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MICROBIOLOGICAL AND PHYSICOCHEMICAL PROPERTIES OF DRINKING WATER SOLD THROUGH WATER VENDING MACHINES INSTALLED IN URBAN AND RURAL SETTLEMENTS

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Abstract

Background. Water vending machines (WVMs) that treat piped water on site and dispense it into consumers' containers have become a widespread source of drinking water in settlements of Uzbekistan, yet their hygienic performance has not been systematically evaluated.

Aim. To assess the microbiological and physicochemical properties of drinking water sold through WVMs in urban and rural settlements of Samarkand region and to identify the technical and sanitary factors that determine its safety.

Methods. Forty WVMs (26 urban and 14 rural) were inspected with a standardized checklist and sampled in spring, summer and autumn 2025 (120 product samples). Feed water (n = 40) and swabs from dispensing nozzles (n = 40) were also examined. Analyses followed ISO and standard methods, and results were compared with O'z DSt 950:2011 and international benchmarks for packaged water.

Results. Reverse osmosis (RO) with ultraviolet (UV) disinfection was used in 77.5% of machines, but UV lamps were non-functional in 27.5% and filter replacement was documented in only 45.0%. RO reduced total dissolved solids from 548 ± 104 to 64 ± 38 mg/L and total hardness from 5.67 ± 1.05 to 0.62 ± 0.41 mmol/L; 83.9% of RO samples contained less than 100 mg/L of dissolved solids and 22.6% had pH below 6.5. Microbiological non-compliance was found in 36.7% of product samples (heterotrophic plate count at 22 °C > 100 CFU/mL in 27.5%, total coliforms in 19.2%, Pseudomonas aeruginosa in 15.0%, Escherichia coli in 6.7%), whereas only 5.0% of feed water samples contained coliforms. Heterotrophic counts rose with the time elapsed since filter replacement. Non-compliance was associated with a non-functional UV lamp (OR 4.87; 95% CI 2.07–11.44), visible soiling of the nozzle (OR 4.33; 1.96–9.59), undocumented filter replacement (OR 3.91; 1.72–8.87) and an unprotected nozzle (OR 3.03; 1.40–6.56), but not significantly with rural location. **Conclusion.** WVMs improve the chemical and organoleptic quality of water but frequently degrade its microbiological quality and produce physiologically inadequate, demineralized water. Mandatory maintenance schedules, monitoring of UV units, protection of dispensing points and remineralization should be enforced within a dedicated regulatory framework.

Keywords: water vending machines; drinking water; reverse osmosis; ultraviolet disinfection; heterotrophic plate count; Pseudomonas aeruginosa; demineralization; sanitary inspection; Uzbekistan

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1. Introduction

Water vending machines (WVMs), often called aquamats, are automated self-service units that take water from the public supply, subject it to additional treatment and dispense it for a fee into containers brought by consumers. Owing to their low price per litre compared with bottled water and the good organoleptic properties of the product, WVMs have spread rapidly in many countries, and their number has grown noticeably in cities and rural settlements of Uzbekistan during the last decade. Typical units combine sediment and activated carbon filtration, reverse osmosis (RO) membranes and a final ultraviolet (UV) lamp, sometimes supplemented by a storage tank and a mineralizing cartridge (Yeremin et al., 2023).

Although WVMs are marketed as a source of "purified" water, studies from different regions show that their product can be of worse microbiological quality than the feed water. In the United States, Pseudomonas aeruginosa was detected in 23% and coliform bacteria in 20% of samples from 30 WVMs, and 73% of samples exceeded 500 CFU/mL of heterotrophic bacteria (Chaidez et al., 1999). In California, total coliforms were found in 32% of WVMs, and molecular methods detected genetic material of several pathogens (Hile et al., 2021); a later study also found heavy contamination of dispenser swabs (Hile et al., 2023). In Bangkok, 37.2% of vended water samples exceeded 500 heterotrophic colonies per millilitre and 21.6% contained P. aeruginosa (Janwithayanuchit et al., 2017), and similar problems were reported from Malaysia and Thailand (Hashim & Yusop, 2016; Wibuloutai et al., 2019). Earlier surveys attributed these failures largely to inadequate maintenance, inspection and sanitation of machines (McSwane et al., 1994; Schillinger & Du Vall Knorr, 2004).

