harm and contribute to better policy making.

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## Declaration of interest

The authors report no conflicts of interest.

## References

Archer, J. R., Hudson, S., Wood, D. M., & Dargan, P. I. (2013). Analysis of urine from pooled urinals - a novel method for the detection of novel psychoactive substances. *Current Drug Abuse Reviews, 6*(2), 86-90. doi:10.2174/18744737066661312051440l4

Bade, R., Bijlsma, L., Sancho, J. V., Baz-Lomba, J. A., Castiglioni, S., Castrignano, E., . . . Hernandez, F. (2017). Liquid chromatography-tandem mass spectrometry determination of synthetic cathinones and phenethylamines in influent wastewater of eight European cities. *Chemosphere, 168*, 1032-1041. doi:10.1016/j.chemosphere.2016.10.107

Bade, R., Rousis, N. I., Bijlsma, L., Gracia-Lor, E., Castiglioni, S., Sancho, J. V., & Hernandez, F. (2015). Screening of pharmaceuticals and illicit drugs in wastewater and surface waters of Spain and Italy by high resolution mass spectrometry using UHPLC-QTOF MS and LC-LTQ-Orbitrap MS. *Analytical and Bioanalytical Chemistry, 407*(30), 8979-8988. doi:10.1007/s00216-015-9063-x

Bijlsma, L., Beltran, E., Boix, C., Sancho, J. V., & Hernandez, F. (2014). Improvements in analytical methodology for the determination of frequently consumed illicit drugs in urban wastewater. *Analytical and Bioanalytical Chemistry, 406*(17), 4261-4272. doi:10.1007/s00216-014-7818-4

Bijlsma, L., Serrano, R., Ferrer, C., Tormos, I., & Hernandez, F. (2014). Occurrence and behavior of illicit drugs and metabolites in sewage water from the Spanish Mediterranean coast (Valencia region). *Science of the Total Environment, 487*, 703-709. doi:10.1016/j.scitotenv.2013.11.131

Brewer, A. J., Banta-Green, C. J., Ort, C., Robel, A. E., & Field, J. (2016). Wastewater testing compared with random urinalyses for the surveillance of illicit drug use in prisons. *Drug and Alcohol Review, 35*(2), 133-137. doi:10.1111/dar.12185

Burgard, D. A., Banta-Green, C., & Field, J. A. (2014). Working upstream: how far can you go with sewage-based drug epidemiology? *Environmental Science & Technology, 48*(3), 1362-1368. doi:10.1021/es4044648

Causanilles, A., Baz-Lomba, J. A., Burgard, D. A., Emke, E., Gonzalez-Marino, I., Krizman-Matasic, I., . . . Bijlsma, L. (2017). Improving wastewater-based epidemiology to estimate cannabis use: focus on the initial aspects of the analytical procedure. *Analytica Chimica Acta, 988*, 27-33. doi:10.1016/j.aca.2017.08.011

EMCDDA. (2016a). *Assessing illicit drugs in wastewater: advances in wastewater-based drug epidemiology*. Luxembourg: Publications Office of the European Union.

EMCDDA. (2016b). *Recent changes in Europe's MDMA/ecstasy market*. Luxembourg: Publications Office of the European Union.

EMCDDA. (2017). *European Drug Report 2016*. Luxembourg: Publications Office of the European Union.

EMCDDA. (2018). Norway Drug Report 2018. Lisbon, Portugal: European Monitoring Centre for Drugs and Drug Addiction.

Favretto, D., Pascali, J. P., & Tagliaro, F. (2013). New challenges and innovation in forensic toxicology: focus on the "New Psychoactive Substances". *Journal of Chromatography A*, 1287, 84-95. doi:10.1016/j.chroma.2012.12.049

Fendrich, M., Johnson, T. P., & Becker, J. (2017). The Use of Biological Measures in Social Research on Drug Misuse. In J. B. VanGeest, T. P. Johnson, & S. A. Alemagno (Eds.), *Research Methods in the Study of Substance Abuse* (pp. 285-313). Cham, Switzerland: Springer International Publishing.

Fendrich, M., Johnson, T. P., Wislar, J. S., & Hubbell, A. (2004). Drug test feasibility in a general population household survey. Drug Alcohol Depend., 73(3), 237-250.

