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The Stability of Urine Chemistry Strip Parameters Stored in BD Vacutainer® UAP Preservative Tubes

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Abstract

Objectives: Urinalysis requires that urine specimens maintain their biochemical and physical integrity from collection through to analysis. Delays between sample collection and laboratory processing can introduce variability due to chemical and cellular degradation.

Design & Methods: This study evaluated the stability of 11 urine strip parameters and two ratios in 100 samples preserved in BD Vacutainer® UAP tubes and stored at room temperature over a 48-hour period, using UC-3500 automated urine chemistry analyzer developed by Sysmex (Japan). Parameters analyzed included colour, appearance, pH, specific gravity, haemoglobin, leukocyte esterase, nitrites, proteins, ketones, albumin, creatinine, protein-to-creatinine ratio (PCR), and albumin-to-creatinine ratio (ACR). Samples were analyzed at baseline (0 hours), 24 hours, and 48 hours.

Results: Data showed minimal changes in all parameters except for a clinically acceptable increase in pH from 5.87 to 6.15 (Standard deviation (SD): 0.11). Specific gravity remained stable from 1015 to 1016 (SD: 0.8). Haemoglobin and leukocyte esterase slightly decreased in 6 samples, reflecting a minor degradation. Colour, appearance, albumin, creatinine, PCR, and ACR remained unchanged over time.

Conclusion: These findings support the efficacy of BD Vacutainer® UAP preservative tubes in maintaining urine sample stability at room temperature for up to 48 hours, thus highlighting their utility in clinical settings where immediate processing is impractical.

Keywords: Urinalysis; Vacutainer; Preservative Tube; Dipstick; Automated Urine Chemistry Analyser

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Introduction

Urinalysis remains a key diagnostic test for a wide range of clinical conditions, including urinary tract infections, kidney diseases, metabolic disorders, and systemic illnesses. The reliability of urinalysis results critically depends on the preservation of urine sample integrity from the point of collection through to laboratory analysis. Urine is a complex biological fluid subject to physicochemical and microbial changes post-collection, especially when immediate analysis is not performed [1]. Key challenges affecting urine stability include bacterial overgrowth, enzymatic degradation, and evaporation, which can lead to alterations in parameters such as pH, protein concentration, and cellular constituents [2]. Moreover, these preanalytical variables could cause significant diagnostic inaccuracies, including false positives or negatives, potentially impacting patient care [3].

Current guidelines from the Clinical and Laboratory Standards Institute (CLSI) and European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) recommend that urine samples should be analyzed within two hours of collection or stored at 2–8°C to delay degradation [4, 5]. However, logistical challenges in many clinical laboratory, including primary care, rural facilities, and large centralized laboratories, often preclude immediate analysis or refrigeration. In recent years, urine preservatives have been developed to stabilize samples by inhibiting bacterial growth and enzymatic activity. Several commercial preservative tubes are available to maintain the chemical and cellular stability of urine parameters, extending the possibility for reliable testing [6, 7]. Studies by Ercan and coauthors demonstrated that urine specimens stored without preservatives at room temperature rapidly degrade within 24 hours, while refrigeration or preservatives significantly

improve stability [8]. Karakoyun and coauthors further reported that chlorhexidine-based preservative tubes effectively maintain routine urinalysis parameters for up to 24 hours at room temperature [7].

Despite these improvements, comprehensive evaluations of the BD Vacutainer® UAP preservative tubes over extended periods, especially at room temperature, remain limited. This study aims to address this gap by assessing the stability of a broad panel of routine urinalysis parameters, including visual inspection, chemical, and calculated ratios (PCR and ACR), over 48 hours at room temperature in urine samples preserved using BD Vacutainer® UAP tubes.

Materials and Methods

Sample Collection and Preservation

One hundred urine specimens were analyzed. Midstream clean-catch urine samples were obtained according to standard guidelines [4, 5]. After collection, each sample was transferred into a BD Vacutainer® UAP preservative tube (Ref no. 365017; Becton, Dickinson and Co., Franklin Lakes, NJ, USA), which contains chemical preservatives designed to inhibit bacterial growth and enzymatic degradation. The BD Vacutainer® UAP are polyethylene terephthalate (PET) plastic tubes (8 mL) containing 0.4% chlorhexidine, 5.6% ethyl paraben, and 94% sodium propionate. All urine samples were stored at room temperature (22 ± 2°C) and protected from direct sunlight until analysis.