The mechanisms of deterioration are well understood. Activated carbon removes residual chlorine and accumulates organic matter, creating favourable conditions for bacterial growth; filters, membranes, storage tanks and nozzles develop biofilms that protect bacteria from disinfection (Flemming & Wingender, 2010); and UV disinfection is effective only when the lamp intensity, exposure time and water transmittance are adequate (Hijnen et al., 2006). Although heterotrophic plate counts (HPC) do not in themselves indicate a health risk, they are sensitive indicators of regrowth and loss of process control (Allen et al., 2004; Bartram et al., 2003), and P. aeruginosa is an opportunistic pathogen of concern for vulnerable consumers (Mena & Gerba, 2009; World Health Organization [WHO], 2022).

A second hygienic issue is demineralization. RO removes 90% or more of dissolved salts (Greenlee et al., 2009), and without subsequent mineral correction the product may be deficient in calcium and magnesium, which is undesirable for long-term consumption (Kozisek, 2005, 2020). A recent review concluded that the safety of vended water depends not only on source quality and treatment but also on the location and design of machines, operator practices and consumer containers, and that regulatory approaches differ widely between countries (Yeremin et al., 2023). In Uzbekistan, the quality of piped water is regulated by O'z DSt 950:2011 (Uzstandard, 2011) and that of packaged water by a general technical regulation (Cabinet of Ministers of the Republic of Uzbekistan, 2019), but specific hygienic requirements for WVMs are not established.

The aim of this study was to assess the microbiological and physicochemical properties of drinking water sold through WVMs in urban and rural settlements of Samarkand region and to identify technical and sanitary factors associated with non-compliance. We hypothesized that the product quality is determined primarily by the maintenance status of machines rather than by the quality of the feed water.

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2. Materials and Methods

2.1. Study design and machine selection

A cross-sectional study with three sampling rounds (spring, summer and autumn 2025) was conducted in Samarkand city and in rural settlements and district centres of Samarkand region. A list of operating WVMs was compiled by field survey, and 40 machines were selected by stratified random sampling: 26 in the city (urban stratum) and 14 in rural settlements (rural stratum). All selected machines were supplied from centralized or local piped water systems. Operators were informed about the purpose of the study and agreed to participate; machines were not identified by owner in the analysis.

2.2. Sanitary and technical inspection

At the first visit, each machine was inspected with a standardized checklist covering: location and exposure to direct sunlight; treatment scheme (presence of an RO stage); functioning of the UV lamp (indicator and visual check); availability of a maintenance log and documented replacement of filters and membranes within the manufacturer's interval; design of the dispensing point (closed compartment or self-closing door); visible soiling, scale or biofilm on the nozzle and dispensing niche; and availability of a laboratory quality certificate. The time elapsed since the last filter or membrane replacement was taken from the maintenance log or, when absent, from the operator's report and classified as ≤ 1, 1–3, 3–6 and > 6 months or unknown.

2.3. Sampling

In each round, a product sample was collected from every machine after dispensing and discarding the first 2 L, which reflects the water received by a typical consumer after a short idle period (n = 120). Feed water was sampled from the inlet tap of each machine in the first round (n = 40), and the internal surface of each dispensing nozzle was swabbed over a standardized area (n = 40). Sampling complied with ISO 5667-5 (International Organization for Standardization [ISO], 2006b). Water for microbiological analysis was collected in sterile bottles containing sodium thiosulfate and transported at 2–8 °C; analyses began within 6 h. Water temperature was measured on site.

2.4. Laboratory analyses

Physicochemical indicators (pH, turbidity, total dissolved solids (TDS), total hardness, calcium, magnesium, nitrates, chlorides, sulfates, fluoride, total iron, permanganate oxidizability and free residual chlorine) were determined by potentiometric, gravimetric, titrimetric, photometric and ion-selective methods in accordance with the Standard Methods (Baird et al., 2017). Heterotrophic plate counts at 36 ± 2 °C and 22 ± 2 °C were determined by pour plating on yeast extract agar (ISO, 1999). Total coliforms and Escherichia coli (ISO, 2014), intestinal enterococci (ISO, 2000) and P. aeruginosa (ISO, 2006a) were enumerated by membrane filtration of 250 mL. Swabs were cultured for HPC and total coliforms. Duplicate analyses, blanks and positive controls were included in each batch.

