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Original Research Article

Investigation of the Impact of Various Type of Drug Abuse on the Potential Hydrogen, Specific Gravity, and Creatinine Level within Human Urine Samples

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Abstract:

The study aimed to assess the impact of various drugs of abuse (DOA) on urine parameters, specifically potential hydrogen (pH), specific gravity (SG), and creatinine (CART) levels within a dataset of 401 human urine samples. The Enzyme-Linked Immuno-Assay (ELISA) method was employed to measure these parameters, and subsequent statistical analysis using Systat statistical software was conducted following data cleaning. When comparing the pH, SG, and CART values of samples with and without DOA presence, no statistically significant differences were observed. Initially suggesting that the presence of DOA does not substantially affect these urine parameters. However, these preliminary findings may imply that pH, SG, and CART levels in urine samples are not directly influenced by the presence of DOA; instead, other factors such as diet, hydration, and individual physiology may play a more significant role. The limited dataset size may contribute to this outcome, emphasizing the need for a more comprehensive investigation with a larger and more diverse dataset. In future, further analysis will be carried out with a control group with carefully collected, unadulterated urine samples from healthy individuals with larger dataset. It will allow the meaningful comparisons and a deeper understanding of the relationship between DOA and urine parameters. Furthermore, various alternatives parameters will be explored to improve drug abuse detection and prevention strategies.

Keywords: Forensic Science, Urine, pH, SG, Specific Gravity, Creatinine, Drugs Of Abuse, DOA, Urine Drug Testing, Sample Validity, Forensic Toxicology.

Introduction

Drug abuse is a pervasive global public health and forensic concern, driving the need for reliable drug detection methods in both clinical and criminal justice settings [1,2]. Urine-based testing for DOA has become particularly indispensable due to its non-invasive nature, rapid results, and broad detection window. Over the past few decades, many countries have widely adopted urine DOA screening in workplaces, criminal justice, and healthcare programs. For example, the United States saw a 66% decrease in positive workplace urine tests from 1988–2004 even as self-reported drug use rose, highlighting both the impact of testing programs and the challenges of evasion [3]. In East and Southeast Asia, methamphetamine has emerged as the most commonly detected illicit drug in urine testing – notably dominating abuse data in regions like Brunei, Japan, the Philippines, UK and Korea [4]. In Taiwan, analyses of arrestee urine samples from 1999–2011 showed methamphetamine to be the single most prevalent illicit substance, outpacing even heroin [5,6]. In the Middle East, drug use trends are shifting as well. Gulf Cooperation Council (GCC) countries have historically enforced strict anti-drug policies, yet recent reports indicate growing misuse of prescription medications such as tramadol and pregabalin. In the United Arab Emirates (UAE), a cohort study of rehabilitation patients found that 67.2% of opioid users misused tramadol, far exceeding heroin use. The same study noted extensive non-medical use of pregabalin (averaging over 8 capsules daily) among poly-substance abusers, underscoring the rising regional challenge of prescription drug abuse. These global and regional patterns underscore the forensic importance of robust DOA testing programs both to inform public health interventions and to enforce drug laws in places like the UAE and the broader GCC [7]. Urine drug testing (UDT) protocols typically involve an initial immunoassay screening followed by confirmatory testing. ELISA and other immunoassays are popular for preliminary screening due to their speed and high throughput. Indeed, immunoassay-based urine screens for

amphetamines, benzodiazepines, cannabis (THC), opiates, and other drugs are routine in many laboratories [8]. However, immunoassays have known limitations, including cross-reactivity and finite sensitivity, which can yield "false-positive" or "false-negative" results [9]. For example, over-the-counter medications and innocuous compounds may trigger false positives, while certain synthetic or semi-synthetic drugs may go undetected. To ensure accuracy, presumptive positives from immunoassays are always confirmed using specific techniques like gas chromatography–mass spectrometry (GC–MS) or liquid chromatography–MS, which can unequivocally identify drug compounds at low concentrations. These confirmation methods significantly reduce false results by separating individual drugs and metabolites and detecting them with high specificity. Given the cost and expertise required for confirmatory assays, UDT programs must balance comprehensive detection with practical feasibility [10]. In the UAE and GCC, where a wide range of substances – from traditional narcotics to prescription analgesics – are encountered, testing protocols have been adapting to include newer drug panels and more sensitive assays. Internationally, there is also movement toward on-site rapid testing and alternative specimens (saliva, sweat, hair) to supplement urine screens, aiming to deter tampering and expand the situations in which drug testing can be performed [11]. Such innovations in testing protocols reflect a global impetus to strengthen drug screening as a public health tool.

