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## A 7-year study of workplace drug testing in two major cities in Türkiye

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### ABSTRACT

**Background:** Workplace Drug Testing (WDT) is a worldwide practice that is crucial in anti-drug efforts. This study aimed to provide comprehensive information about the WDT applications in Türkiye with data obtained from two major cities in two different regions of Türkiye.

**Methods:** The retrospective assessment of forensic toxicological analysis results in urine, hair or urine&hair samples collected from 220 WDT cases in İzmir (3<sup>rd</sup> largest city) and Adana (5<sup>th</sup> largest city) from 2016 to 2022 years was carried. The screening and confirmation analysis were performed with immunoassay and liquid chromatography-tandem mass spectrometry (LC-MS/MS), respectively.

**Results:** Cases with a positive result from one of urine or hair tests were accepted as positive. 20.5% ( $n=45$ ) of the cases were detected positive, and most applicants were identified as drivers (84.4%,  $n=38$ ). Urine samples of 23.5% ( $n=52$ ) were screened by immunoassay, and THC ( $n=1$ ) and opiate ( $n=1$ ) were detected. In the LC-MS/MS analysis, benzodiazepines were detected most in urine as a single substance, while cannabinoids and codeine were detected in the hair. Multidrug combinations used in treatment were detected in urine and hair. It was detected both alone and among the substances accompanying cannabinoids and amphetamines.

**Conclusions:** This study presents 7-year WDT results of two different major cities in Türkiye. The method of WDT application differs across the country so a standard procedure should be performed. In our country, where zero tolerance against substance use is adopted, the detection of illegal and controlled substances in cases clearly shows that WDT practice should be routinized.

### KEYWORDS

Workplace drug testing; forensic toxicology; drug abuse; controlled drugs; pregabalin

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## 1. Introduction

Recently, the use of psychoactive substances, including illicit drugs and prescription medications, has surged. In response, Workplace Drug Testing (WDT) has become a key initiative, playing a crucial role in addressing the issue of psychoactive substance use. The implementation of WDT has expanded across a wide range of countries. Originating in the United States in 1980 due to a series of railway accidents, it was later integrated into the strategies of various industrial and governmental sectors as a major part of the fight against drug abuse (Joseph Rowntree Foundation, 2004). The formation of the European Workplace Drug Testing Society (EWDTs) in 1998 marked a significant development, aligning with establishing WDT standards. Following these events, WDT's importance has increased in Europe, Australia, and other developed countries, broadening its application significantly over the past decade (Caplan & Goldberger, 2001).

Workplace drug testing can be conducted as part of pre-employment procedures, where urine samples are requested from all job candidates, or through post-employment monitoring, which may involve random testing, targeted testing based on comprehensive assessment, or testing under reasonable suspicion from current employees (Normand et al., 1990). These measures ensure adherence to legal mandates by both employers and employees and contribute to reducing workplace accidents and criminal activities, enhancing work quality and productivity, and discouraging drug abuse. In Türkiye, the regulatory framework and implementation practices for drug testing in the workplace are governed by several legal instruments, including the Labor Law, Occupational Health and Safety Law, and specific legislation related to Workplace Medicine and Transportation (covering Aviation, Highways, and Maritime Transport) (Sht-Apam Instructions on Alcohol & Psychoactive Substance Controls of Aviation Personnel Subject to Control (Sht-Apam),, 2019; Occupational Health & Safety Law, 2012). The legislation stipulates that being at the workplace while intoxicated or under the influence of drugs, or consuming alcohol or drugs within the workplace premises is strictly prohibited. In adherence to pertinent legal provisions, the framework for implementing drug testing in workplaces has been established to enhance productivity, quality, and safety in the legislation. This practice has been gradually gaining momentum across Türkiye.

In adherence to legal standards, forensic toxicological assessments conducted to identify abused drugs in biological specimens within workplace contexts are of paramount importance (Aydoğdu et al., 2021). According to international guidelines, the results of workplace drug tests, which must be precise, dependable, and legally defensible, require validation and reporting by a Medical Review Officer (MRO) specialized in this field.

Various factors can influence the outcome of these analyses, such as the type of biological specimen chosen, the analysis timeframe, the physiological characteristics of the individuals being tested, and their consumption of drugs, food, and supplements. In addition to these factors, WDT applications of laboratories that use different analytical approaches may produce different results in drug tests. As the advancement of analytical techniques in recent years and laboratories' preference for alternative biological matrices according to the purpose of analysis has made the detection of psychoactive substances easier, it is inevitable to use different applications.

