Pharmaceutical and Clinical Journal of Nusantara (PCJN)
Volume 03, No. 03, November 2025, Page 156-163
E-ISSN 2985 – 4040 (Online Media)
https://nusantarascientificjournal.com/index.php/pcjn/index
https://doi.org/10.58549/pcjn.v3i03.145

Determination and Validation of Nitrite and Nitrate Levels in Drinking Water from Various Sources in Medan City Using Visible Spectrophotometry

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Abstract

The maximum allowable concentrations of nitrite and nitrate in drinking water are regulated by the Regulation of the Ministry of Health of the Republic of Indonesia No. 492/Menkes/Per/IV/2010. Variations in environmental conditions and agricultural activities may influence the levels of these contaminants in different water sources. This study aimed to determine nitrite and nitrate concentrations in drinking water obtained from several sources in Medan City. Eight samples were collected, consisting of municipally treated water from two drinking water companies, two well water samples, two commercially bottled water brands, and boiled well water. Quantitative analysis was performed using visible spectrophotometry at a wavelength of 536 nm with a Shimadzu UV mini-1240 spectrophotometer. Nitrite concentrations ranged from 0.0273 to 0.6452 mg/L, while nitrate concentrations ranged from 0.0148 to 3.9431 mg/L. All measured levels were below the maximum permissible limits. The validated analytical method demonstrated acceptable accuracy and precision, indicating its suitability for routine monitoring of drinking water quality. Differences in water source and management practices were found to influence nitrite and nitrate concentrations.

Keywords: drinking water, well water, nitrite, nitrate, visible spectrophotometry

Abstrak

Batas maksimum kadar nitrit dan nitrat dalam air minum diatur dalam Peraturan Menteri Kesehatan Republik Indonesia No. 492/Menkes/Per/IV/2010. Variasi kondisi lingkungan dan aktivitas pertanian dapat memengaruhi kadar kedua senyawa tersebut pada berbagai sumber air. Penelitian ini bertujuan untuk menentukan kadar nitrit dan nitrat dalam air minum yang berasal dari beberapa sumber di Kota Medan. Sebanyak delapan sampel dianalisis, meliputi dua air dari perusahaan pengelola air minum, dua air sumur, dua air minum dalam kemasan, dan air sumur yang telah dimasak. Analisis kuantitatif dilakukan menggunakan metode spektrofotometri sinar tampak pada panjang gelombang 536 nm dengan alat Shimadzu UV mini-1240. Kadar nitrit berkisar antara 0,0273–0,6452 mg/L dan kadar nitrat berkisar antara 0,0148–3,9431 mg/L. Seluruh kadar yang diperoleh masih berada di bawah batas maksimum yang diizinkan. Metode yang divalidasi menunjukkan akurasi dan presisi yang baik sehingga layak digunakan untuk pemantauan kualitas air minum secara rutin. Perbedaan sumber dan pengelolaan air memengaruhi kadar nitrit dan nitrat yang terdeteksi.

Kata kunci: *air minum, air sumur, nitrit, nitrat, spektrofotometri sinar tampak*

Received: 03 August 2025 *Revised: 02 October 2025* *Accepted: 08 November 2025* *Publish: 30 November 2025*

INTRODUCTION

Safe drinking water is a fundamental human necessity and a critical determinant of public health. Globally, drinking water quality has become an increasing concern due to contamination by inorganic pollutants, particularly nitrite and nitrate ions, which primarily originate from anthropogenic activities such as intensive agriculture, excessive nitrogen-based fertilizer application, domestic wastewater discharge, and industrial development[1]. Prolonged exposure to elevated concentrations of these nitrogen compounds has been associated with significant health risks, including methemoglobinemia in infants, impaired oxygen transport in the bloodstream, and the potential endogenous formation of carcinogenic N-nitroso compounds[2]. Therefore, continuous monitoring of nitrite and nitrate concentrations in drinking water remains an essential component of environmental and public health surveillance worldwide[3].

To mitigate these risks, many countries have established regulatory standards governing permissible concentrations of nitrite and nitrate in drinking water. In Indonesia, the maximum allowable limits are stipulated under the Regulation of the Ministry of Health of the Republic of Indonesia No. 492/Menkes/Per/IV/2010[4]. Nevertheless, compliance with these standards may vary depending on local environmental conditions, land-use patterns, sanitation systems, and water management practices. Rapid urbanization and population growth, particularly in metropolitan areas, have been reported to increase the vulnerability of water resources to nitrogen-based contamination[5].

