

# Technologies and Techniques for Detection of Nitrate in potable water: A Review

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**Abstract:** In agricultural and industrial production nitrate is widely used. Nitrate is appearing in water, environment, biology and food. Nitrate is a toxic inorganic contaminant, that's why it is perilous to the health living organisms and humans. In recent years, a variety of techniques has been developed for monitoring of nitrate. This paper aggregated as a general survey of the techniques proposed for nitrate monitoring and important monitoring parameters (such as detection limit, working pH, detection range and materials) were classified. This paper is composed of the sort of signal got from techniques, including optical and electrical signals. Electrochemical techniques get an electric signal from dissolved nitrate, with impedimetric, potentiometric and voltammetric techniques are included. Raman Spectrometry, fluorescence and absorption techniques receive an optical signal. In recent years, Biosensors are proposed for monitoring the nitrate in water. The limitations and disadvantages/advantages of the techniques are discussed in this paper. At last, techniques employed to perform nitrate, monitoring is summarized, and their future scope and development are discussed.

**Keywords:** Nitrate, Electrochemical Detection, Spectroscopy Detection

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## 1. INTRODUCTION

Industries like agriculture, food preservation, pharmaceuticals used nitrate [1]. Nitrate also used in meat curing [2, 3], dyes, bleaches [4]. Nitrate is a toxic inorganic contaminant that is dangerous to the living organism and human health. Due to high nitrate concentration "Blue baby syndrome" or Methemoglobinemia [7], gastric cancer [9], abortions [11], carcinogenic nitrosamines [8], spontaneous intrauterine growth restriction [10], happens. Aquatic animals including, fish and crustaceans damages due to the nitrate through its toxicity. As nitrite can oxidize hemoglobin to methemoglobin, which is not capable of carrying oxygen, the latter can reach toxic concentrations in a high-density aquaculture system in contaminated waters. In any case, different studies have proposed that methemoglobinemia may not be the essential system of nitrite poisonous quality. [13]

In aquatic animals, nitrite can be taken up across gill epithelia and can be accumulated to a high concentration in bodily fluids; thus, there is a greater risk for aquatic animals than for terrestrial animals. Moreover, high nitrate concentration has created impressive monetary misfortunes to aquaculture generation. Different type of fish has been researched to decide the relationship between nitrate fixation and fish ailments. The harmfulness of nitrate to fish varies with other fish species. Relative standard has been established to limit the concentration of nitrate in aquaculture water and drinking water because of the damages held by a high concentration of nitrate to aqua animals and human health. The World Health Organization (WHO) set the safe limit of nitrate in potable water is 10 mg/l. [18]. Similar, the safe limit of nitrate in meat production is 250ppm. This same safe limit of nitrate in potable water is followed by other countries. From above limitations, few techniques have been developed for monitoring and removal of nitrate in potable water. This technique includes bio methods [20-22] and electrochemical/chemical methods [19]. Due to the impact

and toxicity of nitrate on biological systems, industry, environment and agriculture, our need and longing to detect this particle are undeniable. Different technology has been produced to overcome potential interferences that would be detected within different solutions and trace level of detection. In Electrochemical methodologies nitrate ions convert into potential difference, current signal [23], this technique includes impedimetric electrodes [28, 29], voltammetric [24, 25] and potentiometric [26, 27]. This technique is simple, requires no time-consuming or complex pre-treatment and also don't requires any external reagent; in addition, this detection system is easily designed and inexpensive. Spectroscopic techniques converts the presence of nitrite ion to optical signals, this techniques include Raman spectroscopy [34, 35], absorption spectrometry [32, 33], fluorescence spectrometry [30, 31]. Spectroscopic techniques can usually reach a very low detection limit with good precision. Combined with separation and enrichment techniques, such as liquid extraction, chromatography, and capillary electrophoresis the detection limit can be further decreased. To execute automatic and continuous detection, related methods and flow injection analysis [30, 32, 36-38] have already been introduced and produced and include microfluidic, on-chip analysis and sequential flow injection analysis. Biosensors [39, 40] can basically be arranged in classes as electrochemical electrodes that get an electrical signal from an analyte through chemical reactions. But biosensors are different from traditional electrodes, to achieve catalytic activity, specificity, and selectivity used biologically active materials. The application of biosensors to nitrite detection has attracted much attention due to their specificity and higher sensitivity.