In this study, data obtained from two principal provinces in different geographical regions of Türkiye and Workplace Drug Testing (WDT) application results were presented. The aim of the study is to reflect a perspective on the forensic, clinical and possible workplace safety consequences of different practices of laboratories within the scope of workplace drug testing. Assuming that the results may differ not only between laboratories that prefer different methodologies, but also due to substantially different regional drug consumption patterns, this study will also provide a common contribution for occupational physicians, employers, lawyers, judicial authorities and workers.

## 2. Methods

This is a cross-sectional study evaluating a total of 220 cases requesting workplace drug testing in Forensic Toxicology Laboratory of Çukurova University in Adana and Addiction Toxicology Laboratory of Ege University in İzmir. In the cases, drug tests were conducted for a reasons such as pre-employment, random drug testing, reasonable suspicion (decreased work efficiency, absent-mindedness) or accidents at workplace. This investigation analyzes workplace drug testing results derived from urine and hair samples collected from cases seeking WDT in İzmir, the third-largest city in western Türkiye, and Adana, the fifth-largest city in southeastern Türkiye, spanning the years 2016 to 2022. The methodologies determined for screening analysis in this study are immunoassay and liquid chromatography-tandem mass spectrometry (LC-MS/MS) for confirmatory analysis. The biological specimens collected from cases undergoing WDT consisted of urine, hair, or a combination of both, depending on each case's specific circumstances and historical context. Instances where substances were detected in either urine or hair samples were categorized as "positive." The ethical clearances for conducting this study were duly obtained from the Ethics Committees of Ege University and Çukurova University under the respective decision numbers 17-2.1/14 and 125.

### **2.1. Sample collection**

The total number of cases admitted to the Addiction Toxicology Laboratory at Ege University in Izmir for WDT analysis between 2017 and 2021 was 53, and the analysis was exclusively performed on urine samples. Before collecting samples, each case was subjected to a preliminary interview by a specialist physician. Following this, consent forms specific to the WDT were filled out, and urine samples were collected by trained personnel adhering to strict chain of custody protocols.

The Forensic Toxicology Laboratory at Çukurova University in Adana, Türkiye, handled 167 WDT inquiries between 2016 and 2022. The analyses in these instances were carried out on urine ( $n=111$ ), hair ( $n=51$ ), or a combination of both ( $n=5$ ), depending on the institution and the purpose of the drug test request. Comprehensive informed consent, both written and oral, was obtained from all participants. The necessary biological specimens were collected by skilled personnel, maintaining the chain of custody. These samples were then appropriately stored under controlled conditions until they were analyzed.

### **2.2. Prescreening analysis**

Urine integrity testing (including urine osmolarity, creatinine, pH, and Intect 7 test for adulterants) was conducted on all urine samples obtained from individuals from both laboratories according to the European Guidelines for Workplace Drug Testing (EWDTS) in urine (EWDTS, 2015). Subsequent to the urine integrity assessment, the urine samples were screened *via* the Drug of Abuse panel encompassing the analytes delineated in Table S1.A fully automated toxicology analyzer (Radox Evidence Automated Biochip Array Analyzer) based on chemiluminescence analysis was used in the screening analysis for the cases in Izmir. Screening analyses were executed utilizing biochip technology with calibrators and quality controls. A 9-point calibration curve was first plotted containing each analyte to be analyzed. To ensure the validity of the method, 2-point quality control samples that were within the calibration range were analyzed. When the control results were valid within the specified range, the samples were injected into the device. Urine integrity testing was conducted in Adana but prescreening analyses of immunoassay analyses were not performed.

### **2.3. Confirmation analysis**

As part of the study, all samples were subjected to confirmation analysis Thermo Scientific Q-Exactive Focus TM liquid-chromatography tandem mass spectrometry (Thermo Scientific, USA) in Izmir and Shimadzu 8040

liquid-chromatography tandem mass spectrometry system (Shimadzu, Kyoto, Japan) in Adana after sample preparation steps. In this study, the confirmation method was validated in terms of the Scientific Working Group for Forensic Toxicology (SWGTOX) (SWGTOX, 2013). The following parameters were evaluated: LLOQ, selectivity, linearity, precision, accuracy, matrix effect and carry-over. All of the results of validation parameters for whole blood, urine and hair matrices were within the target criteria and acceptable range according to the SWGTOX guideline.