Medan, one of the major metropolitan cities in Indonesia, presents complex characteristics in terms of water supply systems and management practices. The community relies on multiple sources of drinking water, including municipally treated piped water, groundwater obtained from bore and dug wells, refill bottled water, and water treated domestically through boiling[6]. Each of these sources is subjected to distinct environmental influences and handling processes, potentially resulting in variations in chemical composition. Despite the widespread consumption of these water sources, systematic comparative data regarding nitrite and nitrate concentrations across different drinking water sources in Medan remain limited[7]. This lack of localized baseline data highlights the need for comprehensive analytical evaluation.

From an analytical standpoint, visible spectrophotometry represents a practical and reliable technique for the quantitative determination of nitrite and nitrate in aqueous samples. The method offers adequate sensitivity, simplicity, reproducibility, and suitability for routine laboratory application. Through appropriate reagent reactions and wavelength optimization, visible spectrophotometry enables accurate assessment of nitrogen compound concentrations in relation to regulatory standards[8].

Based on these considerations, the determination of nitrite and nitrate levels in drinking water from various sources in Medan City is both scientifically relevant and practically important. This study aims to generate baseline data on nitrogen contamination in drinking water, evaluate compliance with national regulatory limits, and provide evidence to support water quality management and public health protection strategies. The findings are expected to contribute to ongoing monitoring efforts and serve as a reference for future environmental and pharmaceutical analytical research.

METHODOLOGY

Instruments

The instruments used in this study included a UV–Visible spectrophotometer (Shimadzu UV mini-1240), an analytical balance with an accuracy of ±0.1 mg, 1 cm quartz cuvettes, a thermostatically controlled water bath, filtration apparatus with filter paper, rubber pipette bulbs, and standard laboratory glassware required for the analysis.

Materials

All reagents used were of pro analysis (p.a.) grade supplied by E-Merck, including sodium nitrite ($NaNO_2$), sulfanilic acid, *N-(1-naphthyl)ethylenediamine dihydrochloride*, glacial acetic acid, and zinc powder (Zn) for nitrate reduction. Distilled water was used for reagent preparation and dilution.

Sample Collection

Sample collection was carried out using a purposive sampling method to ensure that the samples represented commonly consumed drinking water sources.

A total of seven types of water samples were collected from several areas in the city of Medan. The sampling locations were categorized as follows:

A1 = PDAM S****l water
A2 = PDAM D*** T** water
A3 = Bore well water
A4 = Dug well water
A5 = Commercially packaged drinking water (A**s brand)
A6 = Commercially packaged drinking water (P***a brand)
A7 = Dug well water after boiling treatment

Samples were collected in clean polyethylene bottles and analyzed as soon as possible after collection.

Determination of Nitrite Levels in Water Samples

A 25 mL aliquot of each sample was filtered, and the first ±10 mL of filtrate was discarded. Subsequently, 10 mL of the filtrate was transferred into a 50 mL volumetric flask. Then, 2.5 mL of sulfanilic acid reagent was added, shaken, and allowed to stand for 5 minutes. Afterward, 2.5 mL of *N-(1-naphthyl)ethylenediamine dihydrochloride* reagent was added.

The solution was diluted to volume with distilled water, homogenized, and allowed to stand. Absorbance was measured at the 12th minute at a wavelength of 536 nm.
Nitrite concentration was calculated using the regression equation:
$Y = aX + b$
Where:
Y = absorbance
X = concentration obtained from the calibration curve (μg/mL)

The concentration of nitrite in the sample was calculated using the following equation:
C_sample = X × F_p
Where:
C_sample = nitrite concentration in the original sample (μg/mL)
X = concentration obtained from calibration curve (μg/mL)
F_p = dilution factor

Determination of Nitrate Levels in Water Samples

A 25 mL sample was filtered and the first ±10 mL of filtrate was discarded. Then, 10 mL of the filtrate was transferred into a 50 mL volumetric flask. A small amount of zinc powder (Zn) was added to reduce nitrate to nitrite and the mixture was allowed to stand for 10 minutes.

After the reduction process, 2.5 mL of sulfanilic acid reagent was added, shaken, and allowed to stand for 5 minutes, followed by the addition of 2.5 mL of *N-(1-naphthyl)ethylenediamine dihydrochloride* reagent.

The solution was diluted to volume with distilled water, homogenized, and the absorbance was measured at the 12th minute at 536 nm.

The nitrate concentration in the sample was calculated using the regression equation:
$Y = aX + b$[9]

Method Validation of Analytical Procedures

The analytical method for the determination of nitrite and nitrate levels in water samples was validated to ensure reliability, accuracy, and precision in accordance with established analytical guidelines[9]. Validation parameters included selectivity, linearity, accuracy (recovery), precision, limit of detection (LOD), and limit of quantification (LOQ).

Selectivity and Identification

Selectivity was evaluated through qualitative identification of nitrite using the Griess reaction. A portion of each sample was reacted with sulfanilic acid and *N-(1-naphthyl)ethylenediamine dihydrochloride* reagents. The formation of a reddish-purple coloration indicated the presence of nitrite, confirming the specificity of the method toward nitrite ions without significant interference from other components in the sample matrix[9].