Fendrich, M., Johnson, T. P., Wislar, J. S., Hubbell, A., & Spiehler, V. (2004). The utility of drug testing in epidemiological research: results from a general population survey. *Addiction, 99*(2), 197-208. doi:10.1111/j.1360-0443.2003.00632.x

Gjerde, H., Gjersing, L., Furuhaugen, H., & Bretteville-Jensen, A. L. (2019). Correspondence between oral fluid drug test results and self-reported illicit drug use among music festival attendees. *Substance Use & Misuse, 54*, 1337-1344. doi:10.1080/10826084.2019.1580295

Gjerde, H., Nordfjærn, T., Bretteville-Jensen, A. L., Edland-Gryt, M., Furuhaugen, H., Karinen, R., & Øiestad, E. L. (2016). Comparison of drugs used by nightclub patrons and criminal offenders in Oslo, Norway. *Forensic Science International, 265*, 1-5. doi:10.1016/j.forsciint.2015.12.029

Gjerde, H., Øiestad, E. L., & Christophersen, A. S. (2011). Using biological samples in epidemiological research on drugs of abuse. *Norsk Epidemiologi, 21*(1), 5-14. doi:10.5324/nje.v21i1.1420

Gjersing, L., Bretteville-Jensen, A. L., Furuhaugen, H., & Gjerde, H. (2019). Illegal substance use among 1,309 music festival attendees: an investigation using oral fluid sample drug tests, breathalysers and questionnaires. *Scandinavian Journal of Public Health*. doi:10.1177/1403494818821481

Gonzalez-Marino, I., Gracia-Lor, E., Bagnati, R., Martins, C. P., Zuccato, E., & Castiglioni, S. (2016). Screening new psychoactive substances in urban wastewater using high resolution mass spectrometry. *Analytical and Bioanalytical Chemistry, 408*(16), 4297-4309. doi:10.1007/s00216-016-9521-0

Gonzalez-Marino, I., Gracia-Lor, E., Rousis, N. I., Castrignano, E., Thomas, K. V., Quintana, J. B., . . . Castiglioni, S. (2016). Wastewater-Based Epidemiology To Monitor Synthetic Cathinones Use in Different European Countries. *Environmental Science & Technology, 50*(18), 10089-10096. doi:10.1021/acs.est.6b02644

Gripenberg-Abdon, J., Elgan, T. H., Wallin, E., Shaafati, M., Beck, O., & Andreasson, S. (2012). Measuring substance use in the club setting: a feasibility study using biochemical markers. *Substance Abuse Treatment, Prevention & Policy, 7*, 7. doi:10.1186/1747-597x-7-7

Harris, K. M., Griffin, B. A., McCaffrey, D. F., & Morral, A. R. (2008). Inconsistencies in self-reported drug use by adolescents in substance abuse treatment: implications for outcome and performance measurements. *Journal of Substance Abuse Treatment, 34*(3), 347-355. doi:10.1016/j.jsat.2007.05.004

Harrison, L., & Hughes, A. (1997). *The validity of self-reported drug use: improving the accuracy of survey estimates. NIDA Research Monograph no. 167*. Rockville MD: National Institute on Drug Abuse.

Hernandez, F., Castiglioni, S., Covaci, A., de Voogt, P., Emke, E., Kasprzyk-Hordern, B., . . . Bijlsma, L. (2018). Mass spectrometric strategies for the investigation of biomarkers of illicit drug use in wastewater. *Mass Spectrometry Reviews, 37*(3), 258-280. doi:10.1002/mas.21525

Hesse, M., & Tutenges, S. (2012). Music and substance preferences among festival attendants. *Drugs and Alcohol Today, 12*(2), 82-88. doi:10.1108/17459261211235100

Hoegberg, L. C. G., Christiansen, C., Soe, J., Telving, R., Andreasen, M. F., Staerk, D., . . . Kongstad, K. T. (2018). Recreational drug use at a major music festival: trend analysis of anonymised pooled urine. *Clinical Toxicology (Philadelphia, PA), 56*(4), 245-255. doi:10.1080/15563650.2017.1360496

Jenkinson, R., Bowring, A., Dietze, P., Hellard, M., & Lim, M. S. (2014). Young Risk Takers: Alcohol, Illicit Drugs, and Sexual Practices among a Sample of Music Festival

Attendees. *Journal of Sexually Transmitted Diseases, 2014*, 357239. doi:10.1155/2014/357239

Johnson, M. B., Voas, R. A., Miller, B. A., & Holder, H. D. (2009). Predicting drug use at electronic music dance events: self-reports and biological measurement. *Evaluation Review, 33*(3), 211-225. doi:10.1177/0193841x09333253

Johnson, P. B., & Richter, L. (2004). Research note: What if we're wrong? Some possible implications of systematic distortions in adolescents' self-reports of sensitive behaviors. *Journal of Drug Issues, 34*(4), 951-970.