### ***2.3.1. Sample preparation for confirmation analysis***

For the urine samples collected in İzmir, a preparation process involving a purification step using a well plate (Recipe chemicals, Germany) was employed. Each urine sample (500 µL) was subjected to hydrolysis in a 1 M phosphate buffer with the pH:6,8 (Sigma Aldrich, Germany) containing the enzyme beta-glucuronidase (activity: 100,000 units/mL) (Cova chem, Canada) for a duration of 1 h in an oven. Post-incubation, the samples were allowed to normalize to room temperature before being centrifuged. The supernatant was then carefully transferred to a polypropylene filter plate called “DeepWell plate” (Thermo Scientific, USA). The filter plate was conditioned with methanol (MeOH) and subsequently sealed with aluminum foil, and finally, the prepared samples were loaded into the analytical instrument.

In the case of samples gathered in Adana, preliminary sample preparation steps were meticulously followed prior to analysis. Urine samples underwent a liquid-liquid extraction process using De-Tox Tubes (A® and B®) as part of the standard laboratory protocol, subsequent to the integrity assessment. For hair samples, a series of preparation steps, including washing, cutting, and solid-phase extraction (SPE), were executed in alignment with established laboratory procedures, culminating in the samples being positioned within the analytical device for further analysis. 50 mg hair strands were weighed into a borosilica glass tube (10 mL). 2 mL of MeOH was added and incubated in an ultrasonic bath (Bandelin Sonorex, Rangendingen, Germany) for 2 h at ambient temperature. After the samples were centrifuged at 4000 rpm, the supernatant that decanted to a borosilica glass tube was left to dry under N<sub>2</sub> gas. The residue was dissolved in 150 µL of MeOH and 850 µL of distilled water and loaded into the SPE cartridge (OASIS HLB, Waters, Milford, MA, USA). The cartridge was washed with 5% MeOH (in H<sub>2</sub>O) and left to dry for 10 min. It was then eluted with 2 mL of MeOH and left to dry under N<sub>2</sub> gas. The residue was dissolved in 100 microliters of MeOH and injected into the LC/ms/mS device in a volume of 10 microliters.

### **2.3.2. Liquid chromatography-tandem mass spectrometry analysis (LC-MS/MS)**

Urine samples from Izmir were subjected to an analysis covering 151 parameters using LC-MS/MS, including but not limited to cannabinoids, synthetic cannabinoids, amphetamine-type stimulants (ATS), opioids, cocaine and its metabolites, fentanyl, cathinones, benzodiazepines, and prescription drugs and related MRM data was given and listed in Supplementary file [Table S2](#). Before this analysis, settings for multiple reaction monitoring (MRM) in electrospray ionization (ESI) mode were configured. The separation was performed on a C18 column (Gemini Phenomenex 150×2.0 mm i.d., 3 µm, USA), with mobile phase A being 5 mM ammonium bicarbonate and water (pH 9), and mobile phase B being methanol. The flow rate was set at 0.3 mL/min, with the total run time per sample at 18 min, excluding washing, and an injection volume of 10 µL.

In Adana, both urine and hair samples from all cases underwent LC-MS/MS analysis for 125 substances, encompassing cannabinoids, synthetic cannabinoids, amphetamine-type stimulants (ATS), opioids, cocaine and its metabolites, fentanyl, khat, benzodiazepines, prescription drugs, and more, as detailed in [Table S3](#). The analysis entailed the integration of the area under the curve for specific multiple reaction monitoring (MRM) settings in electrospray ionization (ESI) mode. Analytes were separated on a C18 column (Allure PFPP 50×2.0 mm i.d., 5 µm). Mobile phase A consisted of 10 mM ammonium formate and water, and mobile phase B was methanol. The flow rate was set at 0.4 mL/min, with a total run time of 18 min per sample, not including the washing phase, and the injection volume was maintained at 10 µL.

### **2.4. Statistical analysis**

Demographic data of the cases were obtained from the hospital management system and file of cases. Descriptive statistics included frequencies, percentages, frequency tables for categorical variables, and means ± standard deviation (SD) for quantitative variables. Categorical variables were evaluated by a 2-independent samples test. Statistical analyses were performed with SPSS 25.0.