Linearity

Linearity was assessed using a series of standard nitrite solutions prepared at different concentrations. The absorbance of each standard solution was measured at 536 nm using visible spectrophotometry. A calibration curve was constructed by plotting absorbance against concentration, and linearity was evaluated based on the regression relationship obtained from the calibration data[9].

Accuracy (Recovery Test)

Accuracy was evaluated using the standard addition method[9]. Known concentrations of nitrite and nitrate standard solutions were added to selected water samples, which were then analyzed using the same procedure as the original samples.
Percentage recovery was calculated using the following equation:
%Recovery = (Measured concentration / Added concentration) × 100

Precision

Precision was evaluated by repeated analysis of homogeneous samples under the same experimental conditions. Precision was expressed as Relative Standard Deviation (RSD) using the following equation:
RSD (%) = (Standard Deviation / Mean) × 100

A low RSD value indicated good repeatability of the analytical method[9].

Limit of Detection and Limit of Quantification

The sensitivity of the method was evaluated by determining the limit of detection (LOD) and limit of quantification (LOQ). These parameters were calculated using the following equations:
LOD = 3.3 × (SD / Slope)
LOQ = 10 × (SD / Slope)
Where:
SD = standard deviation of the response
Slope = slope of the calibration curve

The calculations were performed in accordance with established analytical validation guidelines[9].

Statistical Evaluation

Statistical analysis was conducted to assess the reliability of the analytical results. Data were expressed as mean ± standard deviation. Confidence intervals were calculated to estimate the true concentration of nitrite and nitrate at a defined confidence level.

RESULT AND DISCUSSION

Identification of Nitrite

Qualitative identification of nitrite in water samples was performed using the Griess reaction. The results of nitrite identification in water samples are presented in Table 1.

Table 1. Identification of nitrite in water samples from the City of Medan

No	Sample	Nitrite (Sulfanilic Acid Reagent and NED Solution)
1.	PDAM Su***l Water	Reddish purple
2.	PDAM De****a Water	Faint reddish purple
3.	Bottled Water Sample (P***a brand)	Faint reddish purple
4.	Bottled Water Sample (A**s brand)	Faint reddish purple
5.	Dug Well Water	Intense reddish purple
6.	Bore Well Water	Reddish purple
7.	Boiled Water Sample	Intense reddish purple

As shown in Table 1, all samples produced a reddish to intense reddish-purple coloration, indicating the presence of nitrite. The variation in color intensity reflects differences in nitrite concentration, where a more intense coloration corresponds to higher nitrite levels in the sample.

Nitrite Absorption Curve

Determination of the maximum absorption wavelength was carried out within the range of 400–800 nm. Nitrite measurements were performed at a concentration of 0.8 µg/mL[10]. The maximum absorbance was observed at 536 nm, which was subsequently used for quantitative analysis.

Linearity of the Calibration Curve

Linearity assessment demonstrated a linear relationship between absorbance and concentration within the studied range. The regression equation obtained for nitrite was:

$Y = 2.341143 - 0.006143$

This equation was used to calculate nitrite and nitrate concentrations in the analyzed samples.

Limit of Detection and Limit of Quantification

The limit of detection (LOD) and limit of quantification (LOQ) were calculated based on the regression data from the calibration curve. The LOD for nitrite and nitrate was 0.000056 µg/mL, while the LOQ was 0.000185 µg/mL[10]. These values indicate that the analytical method possesses adequate sensitivity for trace-level determination.

Nitrite and Nitrate Levels in Water Samples in Medan City

Quantitative determination of nitrite and nitrate concentrations was performed at 536 nm. The analytical results are summarized in Table 2.

Table 2. Nitrite and Nitrate Levels in Water Samples in Medan City

No	Sample	Nitrite Level (µg/mL)	Nitrite Level (µg/mL)
1.	PDAM S****l Water	0.0273 ± 0.0108	0.0591 ± 0.0003
2.	PDAM D*** T** Water	0.0314 ± 0.0006	0.0436 ± 0.0006
3.	Bottled Water Sample (P***a brand)	0.0313 ± 0.0018	0.0736 ± 0.0006
4.	Bottled Water Sample (A**s brand)	0.0409 ± 0.0006	0.0818 ± 0.0003
5.	Dug Well Water	0.0236 ± 0.0005	0.0148 ± 0.0015
6.	Bore Well Water	0.0170 ± 0.0010	0.0360 ± 0.0016
7.	Boiled Water Sample	0.6452 ± 0.0005	3.9431 ± 0.0117

Based on Table 2, bottled water of the P***a® **brand showed the lowest nitrite concentration, while bottled water of the** A***s® brand exhibited the lowest nitrate concentration. In contrast, boiled water originating from dug well water demonstrated the highest concentrations of both nitrite and nitrate. This difference may be attributed to variations in water treatment and management practices. Water supplied by public drinking water companies is routinely evaluated for chemical constituents, including nitrite and nitrate, to ensure compliance with quality standards. Conversely, water obtained from traditional well sources is rarely subjected to routine chemical monitoring. Surface water (non-permanent wells) and groundwater (permanent wells) are strongly influenced by environmental conditions and surrounding agricultural activities[11].

