

Removal of Nitrite from Polluted Waters using Bio-sorbents derived from Powders of Leaves, Barks or Stems of Some Herbal Plants

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ABSTRACT

An investigation is made to explore the surface sorption abilities of activated powders of leaves, stems or barks of some herbal plants. *Phyllanthus niruri*, *Annona squamosa*, *Tridax procumbens* and *Morinda tinctori* are found to have affinity towards nitrite. Conditions like pH, time of equilibration and sorbent concentration have been optimized. Stem or bark powders show more sorption than leaves powders. Ten fold excess of common ions present in natural waters show less effect on the % maximum removal of nitrites at the optimum conditions of extraction. Successful sorbents have been developed to remove more than 90% of nitrites from polluted waters. The application of these sorbents has been tested with naturally available polluted water samples.

Keywords: Nitrites; pollution control; bio-adsorbents.

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INTRODUCTION

Nitrite is a pollutant present in waste waters due to incomplete oxidation of nitrogenous organic matter. It is used in western countries as a preservative for meat to prevent botulism and also impart attracting dark red color and this turns to be one of the potential source of nitrite contamination [3,29,30].

Nitrite is skin irritant and its high concentrations cause a state of anoxia in the blood known as methemoglobinemia and it results in cancer due its ability to form Carcinogenic nitrosamine and N-nitroso compounds; half of the Cancer deaths in developed countries is attributed to nitrite though nitrite and nitrate levels are within legal limits[3,11,20,30,42,43].

Nitrite and nitrates are interconvertable through reduction or oxidation [3, 15]. Reducing conditions in waste waters convert nitrate to nitrite. The maximum permissible limits for nitrite and nitrate are: EU: 0.5 ppm and 50 ppm; USA: 1 ppm and 10 ppm respectively. Though the limits seems to be low but daily consumption results in "bio-amplification" of these compounds due to human metabolic activity. China in 2005 passed most stringent laws on nitrite pollution to counter the nitrite prone cancer mortality and reduced the permissible limit to 0.005 to 0.002 ppm in bottled water and set a limit of 0.02 ppm for ground water recharge. Russia, Japan and Taiwan also have imposed strict nitrite pollution levels in view of cancer prone epidemiological findings. Recent research works suggest that nitrite does not convert healthy cell into cancer cells but it stimulate the growth of cancer cells; the degree of stimulation depends upon various factors such as concentration of nitrite and age and type of cancer cell.

There has been attempts to remove nitrite along with other nitrogen compounds [10,12,31] and the methods exclusively developed for the removal of nitrite are few and far between [1,6,7,20,32,35,36]. Katsuya Abe et al [18] investigated the aerial microalgae *Trentepohlia aurea* in relation to the removal characteristics of nitrites. Polatides et al [28] studied the Electro chemical removal of nitrate and nitrite ions from aqueous solutions by pulsing potential electrolysis. De. D. Kalu et al [6] made kinetic studies of the electrochemical treatment of nitrite and nitrate ions on Iridium-modified Carbon Fiber Electrode. Saleem M et. al. [32] studied electrochemical removal of nitrite in simulated aquaculture. Nes,e.Ozturk et al [26] investigated the removal of nitrite from aqueous solutions by adsorption using Sepiolite and powdered activated carbon. A patent product under the band name ALGONE is available for removal of nitrites in the marine aquarium.

The use of agricultural waste products as bio-sorbents for the removal of polluting ions offer a potential alternative to the existing methods of detoxification and recovery of toxic and valuable ions in polluted waters [2,4,13,16,27,34,38,40,41]. These biological approaches in the recent past have stimulated continuous and expanding research in this field [5,8,14,17,19,21-23,33,39]

In the present work an attempt is made to explore the use of leaf, stems or bark powders of some herbal plants in controlling the nitrite concentrations in polluted waters.

MATERIALS AND METHOD

Chemicals and solutions

All Chemicals used were of analytical grade.

- **Stock Solutions of Nitrite:** 500 ppm stock was prepared by dissolving suitable amount of A.R. grade Sodium Nitrite in a suitable volume of double distilled water and it is diluted as per the need.
- **Sulphanilic acid reagent:** 0.6 g of A.R. Sulphanilic acid was dissolved in 100 ml of 20% v/v hydrochloric acid.
- **α -Naphthylamine reagent:** 0.48 g of A.R. α -naphthylamine was dissolved in 100 ml of 1.3 % v/v hydrochloric acid.
- **Sodium acetate, 2 M:** 16.4 g of anhydrous sodium acetate was dissolved in 100 ml of distilled water.