## **3. Results**

In this research, a total of 220 cases across two different regions were examined. Cases were classified as positive if at least one substance was detected through either immunoassay or LC-MS/MS analysis. The mean age of the individuals involved in the study was  $43.7 \pm 11.8$  years. Out of the total cases, 20.5% ( $n=45$ ) yielded positive results, with the vast majority

of these positive cases, 87.3%, being male. The most common occupational group among the cases was drivers. Detailed socio-demographic characteristics of the individuals are outlined in Table 1.

Figure 1 depicts substance detection rates for cases presenting for WDT analysis, which commenced on divergent dates contingent on the city. No significant differences were discerned in substance detection proportions per month ( $p=0.745$ ) or year ( $p=0.164$ ).

Data on the comparison of the results in urine samples for WDT applications in Izmir and Adana provinces are given in Figure 2.

Positivity and negativity frequencies (n) according to the analyzed biological sample types (whole blood, urine, and hair) were shown in Figure 3.

Preliminary immunoassay screening was conducted on urine samples from 23.46% ( $n=52$ ) of the cases, resulting in the detection of THC and opiates in one case each. In the LC-MS/MS analysis, benzodiazepines emerged as the most frequently identified single substance in urine samples

**Table 1.** Demographic information of the cases.

| n (%)                  | Negative cases | Positive cases | Total           | Significance test |
|------------------------|----------------|----------------|-----------------|-------------------|
| <b>Number of cases</b> | 175 (79.5)     | 45 (20.5)      | 220 (100.0)     |                   |
| <b>Age, years</b>      |                |                |                 |                   |
| Mean $\pm$ SD          | 43.1 $\pm$ 0.9 | 46.0 $\pm$ 1.9 | 43.7 $\pm$ 11.8 | $p=0.194$         |
| Min-max                | (19–64)        | (24–73)        | (19–73)         |                   |
| 18–24                  | 11 (6.3)       | 2 (4.4)        | 13 (5.9)        | $p=0.262$         |
| 25–34                  | 31 (17.7)      | 7 (15.6)       | 38 (17.3)       |                   |
| 35–44                  | 44 (25.1)      | 8 (17.8)       | 52 (23.6)       |                   |
| 45–54                  | 47 (26.9)      | 16 (35.6)      | 63 (28.6)       |                   |
| 55–64                  | 34 (19.4)      | 9 (20.0)       | 43 (19.5)       |                   |
| $\geq 65$              | –              | 1 (2.2)        | 1 (0.5)         |                   |
| <b>Sex</b>             |                |                |                 |                   |
| Female                 | 25 (14.3)      | 3 (6.7)        | 28 (12.7)       | $p=0.172$         |
| Male                   | 150 (85.7)     | 42 (93.3)      | 192 (87.3)      |                   |
| <b>Occupation</b>      |                |                |                 |                   |
| Unknown                | 49 (28.0)      | 4 (8.9)        | 53 (24.1)       |                   |
| Employee               | 10 (5.7)       | 2 (4.4)        | 12 (5.4)        |                   |
| Pilot                  | 2 (1.1)        | 1 (2.2)        | 3 (1.4)         |                   |
| Driver                 | 114 (65.1)     | 38 (84.4)      | 152 (69.1)      |                   |