Johnson, T., & Fendrich, M. (2005). Modeling sources of self-report bias in a survey of drug use epidemiology. *Annals of Epidemiol, 15*(5), 381-389. doi:10.1016/j.annepidem.2004.09.004

Johnson, T. P. (2014). Sources of Error in Substance Use Prevalence Surveys. *Int Sch Res Notices, 2014*. doi:10.1155/2014/923290

Johnson, T. P., & VanGeest, J. B. (2017). Using Surveys to Study Substance Use Behavior. In J. B. VanGeest, T. P. Johnson, & S. A. Alemagno (Eds.), *Research Methods in the Study of Substance Abuse* (pp. 251-283). Cham: Springer International Publishing.

Lim, M. S., Hellard, M. E., Hocking, J. S., & Aitken, C. K. (2008). A cross-sectional survey of young people attending a music festival: associations between drug use and musical preference. *Drug and Alcohol Review, 27*(4), 439-441. doi:10.1080/09595230802089719

Mardal, M., Kinyua, J., Ramin, P., Miserez, B., Van Nuijs, A. L., Covaci, A., & Meyer, M. R. (2017). Screening for illicit drugs in pooled human urine and urinated soil samples and studies on the stability of urinary excretion products of cocaine, MDMA, and MDEA in wastewater by hyphenated mass spectrometry techniques. *Drug Testing & Analysis, 9*(1), 106-114. doi:10.1002/dta.1957

Miller, B. A., Byrnes, H. F., Branner, A. C., Voas, R., & Johnson, M. B. (2013). Assessment of club patrons' alcohol and drug use: the use of biological markers. *American Journal of Preventive Medicine*, 45(5), 637-643. doi:10.1016/j.amepre.2013.06.014

Miller, B. A., Furr-Holden, D., Johnson, M. B., Holder, H., Voas, R., & Keagy, C. (2009). Biological markers of drug use in the club setting. *Journal of Studies on Alcohol and Drugs*, 70(2), 261-268.

Miller, P., Curtis, A., Jenkinson, R., Droste, N., Bowe, S. J., & Pennay, A. (2015). Drug use in Australian nightlife settings: estimation of prevalence and validity of self-report. Addiction, 110(11), 1803-1810. doi:10.1111/add.13060

Mohr, A. L. A., Friscia, M., Yeakel, J. K., & Logan, B. K. (2018). Use of synthetic stimulants and hallucinogens in a cohort of electronic dance music festival attendees. *Forensic Science International, 282,* 168-178. doi:10.1016/j.forsciint.2017.11.017

Nelson, M. E., Bryant, S. M., & Aks, S. E. (2014). Emerging drugs of abuse. *Emergency Medicine Clinics of North America, 32*(1), 1-28. doi:10.1016/j.emc.2013.09.001

Newcombe, R. G. (1998). Two-sided confidence intervals for the single proportion: comparison of seven methods. *Statistics in Medicine*, 17, 857-872. doi:10.1002/(SICI)1097-0258(19980430)17:8<857::AID-SIM777>3.0.CO;2-E

Oliver, C. F., Palamar, J. J., Salomone, A., Simmons, S. J., Philogene-Khalid, H. L., Stokes-McCloskey, N., & Rawls, S. M. (2019). Synthetic cathinone adulteration of illegal drugs. *Psychopharmacology (Berl),* 236(3), 869-879. doi:10.1007/s00213-018-5066-6

Palamar, J. J., Acosta, P., Sherman, S., Ompad, D. C., & Cleland, C. M. (2016). Self-reported use of novel psychoactive substances among attendees of electronic dance music venues. Am J Drug Alcohol Abuse, 42(6), 624-632. doi:10.1080/00952990.2016.1181179

Palamar, J. J., Salomone, A., Gerace, E., Di Corcia, D., Vincenti, M., & Cleland, C. M. (2017). Hair testing to assess both known and unknown use of drugs amongst ecstasy

users in the electronic dance music scene. *International Journal of Drug Policy*, 48, 91-98. doi:10.1016/j.drugpo.2017.07.010