A persistent concern in urine-based DOA testing is sample adulteration and substitution, wherein donors intentionally tamper with urine specimens to produce a false-negative result [12,13]. Common adulteration techniques include diluting the urine (e.g. by consuming excessive fluids or adding water) and adding household chemicals or commercial adulterant products to the sample. These adulterants – ranging from simple substances like water, vinegar, lemon juice, salt, or bleach, to commercial products containing glutaraldehyde (e.g. "UrineAid"), nitrites (e.g. "Klear"), or oxidants (e.g. "Stealth") – can significantly distort immunoassay results. For instance, one study found that adding vinegar (acetic acid) to urine could abolish positive immunoassay signals for almost all drug classes, yielding false-negatives while barely altering the urine's appearance. Similarly, oxidative adulterants like bleach or nitrite can destroy THC metabolites and other drug compounds, or interfere with the antibody-based detection, thereby producing false-negative outcomes. Without safeguards, these forms of tampering can severely undermine the integrity of drug screening programs [14]. To combat this, laboratories employ specimen validity testing (SVT) alongside drug assays. SVT involves measuring urine parameters such as pH, SG, CART, and checking for oxidants/nitrites to detect dilution or adulteration. A urine sample with abnormally low CART and SG is flagged as dilute, indicating possible water-loading; conversely, presence of oxidants or an extreme pH may signal chemical adulterants. Guidelines define quantitative cut-offs: for example, the U.S. Substance Abuse and Mental Health Services Administration (SAMHSA) considers urine with CART <20 mg/dL and SG <1.0030 as "dilute," and CART <5 mg/dL with SG ~1.000 as "substituted" (non-human) [15]. Routine urine CART analysis has proven to be a simple and effective authenticity check that can greatly reduce false-negatives by flagging overly dilute samples. Lafolie et al. (1991) demonstrated that incorporating CART criteria into DOA screening prevented many false-negative results that would have occurred from undetected dilution [16]. Thus, pH, SG and CART serve as crucial indicators of sample integrity: a physiologically normal range (approximately pH 4.5–8.5, SG ~1.005–1.030, CART >20 mg/dL) is expected in genuine urine, whereas values outside these ranges raise suspicion of adulteration or invalid samples. Modern urine testing programs, including those in the GCC, have increasingly standardized specimen validity criteria to ensure that a "negative" drug test truly means drug-absence and not sample tampering. However, despite these measures, the ingenuity of cheaters continues to pose challenges. Recent evaluations of adulterant-detection kits (e.g. specialized test strips like CEDIA® Sample Check) reveal that not all adulterants are identified; in one study, only 5 out of 9 deliberate adulterations were detected by the SVT strip, allowing certain additives (such as sodium azide) to evade detection and produce undetected false-negatives [13,17]. This highlights an ongoing "arms race" in forensic toxicology between adulteration methods and detection techniques. Laboratories must continuously update their validity testing protocols (for example, adding tests for novel adulterants or using more sensitive instrumentation) to close these gaps.

In parallel with classical wet-lab techniques, the emerging role of advanced data analytics and artificial intelligence (AI) in toxicology warrants discussion. As datasets from drug testing programs grow (e.g. thousands of urine results with multiple variables), statistical computing tools have become invaluable for uncovering patterns and trends. In this study, for instance, we applied Systat software for statistical comparisons and Python libraries for data visualization (such as heatmaps) to interpret the relationships between DOA presence and urine parameters. The use of Python and similar data science platforms in forensic science enables reproducible, flexible analysis from simple t-tests to complex multivariate modeling which enhances the rigor of results. More broadly, AI and machine learning techniques are being explored in toxicology

laboratories worldwide [18]. These methods can detect subtle, non-linear patterns in large datasets that human analysts might overlook. Potential applications range from flagging atypical result combinations that could suggest adulteration or novel psychoactive substances, to predicting an individual's drug use trajectory or relapse risk based on their longitudinal test data. Early reports highlight AI's promise in improving both the efficiency and accuracy of drug screening, for example by rapidly analyzing mass spectrometry outputs or by correlating patient factors with drug metabolism to predict false-negatives. While still in nascent stages for forensic applications, such approaches could complement traditional analysis. In the context of urine DOA testing, one could imagine a machine learning model that, given a large repository of test results, learns to identify an "adulterated" urine sample pattern or even suggests when a negative immunoassay might be discrepant with expected pharmacokinetics (flagging it for confirmatory re-testing). Incorporating these innovative tools aligns with the UAE's broader interest in smart technologies and may pave the way for more robust drug surveillance systems. Of course, algorithmic approaches will require validation and oversight, but their inclusion in toxicology research signals a forward-looking trend in the field [19].

An additional motivation for ongoing research in DOA testing is the development of low-cost, non-invasive alternatives to standard assays. Urine testing itself is non-invasive compared to blood draws, but it still faces issues like sample adulteration and privacy concerns during collection. This has led to investigations of alternative biological matrices (saliva, sweat, hair) which are harder to adulterate and can be collected more conveniently. Oral fluid (saliva) testing, for example, allows for directly observed collection and has seen increasing use in roadside and workplace screenings. While these alternatives can reduce cheating, they come with their own limitations (such as shorter detection windows for saliva and higher costs for hair analysis), so urine remains the gold-standard matrix for broad DOA screening. Therefore, improving urine tests themselves – making them more foolproof and cost-effective; is an ongoing priority. One approach is to refine point-of-care testing kits to be cheaper and more reliable, which could benefit from the findings of studies like this. If it is confirmed that drugs do not meaningfully affect urine pH, SG, or CART, then resources can be focused on better direct detection of drugs and better detection of adulterants, rather than pursuing indirect markers [20]. On the other hand, if any subtle patterns are observed (even something as simple as chronic drug users having, say, marginally more dilute urine due to polydipsia), these could spur development of new screening indicators or risk scores. Ultimately, the goal is to enhance DOA screening in a way that is accessible and trustworthy. This is particularly crucial for the UAE/GCC region, where expanding testing capacity and coverage is needed alongside prevention efforts. Some authors have noted a relative lack of published research on substance abuse trends and testing outcomes in the GCC [21], suggesting that current efforts might benefit from more data-driven policy. By investing in low-cost screening innovations and data analysis, GCC countries can improve early detection of drug abuse while keeping testing feasible in large populations.

Overall, the literature reveals that urine-based drug testing is a well-established but continually evolving practice at the intersection of forensic science and public health. Immunoassay-based urine screens (such as ELISA) and confirmatory GC/MS remain the cornerstone of DOA detection, and their use in the UAE and GCC provides a critical line of defense against drug misuse. At the same time, specimen validity parameters including pH, specific gravity, and CART are essential tools to ensure sample integrity and to thwart adulteration attempts [22]. The consensus from prior studies is that these urine parameters are influenced by dilution and adulterants, but not inherently by the presence of drugs.