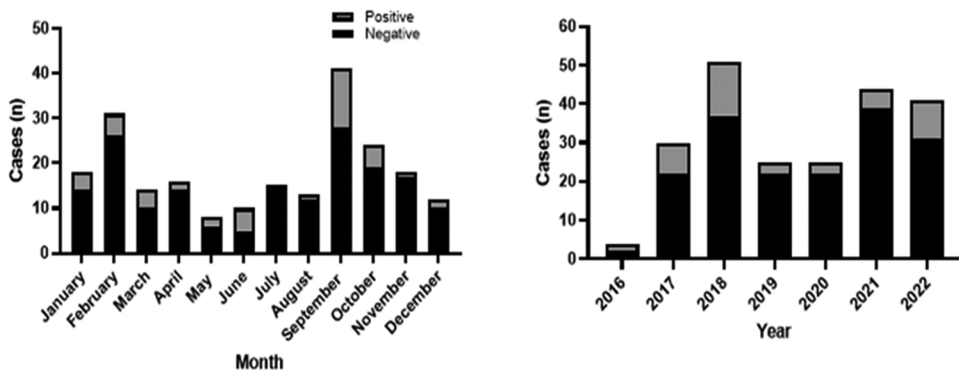


**Figure 1.** Drug detection status in the cases according to months and years.

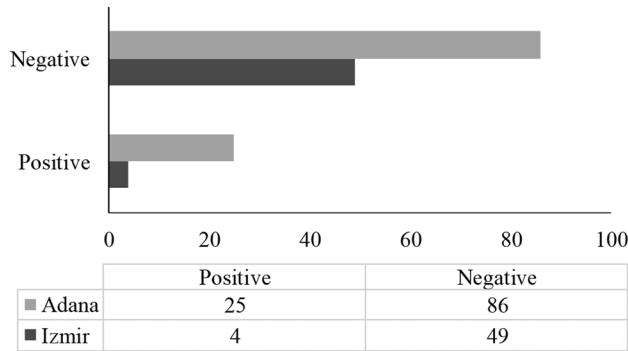


**Figure 2.** WDT results according to province.

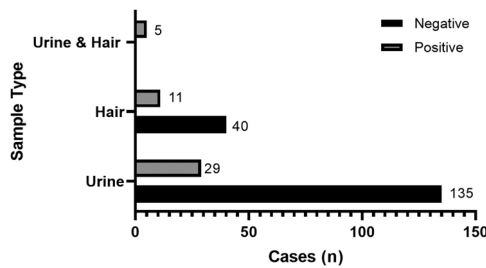


**Figure 3.** Positivity and negativity frequencies (n) according to biological sample types.

(Figure 4), whereas codeine was the predominant substance detected in hair samples that given in Figure 5.

In instances of multi-substance detection, therapeutic compounds such as antipsychotics, antidepressants, antihistamines, and benzodiazepines were prevalent. It is noteworthy that pregabalin was classified as a controlled prescription drug in our country in 2019. Despite not being a part of the standard screening regimen, pregabalin was assessed in 54 cases within this study, with 5.5% ( $n=3$ ) testing positive for the substance. Pregabalin, often found in toxicological evaluations involving polydrug use, was identified both as a single substance and in conjunction with cannabinoids and amphetamines, as illustrated in Figure 6.

#### 4. Discussion

The current study is the first to provide information on WDT in Türkiye containing data of Türkiye's 3<sup>rd</sup> (Izmir) and 5<sup>th</sup> largest (Adana) cities. There is no data in the literature on drug use among Turkish workers *via* a WDT application. In addition, it is difficult to compare the findings of this study with other similar studies on substance abuse prevalence due to the differences in WDT practices between the two cities, such as analytical procedure and biological sample type (Lamberg et al., 2008; Lillsunde