Rendon, A., Livingston, M., Suzuki, S., Hill, W., & Walters, S. (2017). What's the agreement between self-reported and biochemical verification of drug use? A look at permanent supportive housing residents. *Addictive Behaviors, 70*, 90-96. doi:10.1016/j.addbeh.2017.02.011

Riley, S. C., James, C., Gregory, D., Dingle, H., & Cadger, M. (2001). Patterns of recreational drug use at dance events in Edinburgh, Scotland. *Addiction, 96*(7), 1035-1047. doi:10.1080/09652140120053093

Rosay, A. B., Najaka, S. S., & Herz, D. C. (2007). Differences in the validity of self-reporting drug use across five factors: gender, race, age, type of drug, and offense seriousness. *Journal of Quantitative Criminology, 23*, 41-58. doi:10.3886/ICPSR02706.v1

Salomone, A., Palamar, J. J., Gerace, E., Di Corcia, D., & Vincenti, M. (2017). Hair Testing for Drugs of Abuse and New Psychoactive Substances in a High-Risk Population. *Journal of Analytical Toxicology,* 41(5), 376-381. doi:10.1093/jat/bkx020

Scherbaum, N., Schifano, F., & Bonnet, U. (2017). New Psychoactive Substances (NPS) - a Challenge for the Addiction Treatment Services. *Pharmacopsychiatry, 50*(3), 116-122. doi:10.1055/s-0043-102059

Schifano, F., Orsolini, L., Duccio Papanti, G., & Corkery, J. M. (2015). Novel psychoactive substances of interest for psychiatry. *World Psychiatry, 14*(1), 15-26. doi:10.1002/wps.20174

Sloboda, Z. (2002). Drug abuse epidemiology: an overview. *Bulletin on Narcotics, 54*(1-2), 1-13.

Tanner-Smith, E. E. (2006). Pharmacological content of tablets sold as "ecstasy": results from an online testing service. *Drug and Alcohol Dependence, 83*(3), 247-254. doi:10.1016/j.drugalcdep.2005.11.016

Thomas, K. V., Bijlsma, L., Castiglioni, S., Covaci, A., Emke, E., Grabic, R., . . . de Voogt, P. (2012). Comparing illicit drug use in 19 European cities through sewage analysis. *Science of the Total Environment, 432*, 432-439. doi:10.1016/j.scitotenv.2012.06.069

Togni, L. R., Lanaro, R., Resende, R. R., & Costa, J. L. (2015). The variability of ecstasy tablets composition in Brazil. *Journal of Forensic Sciences, 60*(1), 147-151. doi:10.1111/1556-4029.12584

UNODC. (1961). Single Convention on Narcotic Drugs. Vienna, Austria: United Nations Office on Drugs and Crime.

UNODC. (1971). Convention on Psychotropic Substances. Vienna, Austria: United Nations Office on Drugs and Crime.

UNODC. (2016). *New Psychoactive Substances: Overview of Trends, Challenges and Legal Approaches (E/CN.7/2016/CRP.2)*. Vienna, Austria: United Nations Office on Drugs and Crime.

Verstraete, A. G. (2004). Detection times of drugs of abuse in blood, urine, and oral fluid. *Therapeutic Drug Monitoring, 26*(2), 200-205.

Vogels, N., Brunt, T. M., Rigter, S., van Dijk, P., Vervaeke, H., & Niesink, R. J. (2009). Content of ecstasy in the Netherlands: 1993-2008. *Addiction, 104*(12), 2057-2066. doi:10.1111/j.1360-0443.2009.02707.x

Zuccato, E., Castiglioni, S., Senta, I., Borsotti, A., Genetti, B., Andreotti, A., . . . Serpelloni, G. (2016). Population surveys compared with wastewater analysis for monitoring illicit drug consumption in Italy in 2010-2014. *Drug and Alcohol Dependence, 161*, 178-188. doi:10.1016/j.drugalcdep.2016.02.003

Zuccato, E., Chiabrando, C., Castiglioni, S., Bagnati, R., & Fanelli, R. (2008). Estimating community drug abuse by wastewater analysis. *Environmental Health Perspectives, 116*(8), 1027-1032. doi:10.1289/ehp.11022