Adsorbents

Thermally activated powders of leaves, stems or barks of many plants have been employed in this work. It is found that leaves, stems or barks of *Phyllanthus niruri*, *Annona squamosa*, *Tridax procumbens* and *Morinda tinctoria* have been found to have affinity towards nitrites and hence the methodology presented here, pertains to these sorbents only.



Phyllanthus Niruri

Annona squamosa

Tridax procumbens

Morinda tinctoria

Phyllanthus Niruri is a herb belongs to *Phyllanthaceae* family and is found in Central and Southern India; it is found to have many therapeutic values in curing jaundice, diabetes, dyspepsia, ulcers, sores, swellings, ophthalmia and chronic dysentery. *Annona squamosa* is a small well-branched shrub that bears edible fruits called sugar-apple; belongs to *Annonaceae* family and grows well in lower altitudes. *Tridax procumbens* is a species of flowering plant in the daisy family and is best known as a widespread weed and pest plant. It grows in tropical, subtropical and mild temperate regions worldwide. It possesses medicinal uses especially for diabetic treatment. The oral administration of leaf extracts at doses of 200 mg kg⁻¹ lead to a significant blood glucose reduction. This laid the foundation to study the active compounds of such anti-diabetic plants that are responsible for the hypoglycemic activities. It also proves the traditional claim of Mandesh region with regard to *Tridax procumbens* for its anti-diabetic activity [9]. *Morinda tinctoria*, commonly known as Aal or Indian Mulberry is a species of flowering plant and belongs to *Rubiaceae* family and is native to South Asia. It is an evergreen shrub growing to 5-10 m tall. The plant is extensively cultivated in India and its leaves and roots are used in traditional system of medicine, as astringent, deobstrent, emmenagogue and to relieve pain in the gout [25]. It is reported to have anticonvulsant activity [37].

Sorbent Preparation

The leaves or stems or barks of *Phyllanthus Niruri*, *Annona squamosa*, *Tridax procumbens* and *Morinda tinctoria* were cut or scrapped freshly, washed with tap water, then with distilled water and then sun dried. The dried materials were powdered to a fine mesh of size < 75 μ and activated at 105^o C for 4 hrs. in an oven and then these were employed in this work.

Adsorption Experiment

The Batch system of extraction procedure was adopted [13, 24, 38]. Carefully weighed quantities of adsorbents were taken into previously washed 1 lit/500 ml stopper bottles containing 500 ml /250 ml of nitrite solutions of predetermined concentrations. The various initial pH values of the suspensions were adjusted with dil. HCL or dil. NaOH solution using pH meter. The samples were shaken in Mechanical shakers for a desired period and after the equilibration period, an aliquot of the sample was taken, filtered and the nitrite content in it is determined spectrophotometrically [44].

Estimation of Nitrites

A measured quantity of the nitrite sample was taken in to a 50 ml volumetric flask. To it 1 ml of Sulphanilic acid reagent solution was added, mixed well and was allowed to stand at least 3 min and not more than 10 min at room temp in diffused light. Then 1 ml of the α -Naphthylamine solution and 1 ml of 2M-Sodium acetate solution to act as buffer (pH:2.0-2.5) were added. The solution then diluted to 50 ml and mixed well. After 10 min, but not later than 30 min, the Optical Density of the reddish-purple color developed was measured at 520 nm against blank using U.V and visible spectrophotometer (Systronics make). Previously, a standard graph was constructed between O.D. and known concentrations of nitrite, in conformation of Beer's law adopting Least Square Method for linear graphs. The measured O.D. values for the unknown solutions were referred to standard graph to determine the concentrations of nitrite in un-known samples.

Effect of Interfering ions

The interfering ions chosen for study are the common ions present in natural waters, viz., phosphate, sulphate, fluoride, chloride, carbonate, calcium, magnesium, copper and zinc. The synthetic mixtures of nitrite and one of the interfering ions were so made that the concentration of the interfering ions maintained at ten fold excess than the nitrate ion concentration. 500 ml of these solutions were taken in stopped bottles and then correctly weighed optimum quantities of the promising sorbents were added. Optimum pH was adjusted with dil. HCl or dil. NaOH using pH meter. The samples were shaken in shaking machines for desired optimum periods and then the samples were filtered and analyzed for nitrites. % of extraction was calculated from the data obtained. The results are presented in the Table-1.