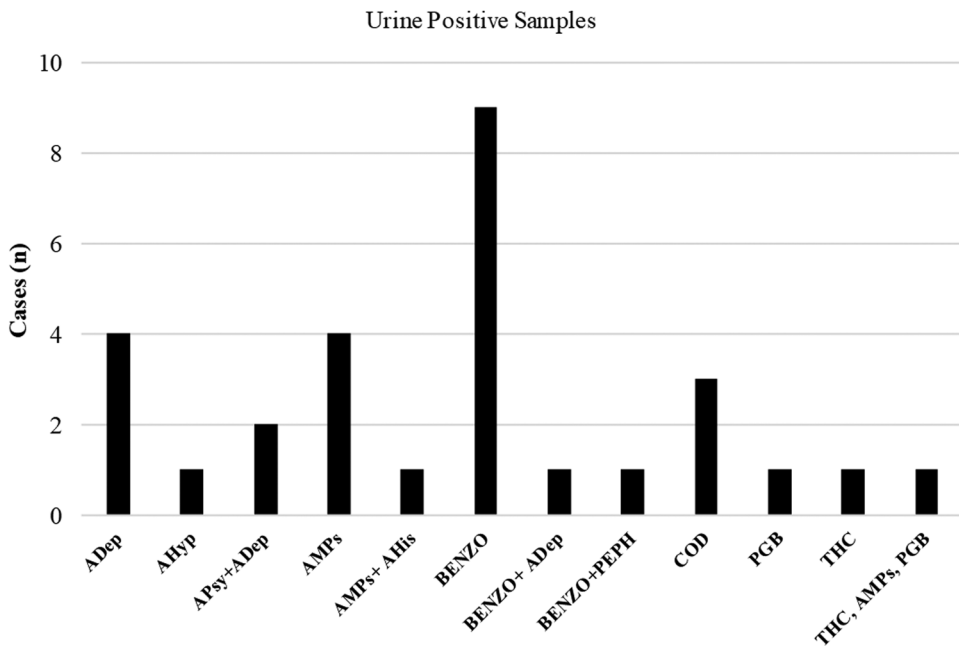


**Figure 4.** Distrubution of detected substances in total urine samples.

\*ADep: Antidepressant, ADia: Antidiabetic, AEme: Antiemetic, AEpi: Antiepileptic AHis: Antihistamine, AHyp: Antihypertensive, ATS: Amphetamines, ANALG: Analgesic, ANEST: Anesthetic, APsy: Antipsychotic, BENZO: Benzodiazepine, COC: Cocaine, COD: Codeine, EPH: Ephedrine, PEPH: Pseudoephedrine, PGB: Pregabalin, THC: Cannabinoid.

et al., 2008; Walsh, 2008). In fact, positivity percentages for drug abuse on WDT in previous studies varied considerably between 1.6% and 30% due to relevant differences such as biological matrix, target substances, and considered cutoff values on their WDT practices (Dalén et al., 2000; EWDTs, 2010; Kazanga et al., 2012; Santoro et al., 2012; Verstraete & Pierce, 2001).

Different substance use profiles between provinces make it difficult to combine the results on a common basis. Comparing the provinces of Adana and Izmir, which have a high rate of illicit drug use, it is an expected result that the demand for workplace drug tests and positive results in Adana an important and risky region are higher in terms of substance use in Türkiye. Although the total number of cases from two cities over 6 years in this study is relatively low compared to published studies from other countries, the positivity rate in this current study (20.5%) is higher than in other studies. Although this study has limitations due to the different practices of the laboratories to be emphasized, for example it is important in terms of providing data in terms of workplace drug tests. The limited demand for WDT could be attributed to several factors, including the legal obligation to report criminal activities, the avoidance of testing expenses by employers, a lack of voluntary