The present study is motivated by the need to confirm this understanding in a local context and to explore any nuanced interactions between drug use and urine properties in samples from the UAE. By statistically evaluating pH, SG, and CART in DOA-positive versus DOA-negative urine samples, we aim to determine whether drug abuse leaves any detectable "fingerprint" on these routine urine measurements. This work also addresses a gap in regional data, providing one of the first systematic looks at urine sample validity markers in relation to prevalent drugs in the Gulf. The findings could reinforce best practices in urine drug screening, for instance, underscoring that a normal pH/SG/CART does not rule out drug use and guide local laboratories in focusing on proven indicators of adulteration. Furthermore, our integration of statistical software and Python-based analysis exemplifies the value of applying modern data science techniques to forensic toxicology questions.

In closing, the literature suggests that while urine DOA tests are robust, maintaining their integrity is an ongoing challenge. Continued research is recommended to (a) improve detection of ever-evolving adulterants [23] (b) update testing panels to include emerging drugs (e.g. synthetic cannabinoids, designer stimulants), and (c) harness new analytical tools (including AI and novel biomarkers) to enhance the sensitivity and cost-effectiveness of drug screening programs. By addressing these areas, forensic scientists and public health authorities can

strengthen urine-based DOA screening, thereby bolstering drug abuse prevention and enforcement efforts in the UAE, the GCC, and worldwide. The current study contributes to this endeavor by shedding light on the interaction between DOA and urine characteristics, and by evaluating the utility of basic urine parameters as part of a comprehensive drug testing strategy.

Materials and Methods

Chemicals: Commercially available assay kits for amphetamine, benzodiazepine, cannabis, cocaine, opiates, phencyclidine, and propoxyphene were obtained from Siemens Healthcare Diagnostics Inc., Newark, USA. Each kit included analyte-specific reagents, calibrators, and quality control materials validated for forensic toxicological analysis. Assays for pregabalin and tramadol were acquired from Ark Diagnostics Inc., Fremont, USA. These kits were selected based on their high sensitivity, specificity, and routine use in forensic and clinical toxicology for initial screening and semi-quantitative assessments.

Materials and Instrumentation Urine samples were collected by trained personnel from individuals under investigation or enrolled in controlled monitoring programs managed by the Drugs Control Department. Total of 401 samples were collected. The collections were conducted in accordance with standard forensic toxicology protocols to ensure the integrity and admissibility of specimens used for drugs DOA testing. Individuals were instructed to provide a midstream urine sample in sterile, tamper-evident containers under supervised conditions to reduce the risk of adulteration or substitution. No dietary or medication restrictions were imposed prior to collection, reflecting typical real-world forensic screening environments.

Each specimen was immediately labeled with a unique identification code and documented using standardized chain-of-custody procedures. Temperature readings were taken within 4 minutes of voiding to confirm sample freshness. The samples were temporarily stored at 2°C to 8°C and transported under controlled conditions to the toxicology laboratory for analysis. Upon receipt, specimens were inspected for signs of tampering and recorded into the laboratory's information system for further testing. Analyses were performed using the V-Twin ELISA analyzer from Siemens Healthcare Diagnostics. This system enables high-throughput screening of multiple drug classes in urine matrices. Calibration and quality control procedures were executed following manufacturer instructions. Specific gravity and pH were measured using Siemens-compatible multi-parameter urine analyzers, and CART levels were determined using the colorimetric Jaffe method, which is widely accepted in forensic laboratories.

Data processing and statistical evaluation were carried out using Systat software (Version 13). Supplementary validation and visual analytics, including heatmap correlation matrices, were created using Python libraries (e.g., Seaborn and Pandas).

Analytical Procedure: Prior to analysis, calibration curves for each DOA and urine parameter (pH, SG, and CART) were constructed using standard calibrators. Curve acceptability was evaluated by running internal quality control materials at both low and high concentrations to ensure accuracy and reproducibility.

Each urine sample was visually inspected for turbidity. Samples that appeared turbid were centrifuged at 3000 revolutions per minute for 3 minutes. Following centrifugation, supernatants were analyzed in duplicate to minimize technical variability. Samples that tested positive for any DOA group via immunoassay were flagged for further evaluation.

Results were recorded electronically and transferred to structured Excel worksheets for statistical analysis. Parameters were compared across groups categorized by DOA status and sample clarity. Thresholds for interpretation and abnormality were established based on standard clinical and forensic guidelines.