Applications of the developed bio-sorbents

The workability of the developed bio-sorbents for removing nitrate ion concentrations in naturally occurring polluted waters was tried. For this purpose, five samples were collected from five lakes polluted with nitrite at different places in Bapatla mandalam of Guntur Dt of Andhra Pradesh and the actual concentration of nitrite present in them was analyzed. Then these samples were subjected to extraction for nitrites using the bio-sorbents developed in this work at optimum conditions of pH, equilibration time and sorbent concentration. The results obtained were presented in the Table-2.

RESULTS AND DISCUSSION

Of the various sorbets tried, the leaves or stems or barks of *Phyllanthus niruri*, *Annona squamosa*, *Tridax procumbens*, *Morinda tinctoria* have been found to have affinity towards nitrites. The sorption characteristics of these adsorbents have been studied with respect to various physicochemical parameters. The results are presented in the Graph No. A1-8, B-1 and 2, C-1 and 2. The following observations are significant:

Time of equilibration

% of extractability increases with time for a fixed sorbent and at fixed pH and after certain time, the extractability remains constant i.e. an equilibrium state has been reached.

pH

% of extraction is found to be pH sensitive. As pH decreases, % of extraction is increased (Vide Graph: B-1 and 2). As for example, with the powder of leaves of *Tridax procumbens*, % of extractability, is found to be 22.4% at pH 10; 34.5% at pH 8; 38.9% at pH 6; 50.6% at pH 4 and 78.0% at pH 2. The same trend is found in other sorbents also.

The % of extractability

It is found to be more with *bark/stem powders* than with *powders of leaves*. As for example, the % of maximum extractability is found to be 70.0% with the activated leaves powder of *Moringa tinctoria* at pH 2 and equilibration time of 6 hrs while with activated bark powders of *Moringa tinctoria*, % of extractability is enhanced to 93.0% at pH 2 and after an equilibration time of 5 hrs. The same trend is found in other sorbents also.

Time of equilibration

It is needed for maximum extractability of nitrite is found to be less for bark or stem powders than with the powder of leaves. With leaves powders of *Phyllanthus niruri*, *Annona squamosa*, *Tridax procumbens* and *Morinda tinctoria*, the equilibration time needed for maximum extraction is found to be 6 hrs at pH 2 while with bark/stems powders of *Phyllanthus niruri*, *Annona squamosa* and *Tridax procumbens*, the equilibration time decreases to five hrs at the same pH and further, in the case of bark powders of *Morinda tinctoria*, 80% extraction is found even at 0.5 hrs of equilibration time (vide Graph Nos:A-1 to 8).

Sorbent Concentration

The adsorbent dosage needed for maximum extraction of nitrite is found to be less in the case of bark/stem powder than the powders of leaves (Vide Graph Nos:C-1 & 2). Sorbent concentration for maximum extraction at optimum

conditions of pH and equilibration time is found to be 1.5 gram/lit for the leaves of *Phyllanthus niruri* while with its barks powder, sorption concentration has been reduced to 1 gm/lit. Similarly, with the powders of leaves of *Annona squamosa*, optimum sorbent concentration is 2 gm/lit but with its bark powder 1 gm/lit is sufficient. In the case of *Tridax procumbens* and *Morinda tinctoria*, the leaves powder needs 4 gms/lit while their bark powders need only 2 gm/lit

The % of maximum extractability is found to be 98% with the powders of *Phyllanthus neruri* leaves at pH 2 and after an equilibration period of 6 hrs with sorbent concentration of 1.5 gms/lit while with its bark powders, the % of extraction has been increased to 100% at pH 2 and after an equilibration period of 5 hrs with sorbent concentration of 1.0 gm/lit. At pH 2 and equilibration period of 6 hrs, % of extraction is found to be 96% with leaves powder of *Annona squamosa* at sorbent concentration of 2 gm/lit; 78.0% with leaves powder of *Tridax procumbens* with adsorbent dosage of 4 gms/lit and 70 % with leaves powder of *Morinda tinctoria* with sorbent dosage of 4 gms/lit.