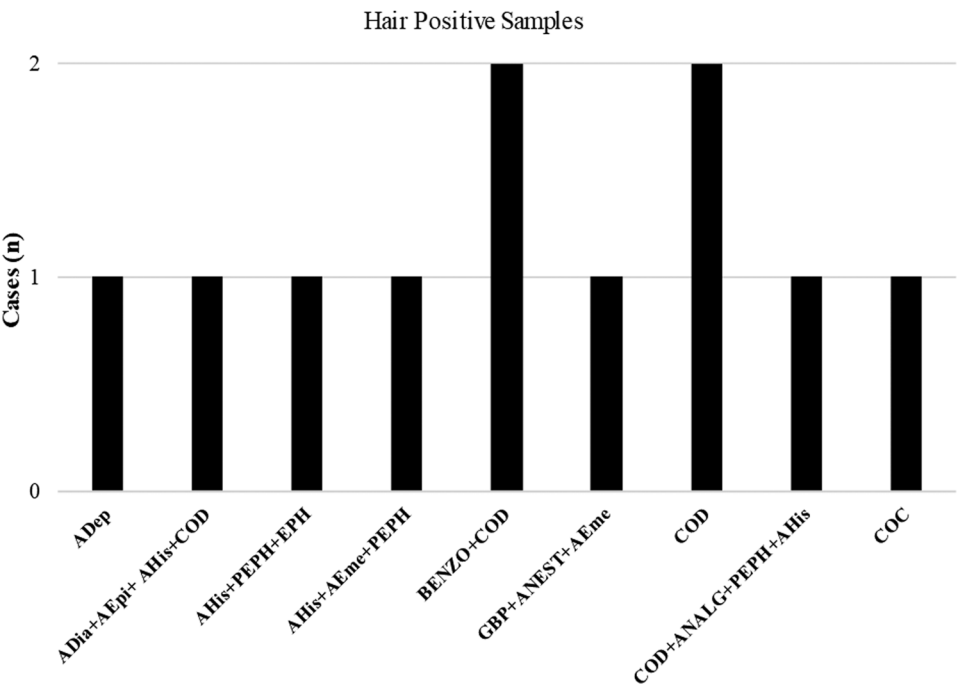


**Figure 5.** Distrubution of detected substances in hair samples.  
\*ADep: Antidepressant, ADia: Antidiabetic, AEme: Antiemetic, AEPi: Antiepileptic AHis: Antihistamine, AHyp: Antihypertensive, ATS: Amphetamines, ANALG: Analgesic, ANEST: Anesthetic, APsy: Antipsychotic, BENZO: Benzodiazepine, COC: Cocaine, COD: Codeine, EPH: Ephedrine, PEPH: Pseudoephedrine, PGB: Pregabalin, THC: Cannabinoid.

participation, and a general shortfall in awareness regarding the benefits and implications of WDT applications.

Several studies have enriched the literature on WDT practices globally. In a study by Santoro et al. involving Italian workers, cannabis emerged as the most frequently used substance, followed by cocaine, which aligns with annual data on illicit substance use (Santoro et al., 2012). A subsequent study on Italian workers yielded similar findings (Kazanga et al., 2012). In contrast to the Italian studies, the present research on Turkish workers highlights a notable prevalence of benzodiazepine positivity when considering both multiple and single substance use. When examining studies reflecting other substance in similar years, analyses of wastewater and surface water to identify the substance profile in Adana showed that benzodiazepines and antidepressants were among the most commonly abused therapeutic drugs (Daglioglu et al., 2019; Kuloglu Genc et al., 2021). In this study, the heightened incidence of benzodiazepine detection over illicit substances may be attributed to the prevalent use of gastro-protectors containing benzodiazepine among Turkish workers, who exhibit a higher average age ( $43.7 \pm 11.8$ ) compared to participants in WDT studies conducted in other countries (Kazanga et al.,