A number of excellent reviews have been compiled over the past decade. [23, 41-47] Also, Detection technology and requirements have been developed in recent years. The purpose of this paper is to study different nitrate detection technology proposed in recent years and also study their technologies, disadvantages/advantages. This paper is

arranged based on the type of signal that obtained from the methods, including optical signals and electric. Biosensors are recommended as a new monitoring method. By tabulating the various analytical parameters (including detection limit, materials, detection range, stability and working pH) of each method, their research and performance trends can be observed. At last limitation and disadvantages/advantages of each method are discussed.

## 2. ELECTROCHEMICAL TECHNIQUES:

Electrochemical detection methods have been examined for real-time monitoring and in situ quantitative analysis of environmental parameters. [23, 43, 46] Methods accustomed to monitoring nitrate can be separated into a number of groups, of which impedimetric methods and voltammetric, potentiometric are regularly introduced. Papers using these groups are compiled, and their performances are calculated.

### 2.1 Potentiometric Electrodes Method:

The potentiometric electrode detects nitrate ions with the help of organic membranes that have a suitable ion-exchanger or ionosphere with precise binding empathy for the objective ion and carries a scrupulous charged species from the sample to the electrode area. Due to the appearance of a charged species, a potential difference is formed between the indicator and reference electrodes, without no any species are produced or consumed and without current flowing between electrodes. The potential difference varies with the logarithm of the concentration under the condition that the concentration of the ion of interest is sufficiently low that the activity coefficients can be considered constant; if not, the response curve should be calibrated.

In recent years, Potentiometer method with ion-selective electrodes has enhanced, especially by achieve

incredibly low detection limits. Signal selectivity to the analytic of interest, low detection limit [23], applicability to turbid samples and color [48] and the ability to probe a huge range of species that are not red ox-active in aqueous environments these are the key advantages of the potentiometric method. The required instrumentation of this method is simple to use, portable, simple to fabricate and cheap. Many researchers have required devising ion-selective electrodes, frequently covered with membranes integrated with suitable ionospheres, for potentiometric detection of nitrate. Certainly, neither option is mainly positive. In potentiometric methods, several issues are survived like interference from other species, low electrode response, potential drift with time and common variation of reference potential, impracticable miniaturization due to unstable potential when the electrode approaches micrometer dimension [23]. In the potentiometric method, two electrodes are used. The working electrodes give a reference from An Ag/AgCl electrode. Sensitivity and selectivity give just before the species of interest from working electrode, is more complicated than the reference electrode. Generally, biosensors go through poor stability due to the frailty of the protein structure, denaturation due to protein unfolding or as the activity of powerless enzymes may be quickly annihilated by inhibition processes, harsh chemical circumstances or high temperatures. In that perspective, immobilized biomimetic compounds were prepared to replace biological macromolecules that mimic the activity of the enzymes on an electrode surface that should be more stable. [49] Consider and co-workers established a cobalt(II) deuteron- porphyrin derived that was electropolymerized with the ability to perform potentiometric recognition of nitrate by recording the change of the decreasing potential of [Dp Co(II) NO<sub>3</sub>-].

Method	Material	Working pH	Detection limit	Linear range	Stability	Reference
Potentiometric	salophen composite in a PVC matrix	4.5-11.9	$8 \times 10^{-7}$ M	$1 \times 10^{-6}$ - $1 \times 10^{-1}$ M	Can be used at least 2 months without deviation	[94]
Potentiometric	Sal opens and Rhodium porphyrins in a polymeric membrane electrode	4.5	$5 \times 10^{-6}$ M	N/A	66days, 89% remained	[27]
Potentiometric	Poly(pyrrole-cobalt(II)Deuter porphyrin)	N/A	N/A	$2 \times 10^{-6}$ M	N/A	[91]
Potentiometric	bulk acoustic wave impedance sensor	N/A	$1.7 \times 10^{-6}$ M	1.00mM	1.76%	[95]