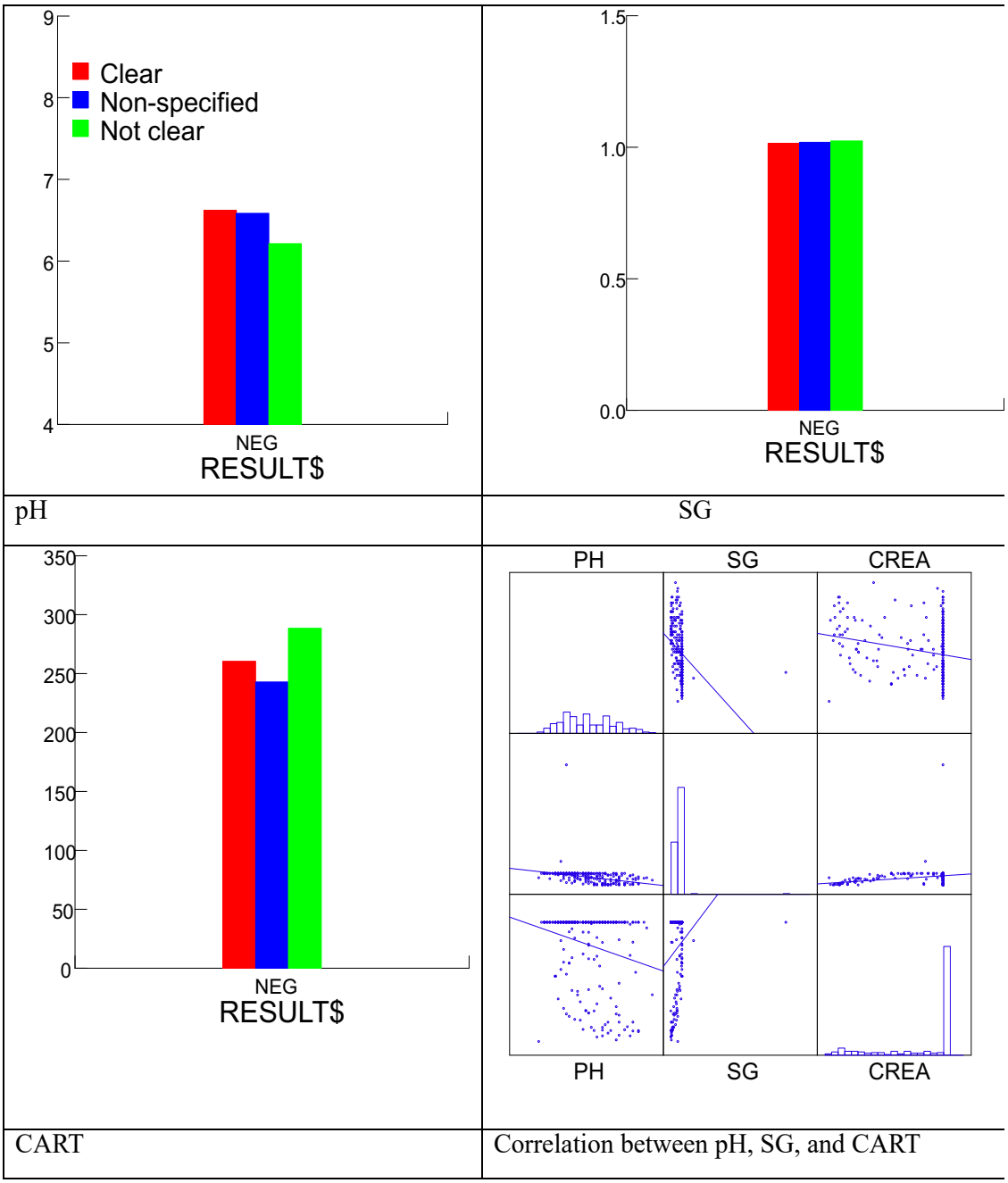
Results

Data Acquisition and preparation: A total of 401 human urine samples were comprehensively analyzed to evaluate the influence of various DOA on critical urine parameters, specifically pH, SG, and CART levels. The samples were methodically divided into two distinct groups based on DOA detection: negative samples (no DOA detected) and positive samples (presence of one or more DOA detected).

Negative Samples Analysis: Detailed observations of negative samples (Table-1) indicated noteworthy trends based on urine clarity classifications: clear, not clear, and non-specified. Samples identified as clear generally demonstrated higher pH values compared to those categorized as not clear or non-specified, suggesting possible variations influenced by physiological or dietary factors unrelated to DOA. Specific gravity, which reflects urine concentration and hydration status, showed minimal variance across all clarity groups. Nonetheless, marginally elevated SG values were recorded in not clear samples, potentially indicative of minor differences in hydration levels or solute concentrations. CART, a critical indicator of renal function and sample validity, was consistently higher in samples labeled not clear, possibly reflecting physiological or metabolic factors. Figure-1 (Correlation Heatmap) elucidated these

observations, confirming no significant statistical correlations between pH and SG, or pH and CART levels. However, a distinct positive correlation between SG and CART was prominently evident, reinforcing the interdependence of urine concentration and renal clearance markers.

Table 1: Negative results affecting pH, SG, and creatine according to the physical properties.



pH | SG

CART | Correlation between pH, SG, and CART

Bar charts show that clear samples exhibited higher pH, while not clear samples had slightly elevated SG and creatinine levels. Variations were minor and not statistically significant.