2.5. Assessment criteria and statistical analysis

Because vended water is sold as a ready-to-drink product, it was assessed against the chemical requirements of O'z DSt 950:2011 (Uzstandard, 2011) and the microbiological benchmarks for water offered for sale in bottles or containers: HPC ≤ 20 CFU/mL at 37 °C and ≤ 100 CFU/mL at 22 °C and absence of coliforms, E. coli, enterococci and P. aeruginosa in 250 mL (Codex Alimentarius Commission, 2001; Council of the European Union, 1998). A sample was considered non-compliant if it failed at least one criterion. Physiological adequacy was assessed by the thresholds of TDS ≥ 100 mg/L and total hardness ≥ 1.5 mmol/L (Kozisek, 2005).

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Continuous variables are presented as mean ± SD or median (IQR) and were compared with the Kruskal–Wallis test with Dunn's post hoc test; HPC values were $\log_{10}$-transformed. Removal efficiency was calculated as $(1 - C_{out}/C_{in}) \times 100\%$. Associations between inspection findings and non-compliance were expressed as crude odds ratios (OR) with 95% confidence intervals (CI) and tested with the χ^2 test; correlations were assessed with Spearman's coefficient (ρ). The level of significance was $p < 0.05$. Analyses were performed in IBM SPSS Statistics 26.0.

3. Results and Discussion

3.1. Technical and sanitary status of the machines

The results of the inspection are presented in Table 1. Most machines (77.5%) included an RO stage followed by UV disinfection, whereas the remaining 22.5% relied on sediment and carbon filtration with UV. However, the UV lamp was non-functional at the time of inspection in 11 machines (27.5%), filter replacement was documented within the recommended interval in only 18 machines (45.0%) and a maintenance log was available in 21 (52.5%). The dispensing nozzle was protected by a closed compartment or self-closing door in 60.0% of machines, and visible soiling, scale or mucous deposits on the nozzle or in the dispensing niche were observed in 37.5%. Only 32.5% of operators could present a laboratory certificate of product quality. All indicators of maintenance were less favourable in rural settlements, where machines were more often installed outdoors near markets and roads.

Table 1. Technical and sanitary characteristics of the surveyed water vending machines, n (%)

Characteristic	Urban (n = 26)	Rural (n = 14)	Total (n = 40)
Reverse osmosis + UV treatment	22 (84.6)	9 (64.3)	31 (77.5)
Filtration + UV without reverse osmosis	4 (15.4)	5 (35.7)	9 (22.5)
UV lamp functional at inspection	21 (80.8)	8 (57.1)	29 (72.5)
Filter replacement documented within interval	14 (53.8)	4 (28.6)	18 (45.0)
Maintenance log available	16 (61.5)	5 (35.7)	21 (52.5)
Nozzle protected (closed niche/self-closing door)	18 (69.2)	6 (42.9)	24 (60.0)
Visible soiling of nozzle or dispensing niche	8 (30.8)	7 (50.0)	15 (37.5)
Exposure to direct sunlight	10 (38.5)	7 (50.0)	17 (42.5)
Laboratory quality certificate available	11 (42.3)	2 (14.3)	13 (32.5)

Note: UV – ultraviolet disinfection unit.

3.2. Physicochemical properties

Feed water corresponded to moderately mineralized tap water that met the national standard, although free residual chlorine was low (0.21 ± 0.11 mg/L) (Table 2). Treatment in RO machines profoundly changed its composition: TDS decreased from 548 ± 104 to 64 ± 38 mg/L, total hardness from 5.67 ± 1.05 to 0.62 ± 0.41 mmol/L, and calcium and magnesium to 8.6 and 2.3 mg/L, respectively.

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In machines without RO, the ionic composition remained close to that of the feed water, and their effect was limited mainly to turbidity, iron and organic matter. Water temperature increased by about 5 °C during residence in the machines, reaching 27–31 °C in summer.