**Table 1.** Characteristics of music festivals and participants.

| | Pop/rock festival (large city) | EDM festival (large city) | Pop/rock festival (small town) |
|---|---|---|---|
| No. of attendees (approximate, per d) | 20,000 | 18,000 | 8,500 |
| No. of study participants | 226 | 205 | 220 |
| Response rate (%) | 60.2 | 53.7 | 77.5 |
| Male sex (%) | 46.5 | 54.1 | 47.3 |
| Age, y (%) | | | |
| 16–23 | 15.5 | 74.1* | 18.6 |
| 24–30 | 47.3 | 21.5 | 41.8 |
| 31–40 | 25.7 | 3.9* | 25.9 |
| 41+ | 10.6 | 0.0* | 13.6 |
| Not recorded | 0.9 | 0.5 | 0.0 |
| Education (%) | | | |
| Bachelor's degree or higher | 79.6 | 33.2* | 80.5 |
| Employment status, previous 30 d (%) | | | |
| Full-time | 72.6 | 42.4* | 69.1 |
| Part-time or student | 23.0 | 52.7 | 24.1 |
| Unemployed | 4.4 | 4.9 | 6.4 |
| Not recorded | 0.0 | 0.0 | 0.5 |
| Self-report drug use, previous 12 mo (%) | | | |
| Amphetamines | 2.7 | 5.9 | 0.5 |
| Cocaine | 6.6 | 7.3 | 2.3 |
| MDMA | 6.6 | 10.0§ | 0.9 |
| Cannabis | 23.5 | 23.9 | 18.2 |
| NPS | 0.0 | 1.5 | 0.9 |
| MOP | 0.0 | 1.5 | 0.0 |

*p<.001, §p<.005 when comparing participants at the EDM and the two pop/rock festivals.

Abbreviations: EDM, electronic dance music; NPS, new psychoactive substances. MOP, a fictitious "dummy substance", to study the extent of overreporting.

**Table 2.** Quantitative and qualitative results of analysis of pooled urine and oral fluid, and self-reported use of illicit drugs and NPS in the previous 48 hours.

| | Pop/rock festival (large city) | | | EDM festival (large city) | | | Pop/rock festival (small town) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Drug testing | | Self-reported use previous 48 h % (95% CI) | Drug testing | | Self-reported use previous 48 h % (95% CI) | Drug testing | | Self-reported use previous 48 h % (95% CI) |
| | Pooled urine (µg/L) | Oral fluid % (95% CI) | | Pooled urine (µg/L) | Oral fluid % (95% CI) | | Pooled urine (µg/L) | Oral fluid % (95% CI) | |
| Amphetamine | 4.9 | 0.0 (0.0-2.1) | 0.0 (0.0-2.1) | 5.4 | 0.0 (0.0-2.3) | 1.0 (0.2-3.9) | 8.3 | 0.0 (0.0-2.1) | 0.0 (0.0-2.1) |
| Methamphetamine | 3.8 | 0.0 (0.0-2.1) | – | 1.6 | 0.0 (0.0-2.3) | – | 1.6 | 0.0 (0.0-2.1) | – |
| Cocaine | 46.2 | 4.0 (2.0-7.7) | 1.3 (0.3-4.2) | 7.9 | 6.8 (3.9-11.4) | 2.0 (0.6-5.3) | 1.7 | 0.0 (0.0-2.1) | 0.0 (0.0-2.1) |
| Benzoylecgonine[a] | 13.4 | 1.8 (0.6-4.8) | – | 11.0 | 2.0 (0.6-5.3) | – | 0.9 | 0.0 (0.0-2.1) | – |
| MDMA (ecstasy) | 28.6 | 4.0 (2.0-7.7) | 1.3 (0.3-4.2) | 38.3 | 7.3 (4.3-12.0) | 3.9 (1.8-7.8) | 3.0 | 0.0 (0.0-2.1) | 0.0 (0.0-2.1) |
| Cannabis | – | – | 5.8 (3.2-9.9) | – | – | 7.8 (4.7-12.6) | – | – | 1.8 (0.6-4.9) |
| THC | n.a. | 8.8 (5.8-13.5) | – | n.a. | 7.3 (4.3-12.0) | – | n.a. | 1.8 (0.6-4.9) | – |
| THC-COOH[b] | 1.3 | n.a. | – | 0.0 | n.a. | – | 3.3 | n.a. | – |
| Ketamine | 0.1 | 0.0 (0.0-2.1) | – | 0.0 | 1.0 (0.2-3.9) | – | 0.0 | 0.0 (0.0-2.1) | – |
| NPS | See below | 1.3 (0.3-4.2) | 0.0 (0.0-2.1) | See below | 0.0 (0.0-2.3) | 0.5 (0.0-3.1) | See below | 0.9 (0.2-3.6) | 0.0 (0.0-2.1) |
| Methcathinone | 0.3 | n.a. | – | 0.0 | n.a. | – | 0.0 | n.a. | – |
| 4-chloro-alpha-PPP | Positive[c] | n.a. | – | n.a. | n.a. | – | n.a. | n.a. | – |
| 2-phenethylamine | Positive | n.a. | – | Positive | n.a. | – | Positive | n.a. | – |
| Alpha-PVP | 0.0 | 0.4 (0.0-2.8) | – | n.a. | 0.0 (0.0-2.3) | – | n.a. | 0.0 (0.0-2.1) | – |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Dimethyltryptamine | n.a. | 0.0 (0.0-2.1) | – | n.a. | 0.0 (0.0-2.3) | – | n.a. | 0.9 (0.2-3.6) | – |
| 2C-B | n.a. | 0.9 (0.2-3.5) | – | n.a. | 0.0 (0.0-2.3) | – | n.a. | 0.0 (0.0-2.1) | – |