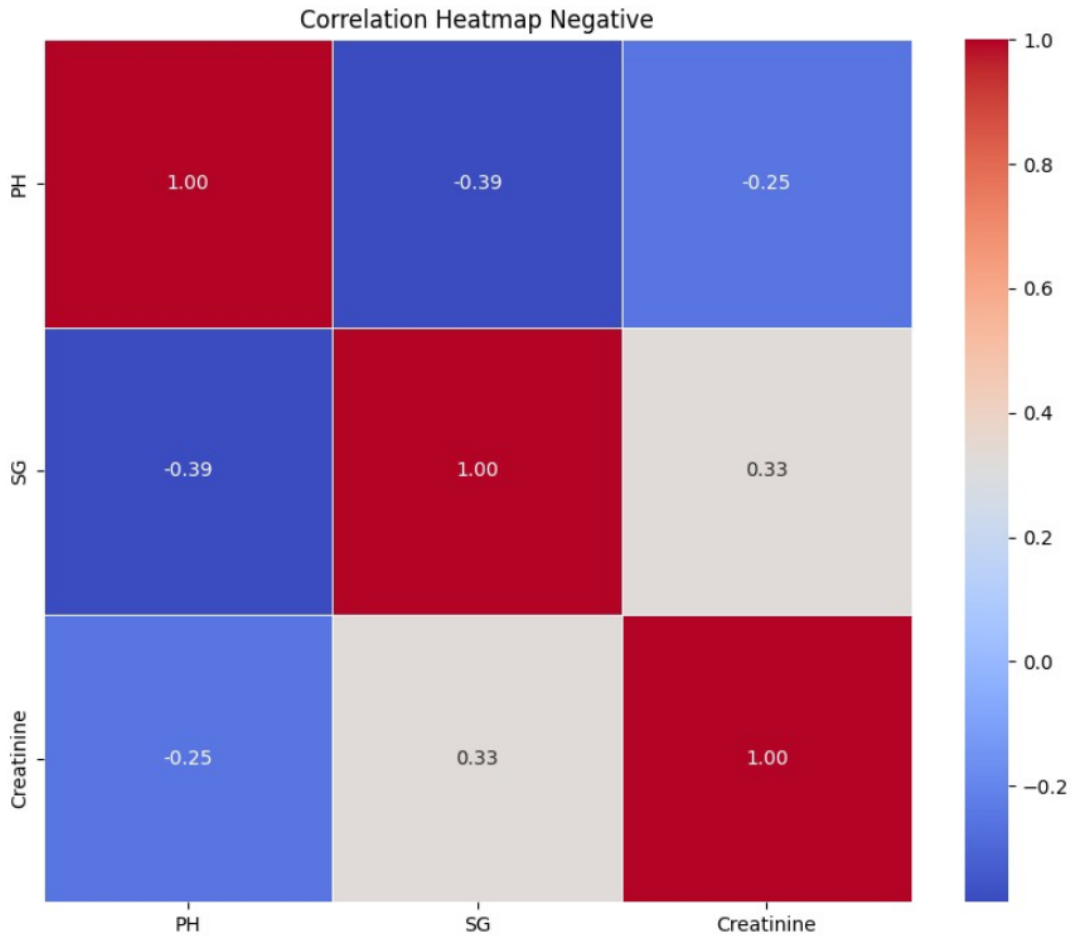


Figure 1: Correlation heatmap of urine parameters in DOA-negative samples.

Color intensity ranges from blue (−0.2) to red (+1.0). A strong positive correlation was observed between SG and creatinine, while pH showed no correlation with either.

Positive Samples Analysis: Analysis of positive samples (Table-2) revealed intriguing associations among the tested substances, including amphetamines (AMPH), benzodiazepines (BENZO), cannabis (THC), pregabalin, and tramadol. Samples classified as clear exhibited notably higher pH values, particularly associated with AMPH and BENZO, possibly reflecting specific metabolic pathways or excretion mechanisms linked to these substances. Specific gravity presented minimal yet observable variations between non-specified and not clear classifications, with THC-positive samples in the not clear category exhibiting notably lower SG values. This finding could indicate distinct excretion patterns or hydration status among THC users compared to other substances. CART levels remained relatively consistent across clarity classifications, although notably lower values were documented in THC-positive samples, potentially signaling altered renal function or metabolic excretion pathways in users of this substance. The correlation analysis displayed in Figure-2 reiterated the absence of significant correlations between pH and SG or pH and CART, but robustly highlighted the strong positive correlation between SG and CART, supporting the physiological interrelation of these parameters.

Specific Gravity Stratified Analysis (SG ≤ 1.025 and SG > 1.025): Further stratified analysis based on specific gravity thresholds (Table-3) yielded critical insights into physiological and hydration factors influencing urine parameters independently of DOA presence. In samples with SG ≤ 1.025, significantly elevated pH values were observed, potentially indicative of a diluted urine state associated with increased fluid intake or lower solute excretion. Conversely, samples exhibiting SG values greater than 1.025 demonstrated significantly elevated CART levels, aligning with expectations of concentrated urine typically resulting from reduced fluid intake or increased solute excretion. This stratified analysis notably did not identify a relationship between pH and CART, highlighting the independence of these parameters under varying hydration conditions.

Overall, comprehensive statistical analyses conducted herein highlight that the presence of DOAdoes not significantly influence urine pH, specific gravity, or CART levels. Observed

variances within these parameters likely reflect individual physiological characteristics, dietary influences, and hydration status rather than direct effects of drug abuse.

Table 2: Positive results affecting pH, SG, and creatinine according to physical properties.