Table 2. Physicochemical properties of feed water and water dispensed by vending machines (mean ± SD)

Indicator	Standard	Feed water (n = 40)	RO + UV product (n = 93)	Non-RO product (n = 27)	p
pH	6.0–9.0	7.46 ± 0.22	6.62 ± 0.41	7.38 ± 0.25	< 0.001
Turbidity, mg/L	≤ 1.5	0.58 ± 0.34	0.14 ± 0.08	0.31 ± 0.17	< 0.001
TDS, mg/L	≤ 1000	548 ± 104	64 ± 38	486 ± 97	< 0.001
Total hardness, mmol/L	≤ 7.0	5.67 ± 1.05	0.62 ± 0.41	4.94 ± 0.98	< 0.001
Calcium, mg/L	–	78.4 ± 14.6	8.6 ± 5.9	68.9 ± 13.2	< 0.001
Magnesium, mg/L	–	21.4 ± 5.2	2.3 ± 1.6	18.3 ± 4.7	< 0.001
Nitrates (NO_3^-), mg/L	≤ 45	15.2 ± 7.1	1.9 ± 1.2	13.8 ± 6.4	< 0.001
Chlorides, mg/L	≤ 350	48.6 ± 12.3	6.4 ± 3.8	45.1 ± 11.7	< 0.001
Sulfates, mg/L	≤ 500	151.3 ± 33.8	9.8 ± 6.1	139.6 ± 31.2	< 0.001
Fluoride, mg/L	0.7–1.5	0.39 ± 0.10	0.05 ± 0.03	0.35 ± 0.09	< 0.001
Total iron, mg/L	≤ 0.3	0.12 ± 0.09	0.02 ± 0.01	0.06 ± 0.04	< 0.001
Permanganate oxidizability, mg O_2/L	≤ 5.0	1.92 ± 0.66	0.48 ± 0.21	1.21 ± 0.45	< 0.001
Free residual chlorine, mg/L	0.3–0.5	0.21 ± 0.11	< 0.05	< 0.05	–
Water temperature, °C	–	17.8 ± 4.6	22.9 ± 4.8	23.4 ± 5.1	< 0.001

Note: standards according to O'z DSt 950:2011; p – Kruskal–Wallis test. Post hoc: the non-RO product did not differ from feed water in TDS, hardness, Ca, Mg, nitrates, chlorides, sulfates and

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fluoride ($p > 0.05$) but differed in turbidity, iron, oxidizability and residual chlorine ($p < 0.01$). RO – reverse osmosis; TDS – total dissolved solids.

The removal efficiencies of the two treatment schemes are compared in Figure 1. RO machines removed 83–94% of all major ions, nitrates and fluoride, which agrees with the typical salt rejection of RO membranes (Greenlee et al., 2009), whereas non-RO machines removed only 7–15% of dissolved ions but 37–50% of iron and organic matter, owing to sediment and activated carbon filtration. None of the machines was equipped with a functioning remineralization cartridge.