[a]Inactive metabolite of cocaine.

[b]Inactive metabolite of THC.

[c]Tested positive, not quantified.

n.a.: not analyzed.

–: not queried or not applicable.

**Supplementary material**

**Table S1.** Cut-off concentrations for illicit substances analyzed in oral fluid or pooled urine samples using quantitative methods.

| Illicit substance | Neat oral fluid (µg/L)[a] | Pooled urine (µg/L) |
|---|---|---|
| *Cannabis* | | |
| Tetrahydrocannabinol | 0.37 | n.a. |
| Carboxy-tetrahydrocannabinol | n.a. | 0.060 |
| *Central stimulants* | | |
| Amphetamine | 15 | 0.10 |
| Benzoylecgonine | 4.3 | 0.060 |
| Cocaine | 1.1 | 0.060 |
| MDMA (ecstasy) | 2.3 | 0.060 |
| Methamphetamine | 8.9 | 0.060 |
| *Illicit opiate* | | |
| Heroin | n.a. | 0.10 |
| 6-monoacetylmorphine | 4.7 | 0.060 |
| *Hallucinogens* | | |
| LSD | 0.019 | n.a. |
| Ketamine | 0.34 | 0.060 |
| Salvinorin A | 3.1 | n.a. |
| *NPS*[b] | | |
| 25B-NBOMe | n.a. | 0.10 |
| 25C-NBOMe | 0.048 | 0.10 |
| 25I-NBOMe | 0.062 | 0.10 |
| 2C-B | 0.23 | n.a. |
| 2C-I | 0.28 | n.a. |
| 3,4-dimethylcathinone | n.a. | 0.060 |
| 3,4-methylenedioxy-pyrovalerone | 0.50 | 0.060 |
| 4-fluoromethcathinone | n.a | 0.060 |
| 4-methylamphetamine | 0.54 | n.a. |
| 4-methylcathinone | n.a. | 0.060 |
| 5F-APINACA | 0.093 | n.a. |
| 5F-PB-22 | 0.091 | n.a. |
| Alpha-PVP | 0.13 | 0.10 |
| AM-2201 | 0.087 | n.a. |
| Buphedrone | n.a. | 0.060 |
| Butylone | n.a. | 0.060 |
| Diclazepam | 0.19 | n.a. |
| Dimethyltryptamine | 0.11 | n.a. |
| Ethcathinone | n.a. | 0.060 |
| Ethylone | n.a. | 0.060 |
| Ethylphenidate | 0.15 | n.a. |
| Etizolam | 0.22 | n.a. |

| | | |
|---|---|---|
| Flubromazepam | 0.20 | n.a. |
| Flubromazolam | 0.22 | n.a. |

**Table S1** continued.

| **Substance** | **Neat oral fluid (µg/L)**[a] | **Pooled urine (µg/L)** |
|---|---|---|
| Mephedrone | 0.11 | 0.060 |
| Methcathinone | n.a. | 0.060 |
| Methedrone | n.a. | 0.060 |
| Methiopropamine | 0.087 | n.a. |
| Methylone | n.a. | 0.060 |
| Naphyrone | n.a. | 0.060 |
| Penthedrone | n.a. | 0.060 |
| Pentylone | n.a. | 0.060 |
| THJ-2201 | 0.087 | n.a. |
| UR-144 | 0.075 | n.a. |

[a]Assuming that 0.4 mL oral fluid was collected and mixed with 0.8 mL preservative buffer.

[b]The listed substances were defined as NPS in this study.

n.a.: not analyzed.