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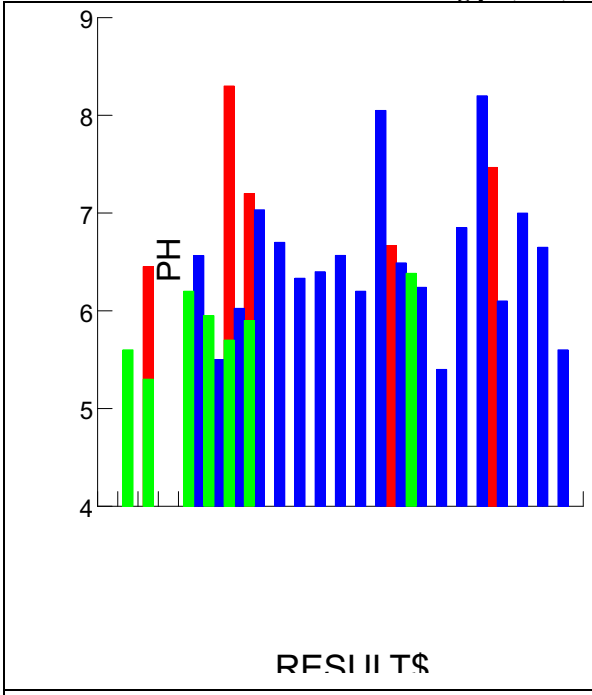
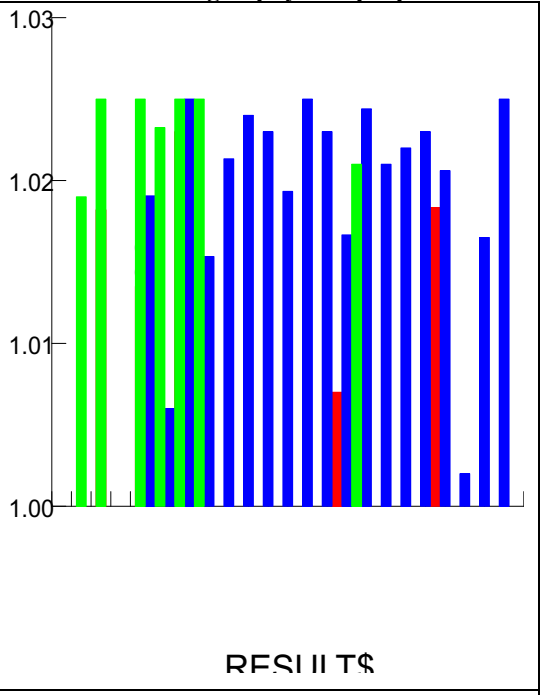
<tr><td> 
</td><td> 
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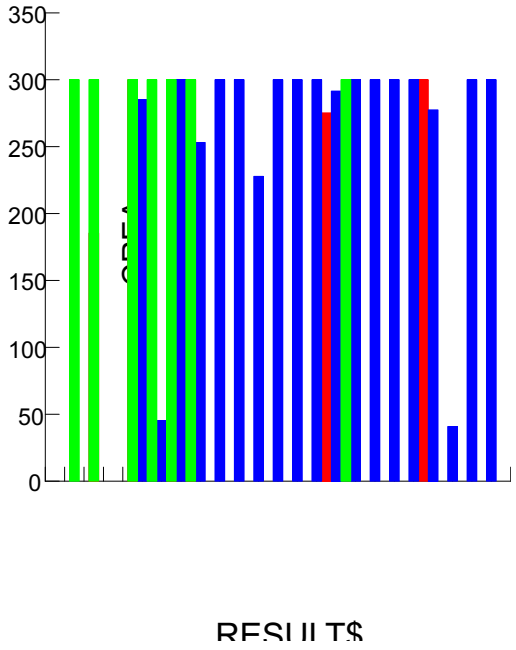
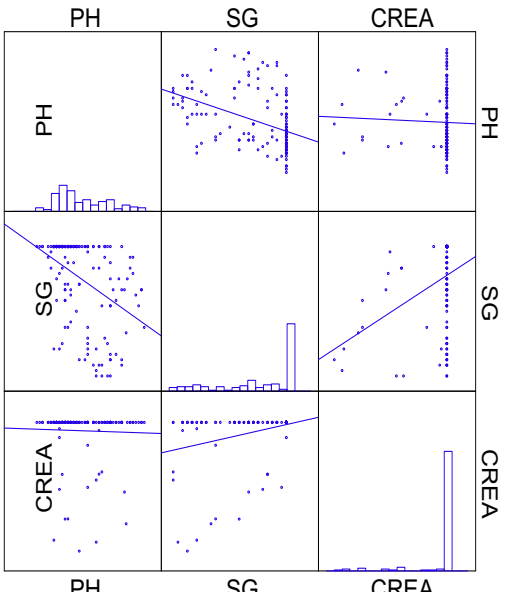
<tr><td> 
</td><td> 
</td></tr>
<tr><td>CART</td><td>Correlation between pH, SG, and CART</td></tr>
<tr><td colspan="2">Bar charts show higher pH in clear samples associated with AMPH and BENZO. SG and creatinine values showed minor variation across groups, with THC-positive samples tending toward lower values.</td></tr>
</table>

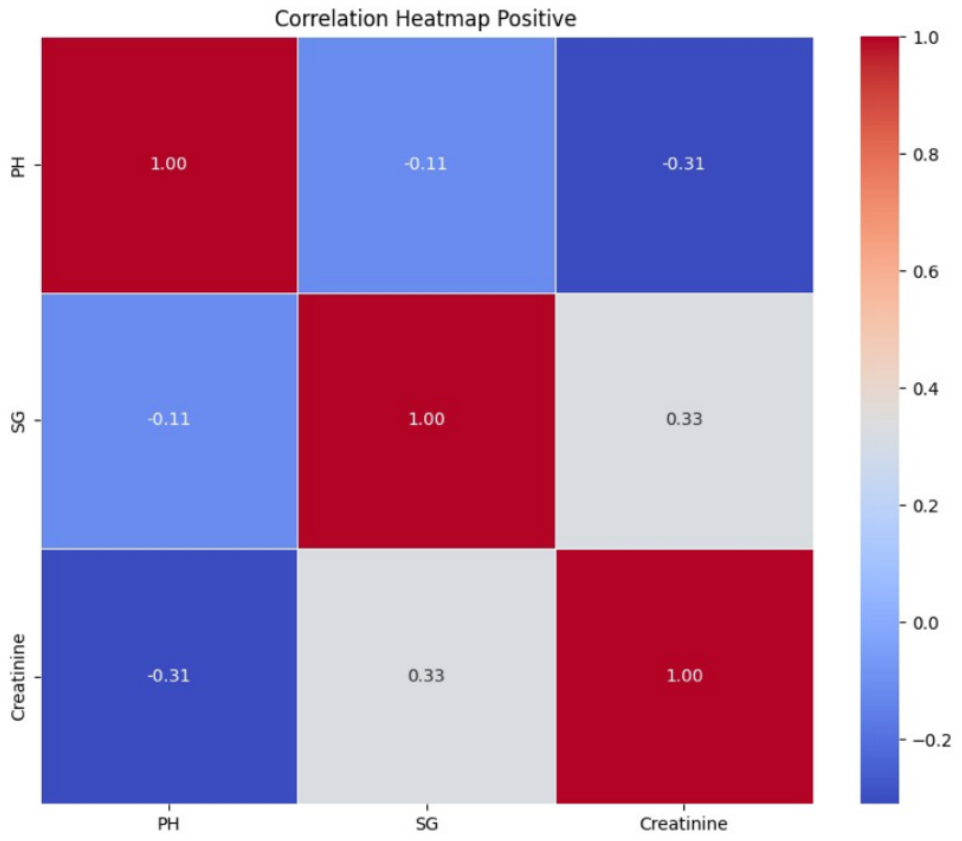


Figure 2: Correlation heatmap of urine parameters in DOA-positive samples.

Color gradient ranges from blue (−0.2) to red (+1.0). A strong positive correlation was observed between SG and creatinine, while no correlation was found between pH and the other parameters.