With bark/stem powders of *Phyllanthus niruri*, *Annona squamosa*, *Tridax procumbens* and *Morinda tinctoria*, the maximum extractability is found to be 100%, 98%, 90% and 93% respectively at pH 2 and after an equilibration time 5 hrs and at optimum dosages of adsorbents. The optimum dosages are found to be 1.0 gm/lit for *Phyllanthus niruri* and *Annona squamosa*; 2 gms/lit for *Tridax procumbens* and *Morinda tinctoria*.

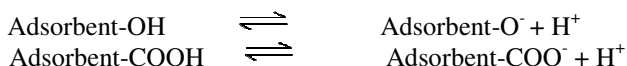
Interfering ions

The extractability of Nitrite ions in presence of ten fold excess of common ions found in natural waters, namely, Chloride, Fluoride, Carbonate, Sulphate, Phosphate, Calcium, Magnesium, Copper and Zinc ions, has been studied. These ions show less effect on the extractability of nitrite ions at the optimum conditions of pH, equilibration time and sorbent dosage (Table 1).

Applications

Sample waters at five lakes polluted with nitrite ions have been collected and the successful sorbents developed in this work have been tried for the removal of nitrite ions at optimum conditions of extraction. It is found that the sorbents developed in this work are successful in removing nitrites considerably in these samples of water at optimum conditions of pH, equilibration time and sorbent dosage. % removal of nitrite in these samples of water is found to be: 90.5 to 94.2% with leaves powder of *Phyllanthus niruri*; 90.6% to 93.1% with leaves powder of *Annona squamosa*; 76.2% to 77.5% with leaves powder of *Tridax procumbens*; and 67.0% to 69.5% with leaves powder of *Morinda tinctoria*. With activated bark powders the extractability is found to be 90.0% to 93.8% for *Phyllanthus niruri*; 92.0% to 96.5% for *Annona squamosa*; 84.2 to 89.1% for *Tridax procumbens* and 87.1% to 89.2% for *Morinda tinctoria* (Table 2).

The remarkable affinity the sorbents, developed in this research work, towards nitrite may be due to the naturally occurring compounds in the leaves, stems or barks and further probe into this aspect of research is beyond the scope of this work. Moreover, these lingo celluloses' materials have -OH/COOH groups and their dissociation is pH dependent. At high pH values, these groups dissociates as:



and imparts weak cation exchange ability and at low pH values, the dissociation is less favored and protonation may also occur:



resulting weak anion-exchange ability. This is reflected in sorption of nitrite. Nitrite being an anion is adsorbed by these materials at low pH values and hence the high % of removal. At high pH values cation affinity persists and not anion and hence, nitrite anion show low % of removal.

The decrease in the rate of sorption with the progress in the equilibration time may be due to the more availability of sorption sites initially and are progressively used up with time due to the formation of adsorbate, nitrite, film on the sites of the sorbent and thus resulting in decrease in sorption ability of the adsorbents.