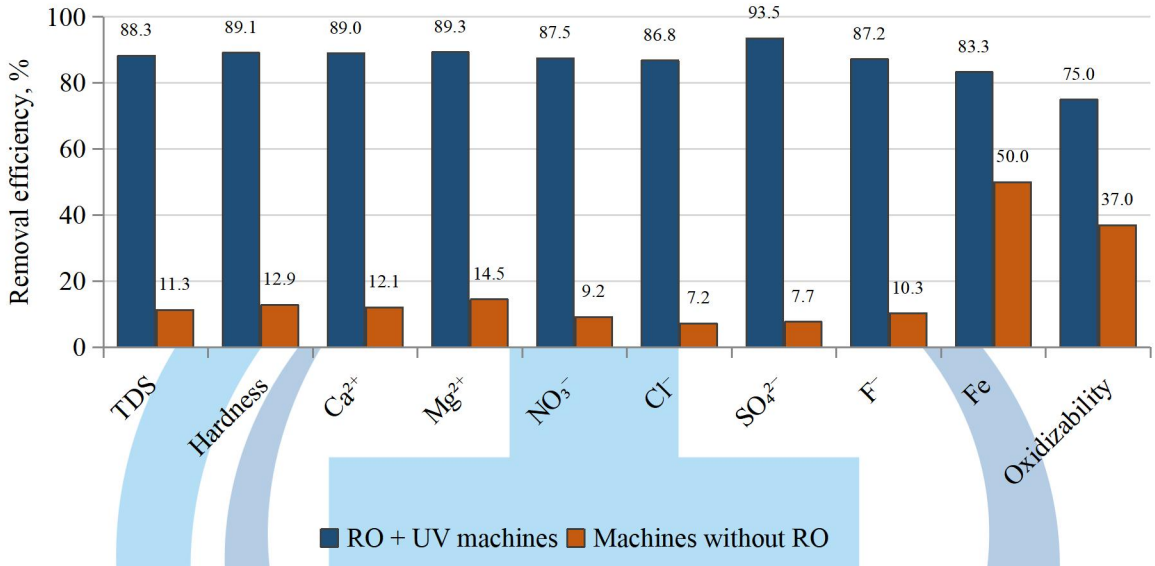


Figure 1. Removal efficiency of physicochemical constituents in vending machines with and without reverse osmosis, %

From the hygienic point of view, RO-treated vended water is chemically safe but physiologically inadequate. TDS was below 100 mg/L in 78 of 93 RO samples (83.9%), total hardness below 1.5 mmol/L in 89 samples (95.7%), and pH was below 6.5 in 21 samples (22.6%), including four samples (4.3%) below the national limit of 6.0. Fluoride fell to 0.05 ± 0.03 mg/L. Long-term consumption of such water as the main source of drinking water, especially for cooking and infant formula, may reduce the intake of calcium and magnesium and increase the aggressiveness of water towards cookware and containers (Kozisek, 2005). Because few regulations prescribe minimum mineral levels (Kozisek, 2020), the issue is often overlooked; the review by Yeremin et al. (2023) similarly emphasized the need to control TDS, calcium and magnesium in vended water produced by RO.

3.3. Microbiological quality

Only two feed water samples (5.0%) contained coliform bacteria, and none contained E. coli; the median HPC at 22 °C was 12 (IQR 5–31) CFU/mL. In contrast, 44 of 120 product samples (36.7%) failed at least one microbiological criterion. HPC at 22 °C exceeded 100 CFU/mL in 27.5% and HPC at 37 °C exceeded 20 CFU/mL in 22.5% of samples, the median HPC at 22 °C increasing to 38 (IQR 9–186) CFU/mL. Total coliforms were detected in 19.2%, P. aeruginosa in 15.0%, E. coli in 6.7% and enterococci in 5.0% of product samples. Swabs from 14 nozzles (35.0%) were positive for coliforms. The deterioration of water between the inlet and the outlet demonstrates that contamination originated predominantly inside the machines and at the dispensing point rather than in the public supply, which

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supports our hypothesis and agrees with observations in other countries (Chaidez et al., 1999; Hile et al., 2021).

The frequency of contamination by type of settlement is shown in Figure 2. All indicators were detected more often in rural machines: total coliforms in 28.6% versus 14.1% and E. coli in 11.9% versus 3.8% of samples, and overall non-compliance reached 47.6% versus 30.8%. However, none of these differences reached statistical significance (for overall non-compliance, $\chi^2 = 3.33$, $p = 0.068$), which indicates that location itself is less important than the maintenance characteristics that differ between urban and rural machines. Non-compliance tended to be highest in summer (47.5%) compared with autumn (37.5%) and spring (25.0%), in line with the higher temperature of the product, although the difference between rounds was not significant ($\chi^2 = 4.38$, $p = 0.112$).