Discussion

The findings of this study indicate that the presence of DOA in urine samples does not result in statistically significant alterations in fundamental physicochemical parameters such as pH, SG, and CART. This observation aligns with prior reports suggesting that these parameters are more strongly influenced by physiological conditions, hydration levels, and dietary habits rather than direct pharmacological action of illicit substances [12,15,16].

The lack of significant variation between DOA-positive and DOA-negative samples implies that routine measurement of pH, SG, and CART remains a valid strategy for evaluating sample integrity but may not directly reflect drug use status. However, a consistent and statistically positive correlation between SG and CART across both positive and negative groups suggests that these two indicators should be interpreted together when assessing sample validity, particularly in suspected dilution cases. This relationship has been highlighted in previous research where CART normalization was used to adjust drug metabolite concentrations for more accurate interpretation [16,24].

It is also notable that THC-positive samples tended to show slightly lower CART and SG values. While the variation was not statistically significant, it raises potential considerations for future research on substance-specific renal excretion patterns or hydration behaviors associated with cannabis use. Other studies have also pointed to varying renal handling and urine concentration tendencies depending on the drug class [22,25].

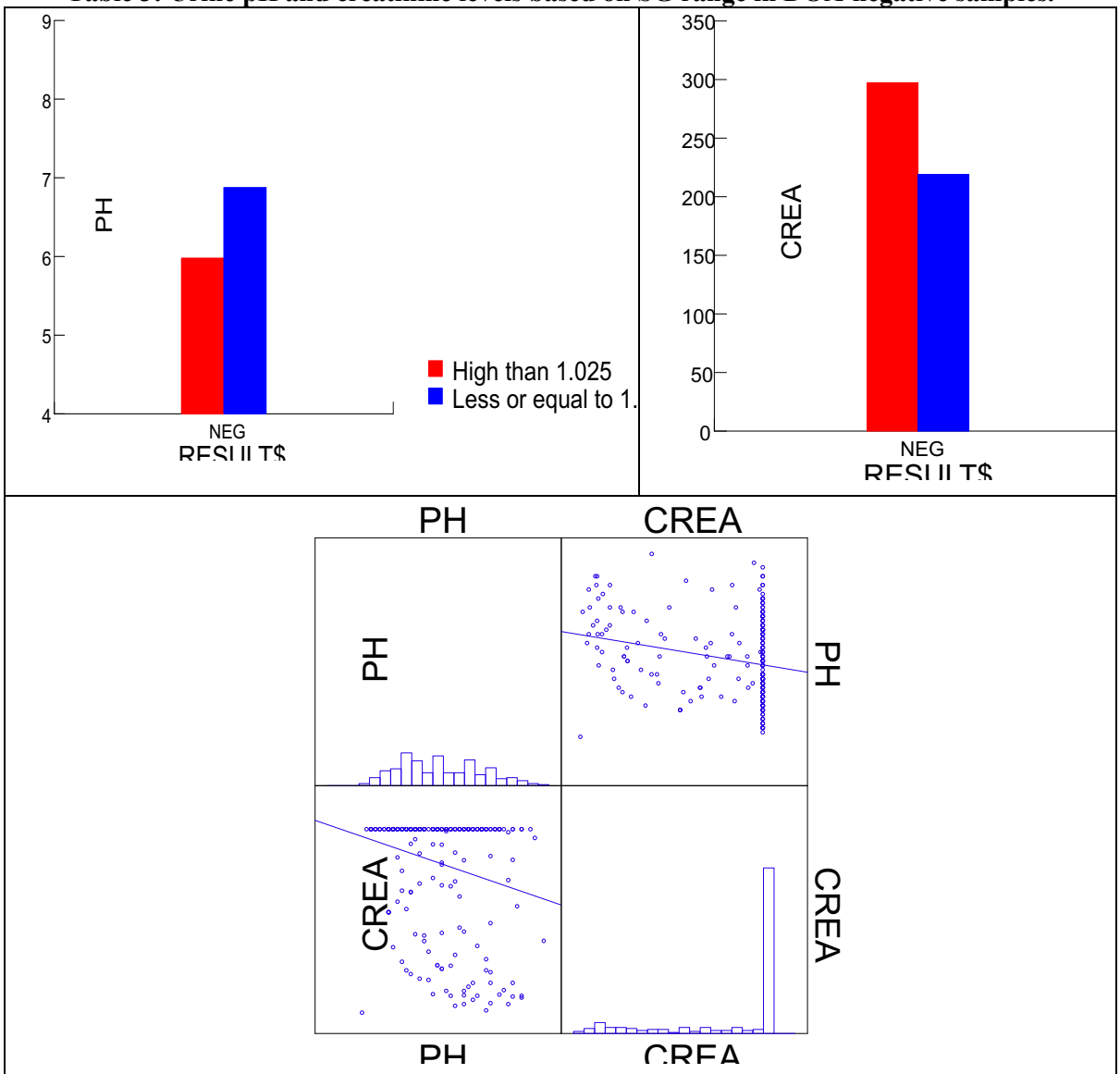
Furthermore, pH values did not significantly differ between DOA-positive and DOA-negative samples, suggesting that the acidic or basic nature of drug metabolites does not considerably affect the overall urine pH. However, when comparing stratified SG ranges, the data did show that samples with SG $\leq$ 1.025 had significantly elevated pH values and those with SG > 1.025 had higher CART concentrations. These findings reinforce the complex interplay between hydration and biochemical excretion, which must be considered when interpreting urine drug test results.

Despite the lack of direct impact from DOA on urine parameters, this study underscores the importance of baseline assessments of pH, SG, and CART to flag potential tampering or dilution. Past studies have

emphasized that some commercial adulterants can significantly skew these parameters, especially SG and pH, to produce false-negative drug test results [17]. Integrating these routine measurements into initial screening workflows provides critical context, especially when confirmatory testing by GC-MS is delayed or unavailable.