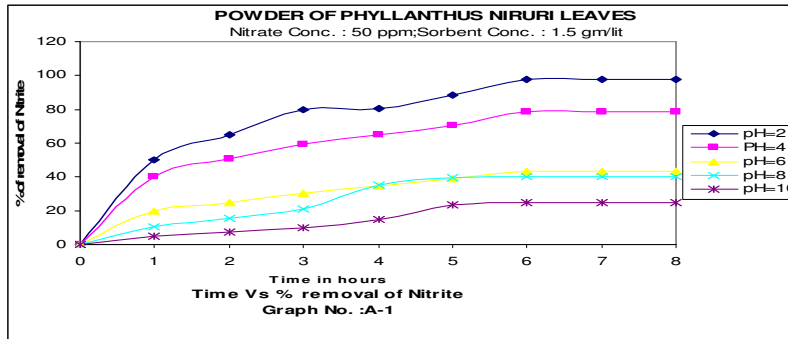


Fig.-1

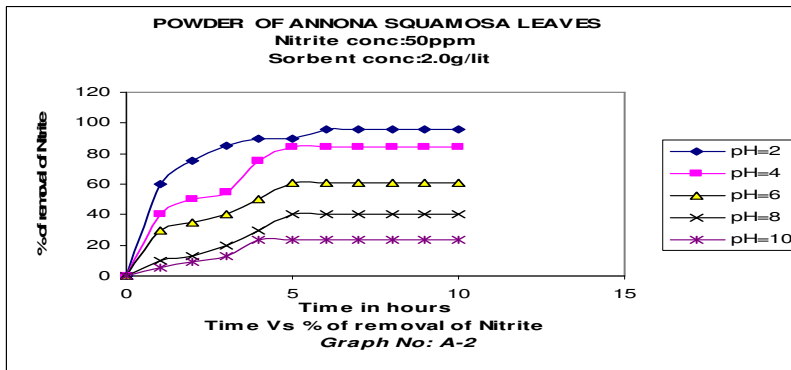


Fig.-2

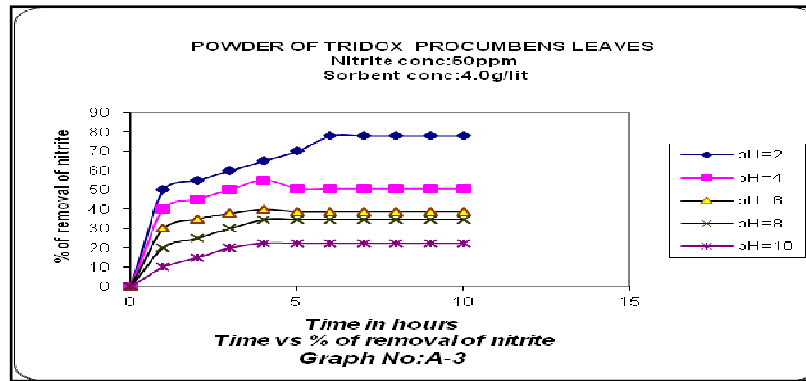


Fig.-3

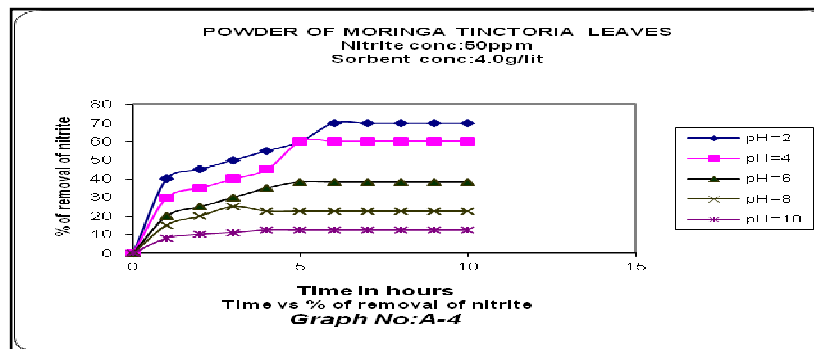


Fig.-4

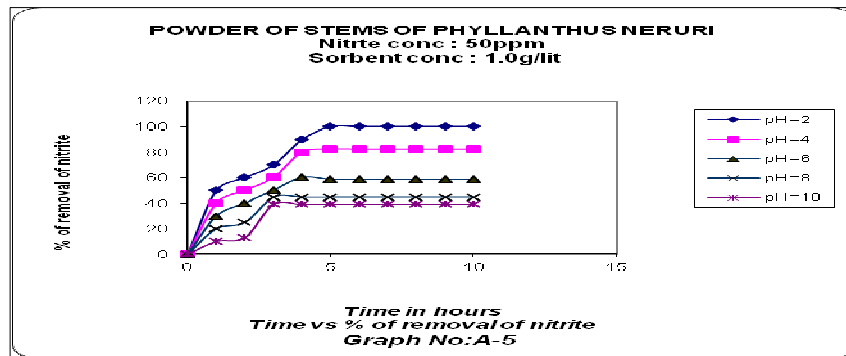