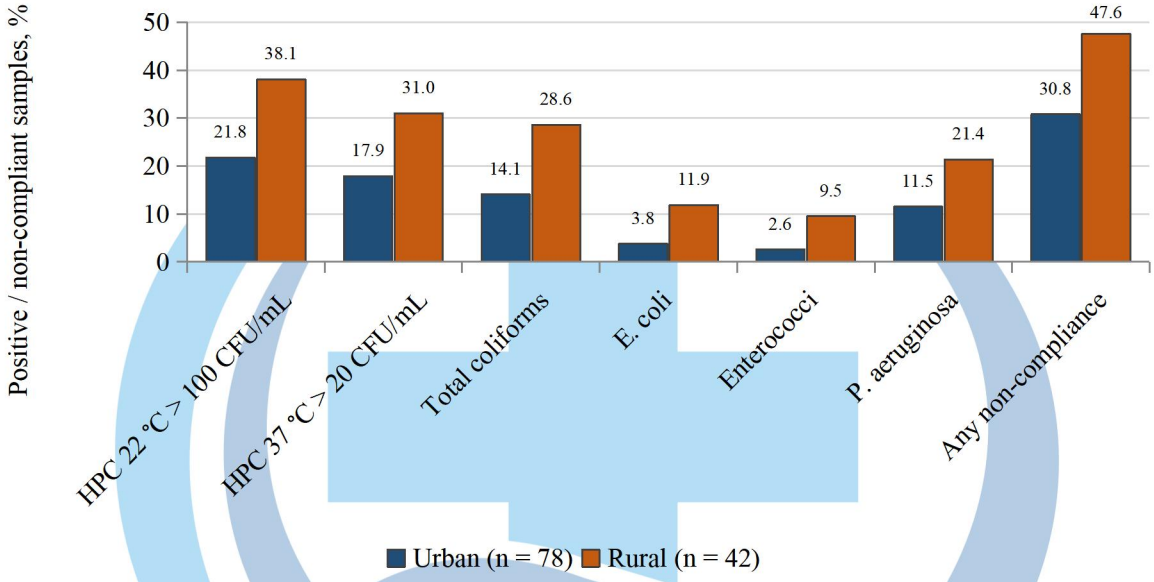


Figure 2. Frequency of microbiological non-compliance of vended water in urban and rural settlements, % of samples

The level of P. aeruginosa contamination (15.0%) was lower than in the North American and Thai studies (Chaidez et al., 1999; Janwithayanuchit et al., 2017) but is nevertheless of concern. This organism readily colonizes filters, tubing and nozzles and, although it poses little risk to healthy people through ingestion, it may cause infections in immunocompromised persons, infants and patients with wounds (Mena & Gerba, 2009). The detection of E. coli and enterococci in 6.7% and 5.0% of samples indicates faecal contamination, most likely introduced at the dispensing point through contact with dirty hands and containers, since these organisms were absent from the feed water (Hunter, 2003; Yeremin et al., 2023).

3.4. Factors associated with non-compliance

Heterotrophic counts increased steadily with the time elapsed since the last replacement of filters and membranes (Figure 3). The mean $\log_{10}$ HPC at 22 °C rose from 1.12 in machines serviced within the previous month to 2.87 in machines serviced more than six months earlier or with unknown service history (Spearman's $\rho = 0.54$, $p < 0.001$). This pattern reflects the progressive colonization of carbon filters and membrane housings by biofilm and the release of cells into the product (Flemming & Wingender, 2010; Liu et al., 2016).

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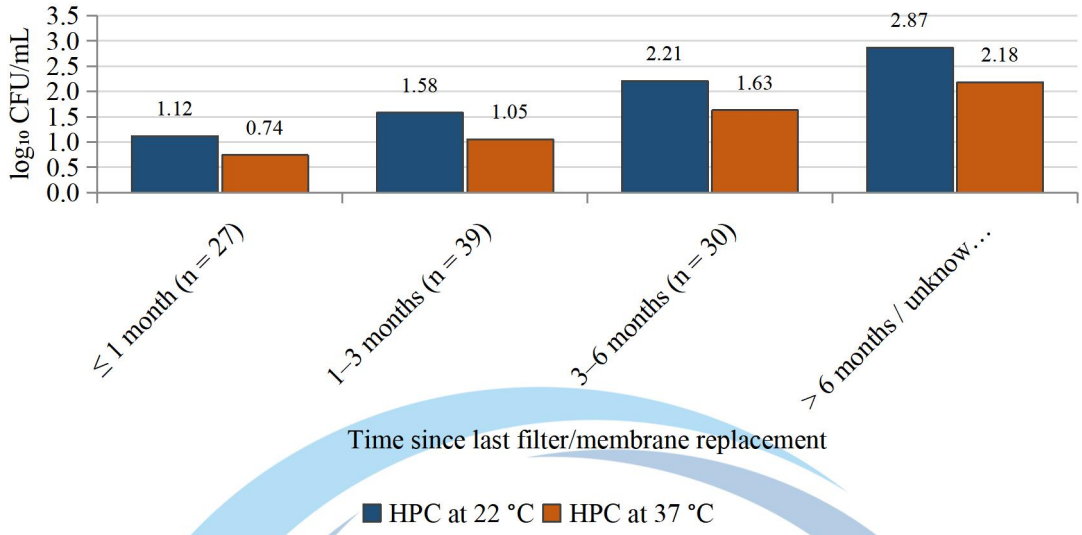