Methods

The study focused on urinalysis parameters spanning chemical and calculated indices: Colour, Appearance, pH, Specific Gravity, Hemoglobin, Leukocyte Esterase, Nitrites, Glucose, Bilirubin, Urobilinogen, Proteins, Ketones, Albumin, Creatinine, and the calculated Ratios Protein-to-Creatinine Ratio (PCR) and Albumin-to-Creatinine Ratio (ACR).

Chemical analyses were performed using the dipstick MEDITAPE UC-11A (Sysmex, Japan) and read on UC-3500 automated urine chemistry analyzer developed by Sysmex (Japan) including colour, appearance, pH, hemoglobin, leukocyte esterase, nitrites, proteins, ketones, albumin and creatinine. UC-3500 is a reflectance photometer instrument that measures the intensity of colorimetric changes in strip pads and converts them to categorical or semiquantitative results. Specific gravity was measured by refractometry. Samples were analyzed at three time points: baseline (0 hours), 24 hours, and 48 hours post-collection.

Statistical Analysis

Statistical analyses were performed using MedCalc for Windows, version 19.4 (MedCalc Software, Ostend, Belgium). Comparisons between groups were performed using the chi-square (χ^2) test and Fisher's exact test, where appropriate. The significance threshold was set at $p < 0.05$.

Cohen's kappa coefficient (κ) was calculated to evaluate the agreement of data. κ values in the ranges of 0.41–0.60, 0.61–0.80 and 0.81–1.00 were considered as moderate, substantial, and perfect agreement, respectively. A $\kappa > 0.80$ was considered as good agreement, indicating that the analytes were stable.

Results

At baseline (0 hours), all 100 urine samples displayed colours ranging from pale yellow to amber, and the appearance was predominantly clear, with only occasional mild turbidity consistent with normal variation. Over the 48-hour period, the samples remained visually stable with only minor subjective differences noted between observers, which were within expected variability and did not demonstrate any consistent temporal trend.

Chemical analysis did not reveal a statistically significant increase in mean pH from 5.87 at baseline to 6.15 (Standard Deviation (SD) = 0.11) after 48 hours ($p > 0.05$), suggesting limited biochemical alteration during storage. Specific gravity measurements showed no significant change over time, with mean values of 1015 at 0 hours and 1016 (SD = 0.8) at 48 hours ($p > 0.05$), respectively, indicating that solute concentration and hydration status remained stable and no significant evaporative loss occurred.

Haemoglobin and leukocyte esterase levels remained largely stable, although a minor decrease was detected in 6 of 100 samples for both parameters. This decrease likely reflects minor enzymatic degradation or haemolysis despite the presence of the preservative. However, it should be considered that the results for haemoglobin and leukocyte esterase may vary slightly due to subtle differences in the colour change of the test pad. However, these changes were minimal and did not affect clinical interpretation thresholds. Samples with negative nitrite results remained consistently stables, indicating effective inhibition of bacterial proliferation by the preservative.

Protein concentrations measured by dipstick analysis remained stable, with no significant variations. Similarly, ketones were negligible and remained unchanged over time points. Albumin and creatinine concentrations did not exhibit significant changes. Consequently, calculated indices such as the protein-to-creatinine ratio (PCR) and albumin-to-creatinine ratio (ACR) demonstrated excellent stability, with no statistically or clinically meaningful differences.

Glucose, Nitrite, and bilirubin were the stable constituents. The number of positive samples was the same throughout the 48 hours, with no changes. A total of 70 Nitrite-negative and 30 Nitrite-positive urine specimens were evaluated. The number of negative and positive specimens did not change. There were only 1 urine sample with high urobilinogen and 3 samples that were positive for ketones, but the results did not change at 48 h.

Overall, these findings indicate that the BD Vacutainer® UAP preservative tubes effectively maintain the integrity of multiple urine chemistry strip parameters, for up to 48 hours at room temperature. κ values underlined the agreement of the results obtained, ranging between 0.92 (haemoglobin and leukocyte esterase) and 1.00 (protein, albumin, creatinine, glucose, nitrite, ketones, and bilirubin) (Table 1).