2.1 Parameters and performances of potentiometric and impedimetric electrodes.

The cobalt (III) based complexes conveyed cannot effectively -categorize in contradiction of the most lipophilic anions like thiocyanate and salicylate.[50] Though, a PVC-based membrane nitrate sensor depend on the Co(II)-salophen complex (CSC) has also been informed that exhibits noble selectivity over fluoride, bromide, iodide, sulphite, nitrite, thiocyanate, triiodide and perchlorate [51] more on, polymeric membrane electrodes depend on rhodium(III) porphyrins and salophen's as Ionophores have been offered with better nitrate selectivity over thiocyanate, perchlorate, and salicylate. The best nitrate selectivity and longest functional lifetimes were gained with membranes doped with Rh-tBTPP and carboxylate PVC, correspondingly. The response time can be moderately reduced by employing polymer matrix additives such as polyurethanes or carboxylate PVC. [27]

PVC membrane electrodes collaborated with nitrate-selective carriers has been invented as a nitrate-responsive detector with good selectivity. [52-55] Though, on certain solid substrates, the PVC membrane has poor adhesion, like silicon chips; therefore, further polymeric matrices have been discovered, containing polyurethane (PU), functionalized PVC, poly (methacrylate) and silicone rubber, attended by poor limited plasticizer compatibility and electrochemical performance. Malinowski examined an anion-selective electrode depend on metal (III) porphyrin ionophores in polyurethane membranes, with potentiometric responses to nitrate obtained. Noteworthy potentiometric anion response and selectivity of the metal (III) porphyrin membranes were also detected in the presence of endogenous cationic sites in PU; in contrast, the anionic sites in PVC have no exogenous lipophilic sites added. [56]

## 2.2 Voltammetric Electrodes:

Amperometric electrode or voltammetry electrode gives a current signal to represent the rate of reactions on the probe surface though a potential is applied to the working electrode. To obtain a strong electrode response to nitrate oxidation, as well as response time and sensitivity and to ignore the oxygen interference, potential applied to the working electrode is calculated. [57] Reference, working, and counter electrodes exist in a typical measuring system. To convert the working probe current to voltage, also amplify this voltage in a suitable range for the analog-to-digital converter to sample, the measuring system requires current-to-voltage converter and voltage amplifier. Then, at last, the result obtain from the measuring system is calculated, displayed on an LCD or local monitor, saved and transmitted.

Voltammetric electrode technique has been employed to monitor and detect nitrate since the early 1900s, at that time glassy carbon electrodes were used for real-time monitoring without any external reagent. An excessive number of substrates have since been examined for voltammetry detection and to increase electrode performance, like nickel, copper, platinum, carbon, cadmium, gold, lead boron-doped diamond, indium tin oxide. [58, 59]

## 3. BIOSENSORS:

For nitrate detection variety of biosensors has been developed. To perform nitrite detection Biosensors are used. [29] As biosensors typically express higher specificity and sensitivity, there is developing an interest in their examination for direct monitoring of nitrate. In biosensors some modifiers are used for nitrate detection in potable water, like copper-containing nitrite reductive, Cu-NiR, PAPS-SO<sub>3</sub>H-V [60], myoglobin [61]. Biosensing, a theory that invented based on nature. Also, biosensing is a topic of great attention in recent years. The biosensor is a systematic device; consist of a biological recognition component like a detector and transducer. The bioreceptor is either closely combined or connected to the transducer. For the generation of a measurable signal, analytic bioreceptor bind event on the surface of biosensor [62–64]. The main challenge related to the fabrication of an electrochemical biosensor lies in the Formation of an electrical communication between the signal transducers and the biological Recognition [65]. By choosing suitable mediators, modes of immobilization nanomaterials immobilization matrices and nanomaterial's, the effectiveness of the electrical communication between an electrode and the detection element is improved.