Fig.-5

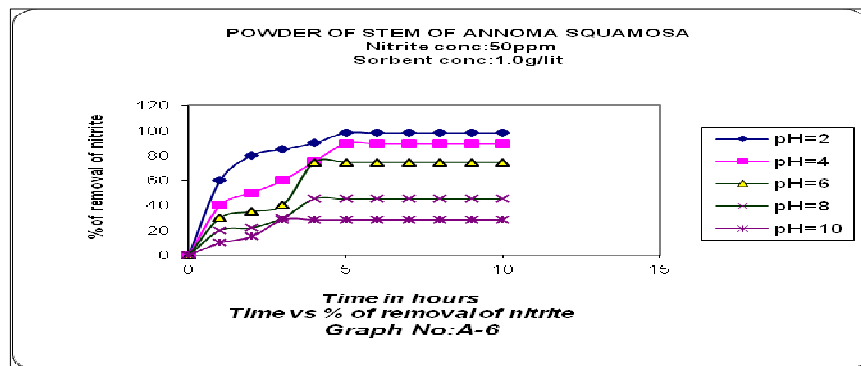


Fig.-6

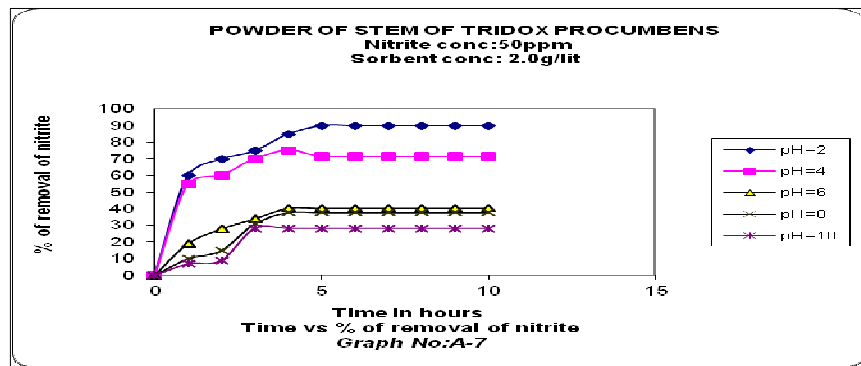


Fig.-7

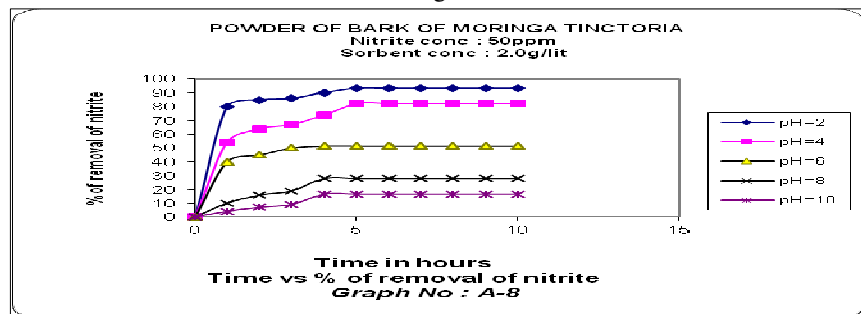


Fig.-8

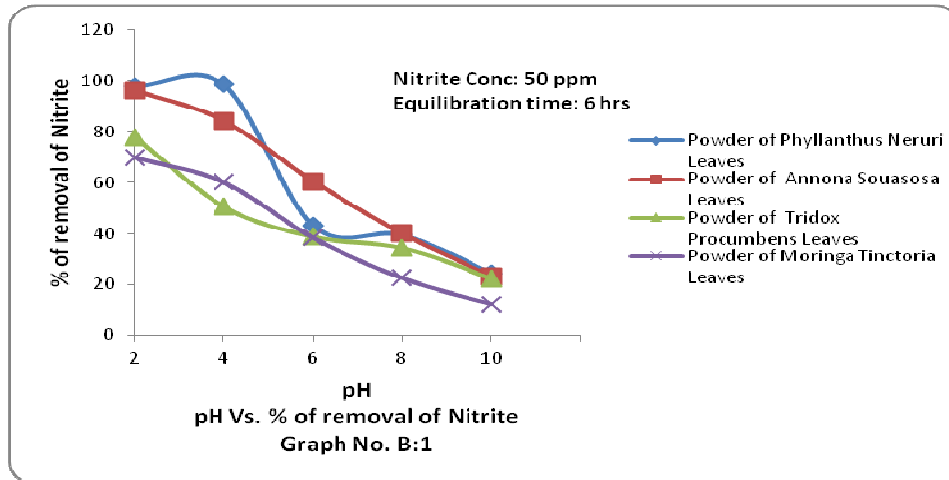


Fig.-9