Figure 3. Heterotrophic plate counts in vended water depending on the time since the last filter replacement, $\log_{10}$ CFU/mL (mean)

The associations between inspection findings and non-compliance are summarized in Table 3. The strongest predictors were a non-functional UV lamp (OR 4.87), visible soiling of the nozzle or dispensing niche (OR 4.33) and undocumented filter replacement (OR 3.91); an unprotected nozzle tripled the odds of non-compliance. Exposure to direct sunlight, rural location and the absence of an RO stage showed positive but non-significant associations. These results are consistent with the view that the quality of maintenance, disinfection of machines and control of the product are decisive for the safety of vended water (Hile et al., 2021; Schillinger & Du Vall Knorr, 2004; Yeremin et al., 2023). UV units provide a reliable barrier against vegetative bacteria only when lamps are replaced and cleaned regularly (Hijnen et al., 2006), and their failure is not apparent to consumers because it does not change the appearance or taste of the water.

Table 3. Association of technical and sanitary characteristics of vending machines with microbiological non-compliance of the product

Factor	Factor present, n/N (%)	Factor absent, n/N (%)	OR (95% CI)	p
UV lamp non-functional	21/33 (63.6)	23/87 (26.4)	4.87 (2.07–11.44)	< 0.001
Visible soiling of nozzle/niche	26/45 (57.8)	18/75 (24.0)	4.33 (1.96–9.59)	< 0.001
Filter replacement not documented	33/66 (50.0)	11/54 (20.4)	3.91 (1.72–8.87)	0.001
Nozzle not protected	25/48 (52.1)	19/72 (26.4)	3.03 (1.40–6.56)	0.004

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Factor	Factor present, n/N (%)	Factor absent, n/N (%)	OR (95% CI)	p
Rural location	20/42 (47.6)	24/78 (30.8)	2.05 (0.94–4.43)	0.068
Direct sunlight exposure	23/51 (45.1)	21/69 (30.4)	1.88 (0.88–3.99)	0.099
No reverse osmosis stage	12/27 (44.4)	32/93 (34.4)	1.52 (0.64–3.64)	0.341

Note: n/N – non-compliant samples/all samples from machines with or without the factor (three samples per machine); OR – crude odds ratio; CI – confidence interval; p – χ^2 test.

The practical implications are straightforward. Hygienic requirements for WVMs should include mandatory replacement of filters and membranes at fixed intervals with documentation, automatic monitoring of UV lamp operation with blocking of dispensing in case of failure, a closed and regularly disinfected dispensing compartment, protection from direct sunlight, remineralization of RO-treated water to at least 100 mg/L of TDS with adequate calcium and magnesium, and information for consumers about the sanitation of their containers. Similar requirements already exist in several jurisdictions where vended water is regulated either as a food product or as a public water system (Yeremin et al., 2023). Periodic laboratory control by operators, with results displayed on the machine, would further improve transparency.

4. Conclusion

Drinking water sold through vending machines in urban and rural settlements of Samarkand region is chemically safe and organoleptically improved compared with the feed water, but its hygienic quality is compromised in two respects. First, 36.7% of product samples failed microbiological criteria, with frequent detection of heterotrophic regrowth, coliforms and P. aeruginosa and occasional faecal indicators, although the feed water was of good microbiological quality; contamination therefore arose within the machines and at the dispensing point. Second, RO-treated water was profoundly demineralized, with TDS below 100 mg/L in 83.9% of samples and hardness, calcium, magnesium and fluoride far below physiologically desirable levels. Non-functional UV lamps, soiled and unprotected nozzles and irregular filter replacement were the main determinants of non-compliance, while rural location had no independent significant effect. The introduction of specific sanitary rules for WVMs, covering maintenance intervals, automatic UV monitoring, protection and disinfection of dispensing points, remineralization and regular laboratory control, is recommended.

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