One limitation of the present study was the absence of a tightly controlled healthy baseline or negative reference population. All samples were collected under field conditions without restriction on diet or medication, which introduces natural variability. While this may limit internal control, it simultaneously increases the ecological validity of the study by reflecting real-world testing conditions in forensic or workplace scenarios. Future investigations should consider including control groups of verified drug-free individuals for stronger comparative analyses.

Table 3: Urine pH and creatinine levels based on SG range in DOA-negative samples.



Samples with SG ≤ 1.025 showed higher pH, while those with SG > 1.025 had elevated creatinine. No correlation was observed between pH and creatinine.

Another objective of this study was to explore the feasibility of using basic urine parameters, pH, specific gravity, and CART as potential proxies for predicting drug abuse status through statistical correlation and data science approaches. If meaningful relationships between these parameters and DOA presence had been established, this could have opened the possibility of using routine, low-cost urine analysis combined with machine learning to flag high-risk samples, thereby reducing reliance on immunoassay kits for initial screening. However, the findings revealed no consistent or statistically significant relationship between DOA presence and individual urine parameters. The only notable

correlation observed was between SG and CART, which is a well-established physiological association. These results suggest that while specimen validity markers are critical for ensuring sample authenticity, they cannot independently substitute or reliably predict DOA positivity. Nevertheless, the application of statistical tools like Systat and Python-based visualization methods demonstrated the value of integrating data science into forensic workflows. These tools may still offer future utility in identifying adulteration patterns or optimizing large-scale data-driven screening models when paired with expanded datasets and additional biological variables.

Overall, the study contributes valuable data to the field of forensic toxicology by confirming that urine validity markers remain essential tools for assessing specimen integrity but are not independently diagnostic of substance abuse. Integrating these parameters into standard screening and interpretive algorithms enhances the reliability of drug testing systems, especially in high-volume or resource-limited forensic environments.

Table 4: Summary of Urine Sample Parameters by DOA Status

DOA Status	Count	pH Mean ± SD	SG Mean ± SD	Creatinine Mean ± SD
Positive	164	6.25 ± 0.59	1.02 ± 0.01	114.81 ± 81.01
Negative	75	6.28 ± 0.62	1.02 ± 0.01	107.03 ± 61.57
Unspecified	162	6.41 ± 0.59	1.02 ± 0.01	96.13 ± 56.35

Table 4 shows the distribution of pH, SG, and CART levels across samples categorized by DOA status. 'Positive' refers to samples with one or more detected DOA, 'Negative' indicates explicitly reported drug-free samples, and 'Unspecified' are those with no information on drug status. Mean values are accompanied by standard deviations (SD) to reflect variability.

Table 5: Cut-off Values for Immunoassay Screen Kits

Drug Class	Cut-off Value (ng/mL)	Source / Reference
Amphetamines	500 (SAMHSA) / 300–1,000	Siemens EMIT II Plus (500 ng/mL standard) [26]
Benzodiazepines	200	Common immunoassay cut-off (Medigold Health matrix guidelines) [27]
Cannabis (THC)	50	Standard immunoassay cut-off (Medigold Health matrix guidelines) [27]
Cocaine (Benzoylecgonine)	150	Medigold Health [27]
Opiates	300	Standard immunoassay cut-off (Medigold Health matrix guidelines) [27]
Phencyclidine (PCP)	25	Standard immunoassay cut-off (Medigold Health matrix guidelines) [27]
Propoxyphene	300	Standard immunoassay cut-off (Medigold Health matrix guidelines) [27]
Pregabalin	500	ARK Pregabalin Urine Assay [28]
Tramadol	100	ARK Tramadol Assay [28]

This table summarizes the threshold concentrations applied to classify urine samples as positive for DOA. Cut-off values were derived from manufacturers' guidelines for the commercial immunoassay kits used, ensuring consistency with standard forensic toxicology practices.

Recommendations

Based on the findings of this study, several practical recommendations can be proposed to enhance the accuracy, efficiency, and cost-effectiveness of urine drug testing protocols. First, the routine inclusion of urine pH, SG, and CART measurements should be maintained as an essential part of all DOA screening workflows to ensure sample validity and detect potential adulteration or dilution. SG and CART should be interpreted in tandem when evaluating borderline or suspicious cases, as their combined assessment improves the reliability of detecting diluted samples. Further, the potential influence of specific drug classes on these urine parameters remains an area of interest and warrants detailed investigation under controlled conditions to determine whether chronic or acute drug use may subtly affect urine chemistry.

Institutions performing large-scale DOA testing are encouraged to establish population-specific reference intervals for pH, SG, and CART, accounting for local environmental, dietary, and

physiological factors. In addition, the integration of advanced adulterant screening techniques such as assays for oxidants and glutaraldehyde should be considered to enhance detection of chemically altered samples and reduce the incidence of false-negative results. Expanding the dataset to include rigorously confirmed negative and positive controls will also strengthen the interpretive value of these physicochemical parameters and improve generalizability.

Finally, the potential role of artificial intelligence and data science in DOA screening should be further explored. While current results do not support replacing immunoassay-based methods with indirect markers such as pH, SG, and CART, the application of machine learning models to large, well-annotated datasets may facilitate intelligent triaging of samples or flagging of cases requiring further investigation. This could be particularly beneficial in resource-limited settings, where cost-effective, data-driven approaches may serve as adjuncts to conventional drug testing strategies.

Conclusion

This study affirms that routine urine parameters including pH, SG, and CART are not reliable indicators of drug presence but remain critical for identifying sample manipulation. Their inability to distinguish DOA-positive from negative cases highlights the limitation of relying solely on basic urine chemistry for screening. However, the structured application of data science reveals potential in strengthening testing frameworks. While these parameters alone cannot replace biochemical assays, their integration into AI-driven models offers a viable path for improving efficiency, especially in resource-sensitive forensic environments. This direction warrants further exploration through larger datasets and algorithmic validation.

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