Nitrite concentrations were consistently lower than nitrate concentrations across all samples. This observation is consistent with the chemical behavior of nitrogen species in aquatic environments, where nitrite is thermodynamically less stable and readily oxidized to nitrate. Consequently, nitrate is generally present at higher concentrations than nitrite in natural waters. Elevated nitrate concentrations may originate from fertilizer application, decomposition of organic matter, dissolved fecal contamination, waste disposal, and industrial effluents[12].

The results also indicate that heated samples exhibited increased levels of both nitrite and nitrate. Oxygen availability influences nitrification processes mediated by nitrifying bacteria. During heating, oxidation processes may be enhanced, promoting conversion of ammonia to nitrite and further oxidation of nitrite to nitrate. As a result, nitrate concentrations in heated samples increased compared to unheated samples. Meanwhile, nitrite levels may also increase due to intermediate oxidation reactions during nitrogen transformation pathways.

Previous studies reported that well water located in rice field and coastal areas generally showed relatively low concentrations of nitrite and nitrate and did not exceed permissible limits established by the Indonesian Ministry of Health. In contrast, well water from plantation areas exhibited relatively high nitrate levels, with most samples exceeding allowable limits. Well water located in horticultural agricultural areas was found to contain nitrate concentrations ranging from 60 to 103 mg/L[13].

Nitrite and nitrate contamination in groundwater is influenced by multiple environmental factors, including proximity to animal or human waste disposal sites, soil characteristics, fertilizer usage, and hydrological conditions. The World Health Organization (WHO) has reported a significant increase in nitrate levels in surface and groundwater over the past decade due to intensified fertilizer application, land-use changes, and continuous discharge of animal waste, thereby increasing the risk of human exposure[14].

Nitrate originating from agricultural activities can infiltrate groundwater systems. The extent of nitrate migration depends on several factors, including ion concentration, carbon-to-nitrogen ratio, soil type, groundwater depth, plant species, seasonal variation, and climatic conditions[15].

Validation Test

Validation parameters evaluated in this study included accuracy (trueness), precision (repeatability), limit of detection, and limit of quantification.

Accuracy Test

Accuracy was expressed as percentage recovery (% recovery) using the standard addition method. The accuracy test was performed using S****l water samples. The standard solution was added to the sample, and absorbance was measured at 535 nm. The results are presented in Table 3 and Table 4.

Table 3. Recovery of nitrite and nitrate using the standard addition method in water from the S**l drinking water company**

Before Standard Addition (µg/mL)	Standard Added (µg/mL)	After Standard Addition (µg/mL)	Recovery (%)
0.0285	1	0.9805	95.20
0.0275	1	1.0195	99.20
0.0275	1	1.0680	104.05

0.0275	1	1.0455	101.85
0.0270	1	1.0610	103.40
0.0265	1	1.0610	103.45
Mean ($\bar{X}$)			**101.19**

Table 4. Nitrate Concentration in S**l Water (µg/mL)**

Before Standard Addition (µg/mL)	**Standard Added (µg/mL)**	**After Standard Addition (µg/mL)**	**Recovery (%)**
0.0590	1	1.0325	97.35
0.0590	1	1.0355	97.65
0.0595	1	0.9955	93.60
0.0590	1	0.9900	93.10
0.0590	1	1.0425	98.35
0.0595	1	1.0505	99.15
Mean ($\bar{X}$)			**96.53**

Based on Tables 3 and 4, the average percentage recoveries for nitrite and nitrate were 101.19% and 96.53%, respectively. These values fall within the acceptable recovery range of 80–120%, indicating good accuracy of the analytical method.

Precision Test

Precision was evaluated using Relative Standard Deviation (RSD). The RSD values obtained were 3.38% for nitrite and 2.64% for nitrate. These values meet the acceptance criteria, as they are below 10–20%, indicating good repeatability of the method.

CONCLUSION

Nitrite and nitrate levels in drinking water from various sources in the City of Medan were successfully determined using visible spectrophotometry at 536 nm. All samples contained detectable concentrations of nitrite and nitrate, and the measured levels remained below the maximum permissible limits established by national drinking water standards. The validated analytical method demonstrated good accuracy, precision, and sensitivity, indicating that it is suitable for routine monitoring of drinking water quality.

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