Discussion

The stability of urine samples between collection and analysis is a critical preanalytical factor that significantly influences the accuracy of urinalysis results. This study demonstrates that BD Vacutainer® UAP preservative tubes effectively maintain the integrity of 14 routine urinalysis strip parameters when samples are stored at room temperature up to 48 hours. The results align with previous research emphasizing the importance of using preservatives or refrigeration to prevent the degradation of urine constituents. Ercan and co-authors reported that urine samples stored at room temperature without preservatives exhibited marked deterioration in both chemical and cellular components within 24 hours. Conversely, the use of preservatives or refrigerated storage effectively preserved these analytes, underscoring their critical role [8].

Table 1: Agreement of urine analytes stored into a BD Vacutainer® UAP preservative tube after 24 and 48 hours compared to initial results.

		Time of analysis (hrs)	K (95% CI)
Chemical parameters	pH (pH units)	24	1.00 (1.00-1.00)
		48	0.98 (0.96-1.00)
	Haemoglobin (mg/dL)	24	0.98 (0.96-1.00)
		48	0.92 (0.84-1.00)
	Leukocyte esterase (cells/µl)	24	0.98 (0.96-1.00)
		48	0.94 (0.88-1.00)
	Nitrite (mg/dL)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)
	Glucose (mg/dL)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)
	Protein (mg/dL)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)
	Albumin (mg/L)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)
	Creatinine (mg/dL)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)
	Ketone (mg/dL)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)
	Bilirubin (mg/dL)	24	1.00 (1.00-1.00)
		48	1.00 (1.00-1.00)

Similarly, Karakoyun and co-authors highlighted the effectiveness of chlorhexidine-based preservative tubes in maintaining the stability of routine urinalysis parameters for 24 hours at room temperature [7]. Our findings extend these observations by demonstrating that BD Vacutainer® UAP tubes can preserve urine sample integrity up to 48 hours at room temperature. Importantly, we incorporated a broader panel of parameters, including albumin, creatinine, and clinically significant indices such as the protein-to-creatinine ratio (PCR) and albumin-to-creatinine ratio (ACR), all of which remained stable throughout the 48-hour period. Given that these markers are key tests for detecting early renal damage and proteinuria, their preservation reinforces the clinical utility of the BD Vacutainer® UAP tubes.

The observed small increase in pH over the 48-hour storage period agreed with findings reported by Ekşioğlu and co-authors and likely reflects slow biochemical transformations such as urea hydrolysis or ammonium accumulation, which may occur despite the presence of preservatives [6]. Nonetheless, this pH shift remained within normal physiological limits. The stability of specific gravity measurements further supports the effectiveness of the preservative in preventing evaporative loss or solute precipitation, contributing to sample integrity.

A minor decrease in haemoglobin and leukocyte esterase was noted in a small subset of samples, reflecting the lability of enzymatic markers and haemoglobin's susceptibility to degradation. Although these changes were limited, they underscore the importance of timely analysis of certain labile parameters to ensure diagnostic accuracy.

This study presents some limitations that should be not neglected. First, it is a single-centre analysis, and a larger cohort is needed to confirm our findings. Second, a review of the scientific literature reveals substantial heterogeneity among studies in terms of sample sizes, time points, instruments, and software used, which may explain discrepancies between our conclusions and those of other works.

Collectively, our data support the use of BD Vacutainer® UAP preservative tubes as a practical and reliable solution for urine sample preservation, especially in clinical settings where immediate refrigeration or analysis is not readily performed. By extending the stability of key urinalysis parameters at room temperature, these tubes enhance workflow flexibility, reduce the risk of sample rejection due to delayed processing, and ultimately improve diagnostic reliability. This is particularly useful in decentralized or resource-limited environments, where maintaining cold chains or rapid sample analysis can be difficult.

Author Contributions

JI and PI wrote the manuscript; JI, PI, and MC designed the study; FB, LF, DM, EZ, and LZ performed the sample analysis; JI, PI, and MC critically revised the manuscript. All authors read and approved the final draft for submission.

Data Availability Statement

The data generated and analyzed are available from the corresponding author on request.

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