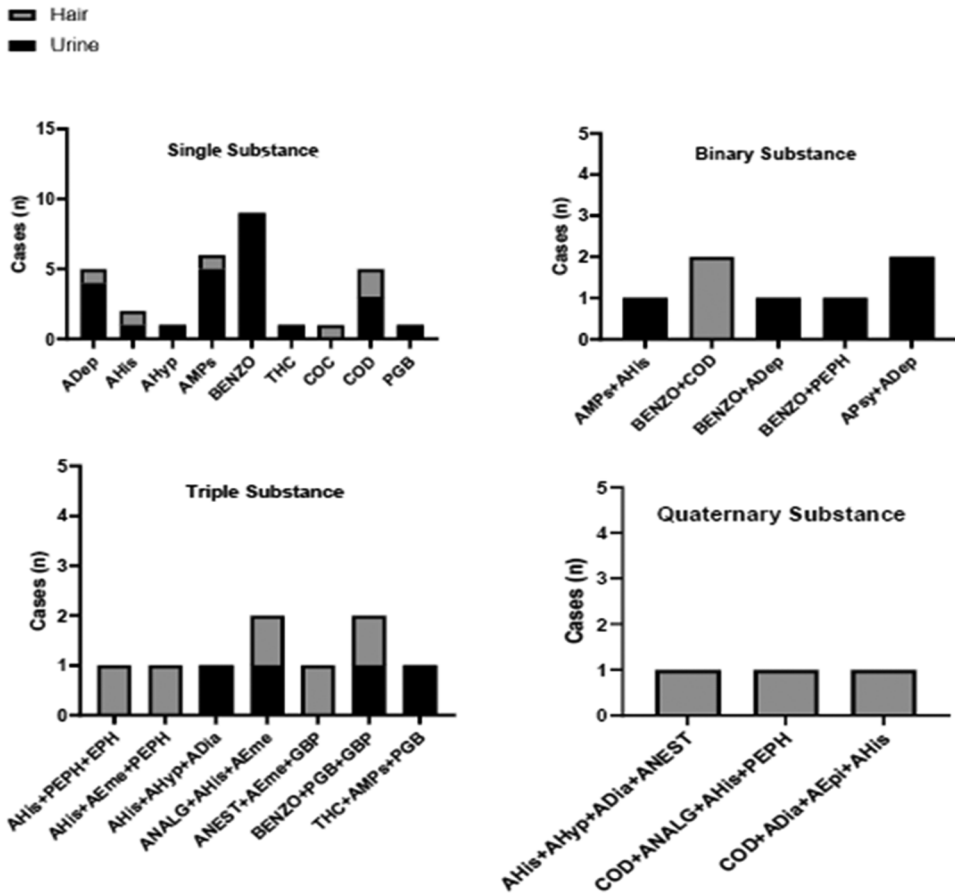


**Figure 6.** Distribution of comorbid substance use data in hair and urine in LC-MS/MS analysis. \*ADep: Antidepressant, ADia: Antidiabetic, AEme: Antiemetic, AEpi: Antiepileptic, AHis: Antihistamine, AHyp: Antihypertensive, ATS: Amphetamines, ANALG: Analgesic, ANEST: Anesthetic, APsy: Antipsychotic, BENZO: Benzodiazepine, COC: Cocaine, COD: Codeine, EPH: Ephedrine, PEPH: Pseudoephedrine, PGB: Pregabalin, THC: Cannabinoid.

2012; Rosso et al., 2017; Santoro et al., 2012). The main data that should be emphasized is the use of multiple substances in Adana. Drugs used in treatment, such as antipsychotic, antidepressant, antihistaminic, and benzodiazepine, were detected in multisubstance combinations in urine and hair. Although it may be possible to determine the purpose (medical or abuse) for which the workers intake this kind of psychoactive drugs at a later stage of the forensic investigation, regulations, and additional procedures to be added to WDT applications could allow the decision-making mechanism to be faster and more accurate. Additionally, incorporating e-prescription details of the individuals into the application files, with their consent and approval, could significantly streamline the judicial investigation process, thereby enhancing the efficacy of WDT policies.

The European Drug Report 2022 highlights that ATs (amphetamine, methamphetamine, ecstasy/MDMA etc.) are the most abused potent central nervous system stimulants worldwide after cannabis. The detection of ATs rather than cannabis in our study may be related to the recent increase in methamphetamine use in our country. For instance, in 2020, Türkiye reported a significant increase in methamphetamine seizures, with over 4 tonnes seized, a substantial rise from the 1 tonne seized in 2019 (European Monitoring Center for Drugs and Drug Addiction (EMCDDA), 2022). The strategic locations of Adana, a transit hub in the south, and İzmir, a port city, may have influenced drug use patterns and, consequently, the positivity rates observed in WDT. Moreover, according to the 2022 EMCDDA report, which includes drug analyses in wastewater, Adana was ranked 16th among 106 European centers in methamphetamine usage, based on wastewater treatment plant samples (EMCDDA, 2023).

The escalation in the non-medical utilization and abuse of prescription medications represents a significant public health concern in numerous countries, including Türkiye. A substantial portion of individuals who abuse these substances harbor the belief that prescription drugs are safer and more trustworthy than illicit substances, coupled with the fact that they are more readily accessible, which contributed to the increase in the consumption of these prescribed drugs (Schifano et al., 2021). Pregabalin was included among the controlled prescription drugs in our country in 2019. Besides medical purposes, data and case reports about pregabalin's potential to be abused and subsequent addiction have increased in recent years (Grosshans et al., 2013; Ovat et al., 2024; Schifano, 2014; Teker et al., 2024; Yargic & Alyanak Ozdemiroglu, 2011). It is known that pregabalin, which is not included in the routine primary WDT analysis scale (European Workplace Drug Testing Society, 2022), is used to prevent the criminal consequences of illicit drug use or traumatic narrative and current living conditions that are difficult to sustain (Servais et al., 2023). It should be noted as a limitation of this study that the inclusion of PGB in the testing protocol was delayed due to the lack of a reference standard for PGB in the laboratory and was first included in the toxicological analysis framework for Workplace Drug Testing (WDT) in Adana in 2021. The increase in the abuse of prescription-controlled drugs should not be overlooked in the evaluation of WDT for workplace safety, productivity, and worker health.

## 5. Conclusions

This study showcased WDT practices across two distinct regions in Türkiye, both of which hold significant relevance in terms of drug abuse. Despite the absence of routine WDT application, variations in the application of non-standardized practices are evident across different areas within the

country. The findings of the current work underscore the necessity for new regulations aimed at enhancing workplace safety and the well-being of workers and the general public impacted by these practices.

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