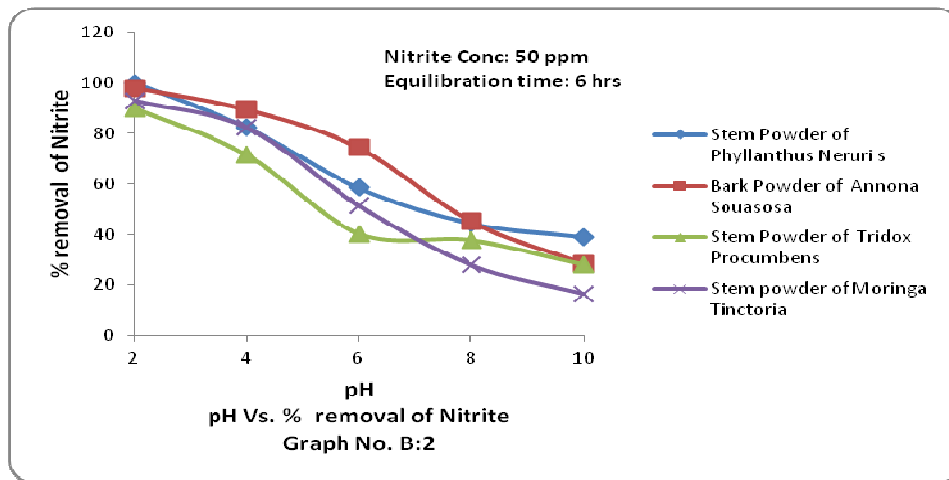


Fig.-10

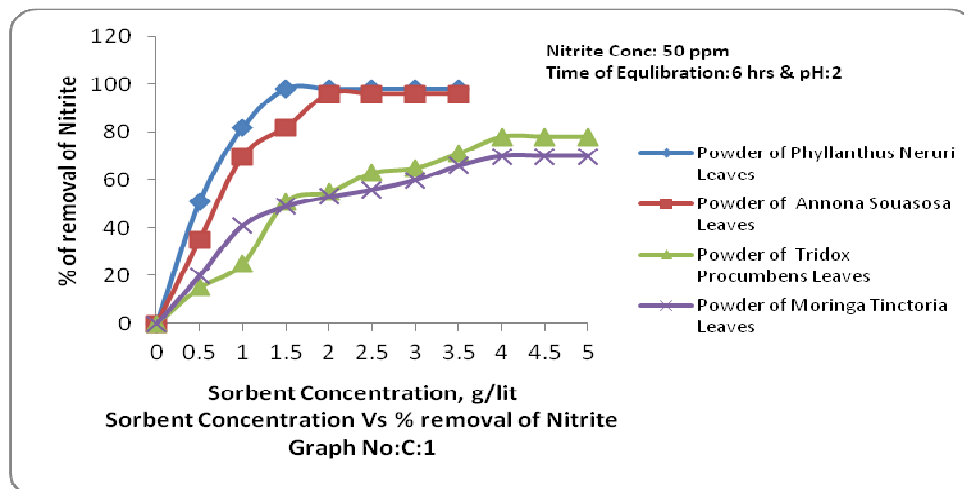


Fig.-11

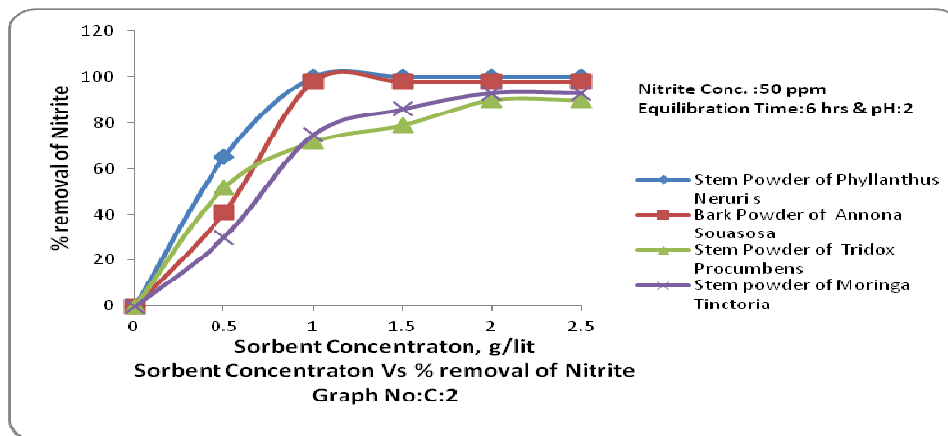


Fig.-12

Table-1: Effect of interfering Ions on the Extractability of Nitrite with different Bio-sorbents