### 3.1 Growth Of The Biosensors Technology:

The chemistry of the enzymatic reduction of nitrate involves a flow of electrons from an electron donor, like a mediator or NADH through the enzyme NaR to achieve reduction into nitrite [66]. To reduce nitrate from water, generally, NaR used two electrons from the electron mediator or donor [67, 68]. Lately, NaR along with a suitable cofactor have been used in dissimilar configurations for the construction of nitrate biosensors. In past, major hard work to expand nitrate biosensors has been directed to immobilization inside or non-conducting polymers and on conducting.

## 4. SPECTROPHOTOMETRIC METHODS:

Spectrophotometric methods are the most widely used method for nitrate detection in water [69]. Nitrate detection by direct UV spectrophotometer provides a rapid analytical method [70], But this technique is topic to severe interferences from ionic species and organic matter. Accordingly, its application is restricted to non-saline waters with high levels of nitrate and very low organic matter content. Consequently, colorimetric methods are mostly used for nitrate detection. The most frequently used colorimetric assay is based on the reaction of nitrite, sulphanilamide, and N-1-naphtylethylenediamine under acidic conditions, which results in the formation of a pink azo dye. Nitrite resulting from nitrate diminution plus any other nitrite present in the sample reacts with sulphanilamide at pH 4.1 and results in the formation of p-diazoniumsulphanilamide. Nitrate diminution is usually achieved via biological or chemical methods. The commonly used chemical reduction methods. The spectroscopic method consists of a source, detector calumniating lens, and signal conditioning. The absorption of a specific is measured to quantify the amount of a specific substance, as the substance can absorb energy (photons)

from radiation of a specific wavelength. The absorption spectrum is usually measured by detecting the intensity of the radiation that passes through the substance upon irradiation with a specific wavelength. Light with different wavelengths has been employed for nitrite detection. In addition, ultraviolet radiation has been used as an absorption photometric method for nitrite detection. Different ions have different absorption peaks at a corresponding wavelength. Therefore, a specific analyte can be detected by measuring the absorbance at a certain wavelength. Other wavelengths are needed to distinguish or detect interfering ions.

## 5. OTHER IMPORTANT NAR BASED METHODS FOR NITRATE DETERMINATION:

Some other strategy has been adopted for nitrate which is originally depending on NaRcatalyzed reduction of nitrate to nitrite. Binnerup et al. developed a highly sensitive denitrification bioassay developed [71] for the detection of  $\text{NO}_3^-$  and  $\text{NO}_2^-$  in rhizosphere soil samples occupied the use of anaerobically grown *Pseudomonas aeruginosa* denitrifying bacteria. The bioassay separates both the  $\text{NO}_3^-$  and  $\text{NO}_2^-$  pools from the other N compounds in the sample and is, therefore, a useful tool for preparing samples for  $^{15}\text{N}$  isotope analysis by mass or emission spectrometry. Jensen et al. [72] used the nitrate microsensor technique to obtain nitrification and denitrification micro profiles from stratified microbial communities. A NaR assay was useful to an autoanalyzer by Corso et al. [73] in an effort to build up an accurate, rapid and convenient spectrophotometric method for the determination of urinary nitrate in humans. These researchers compared the urinary nitrate levels in smokers and non-smokers and indicated that smoking is related to a diminished excretion of nitrate from the body. The method was linear between nitrate concentrations of 0.062 mM and 1.00 mM. Urinary nitrate determination also has potential utility for the evaluation of in vivo synthesis of nitric oxide, which is the major endothelium-derived relaxing factor.

## 6. CONCLUSION:

Electrochemical sensors and biosensors, which are simple, inexpensive and easily miniaturized, have been investigated for many years to improve their selectivity and sensitivity. These sensors are suitable for miniaturization and long-term monitoring. Compared with spectroscopy, their detection limit is somewhat higher. But they are easily used and require no reagents or complex instruments. Nitrate biosensors provide an excellent alternative to the chemical detection methods for nitrate determination because of advantages such as high selectivity, simplicity and ability to be used for online and continuous monitoring. Spectroscopic methodologies can get very low detection limits and can be used to detect trace amounts. At the same time, reagents are required by spectroscopic methods to perform detection.

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