| S.No. | Adsorbent and its concentration | Maximum Extractability at optimum conditions | Extractability of Nitrite in presence Ten fold excess of (500ppm) interfering ions at optimum conditions: Conc of Nitrite: 50 ppm | | | | | | | | |
|-------|---|--|---|------------|-----------------|-------------------------------|----------------|------------------|------------------|------------------|------------------|
| | | | SO ₄ ²⁻ | Phosphates | Cl ⁻ | CO ₃ ²⁻ | F ⁻ | Ca ²⁺ | Cu ²⁺ | Zn ²⁺ | Mg ²⁺ |
| 1 | Powder of Phyllanthus neruri leaves: 0.5 gms/lit | 98.0%; pH 2, 6.0hrs | 83.3% | 94.2% | 96.5% | 97.5% | 94.7% | 94.2% | 90.4% | 94.5% | 92.8 |
| 2 | Powder of Annona squamosa leaves 2.0 gms/lit | 96.0% pH 2, 6.0 hrs | 85.4% | 93.5% | 94.3% | 95.4% | 91.5% | 92.5% | 93.9% | 92.3% | 93.8% |
| 3 | Powder of Tridox procumbens leaves 4.0 gms/lit | 78.0% pH 2, 6.0hrs | 70.2% | 77.1% | 74.1% | 76.3% | 73.6% | 76.0% | 73.1% | 72.2% | 74.0% |
| 4 | Powder of Moringa tinctoria leaves 4.0 gms/lit | 70.0% pH 2, 6.0 hrs | 66.4% | 65.4% | 66.1% | 69.7% | 64.5% | 67.1% | 68.3% | 65.7% | 67.8% |
| 5 | Powder of stems of Phyllanthus neruri 1.0 gms/lit | 100.0% pH 2, 4.0 hrs | 87.8% | 97.2% | 95.4% | 97.5% | 95.5% | 97.0% | 98.3% | 97.8% | 96.4% |
| 6 | Powder of barks of Annona squamosa 1.0 gms/lit | 98.0% pH 2, 5.0 hrs | 84.8% | 96.3% | 95.8% | 96.1% | 94.6% | 94.8% | 93.4% | 95.6% | 94.8% |
| 7 | Powder of stems of | 90.0% pH 2, | 81.3% | 87.4% | 87.9% | 89.6% | 83.1% | 85.4% | 87.3% | 86.5% | 88.2% |

| | | | | | | | | | | | |
|---|---|---------------------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| | Tridox procumbens 2.0 gms/lit | 5.0 hrs | | | | | | | | | |
| 8 | Powder of stems of Moringa tinctoria 2.0 gms/lit | 93.0% pH 2, 5.0 hrs | 86.5% | 91.9% | 89.3% | 92.3% | 87.4% | 90.2% | 89.3% | 87.8% | 88.4% |

Table-2: Percentage of extractability of Nitrites in Natural samples with Bio-sorbents developed in this work

| Bio-Sorbent | % of Extractability of Nitrites | | | | |
|--|---|--|---|--|---|
| | Sample 1: Found to have 5 ppm of nitrite | Sample 2 Found to have 6.9 ppm of nitrite | Sample 3 Found to have 7.2 ppm of nitrite | Sample 4 Found to have 8.1 ppm nitrite | Sample 5 Found to have 6.5 ppm of nitrite |
| Leaves powder of Phyllanthus neruri at pH 2; Equilibration time: 6 hrs and sorbent concentration: 1.5 gm/lit | 93.0% | 94.2% | 92.1% | 90.5% | 92.5% |
| Leaves powder of Annona squamosa : at pH 2; Equilibration time: 6 hrs and sorbent concentration: 2.0 gm/lit | 91.3% | 93.1% | 90.6% | 91.9% | 92.1% |
| Leaves powder of Tridax procumbens at pH 2; Equilibration time: 6 hrs and sorbent concentration: 4.0 gms/lit | 76.2% | 77.1% | 76.8% | 77.5% | 76.4% |
| Leaves powder of Morinda tinctoria at pH 2; Equilibration time: 6 hrs and sorbent concentration: 4.0 gm/lit | 69.5% | 68.9% | 68.2% | 69.2% | 67.0% |
| Stems powder of Phyllanthus neruri at pH 2; Equilibration time: 5 hrs and sorbent concentration: 1.0 | 92.2% | 93.8% | 90.0% | 91.8% | 92.4% |

| gm/lit | | | | | |
|--|-------|-------|-------|-------|-------|
| Barks powder of <i>Annona squamosa</i> at pH 2; Equilibration time: 5 hrs and sorbent concentration: 1.0 gm/lit | 96.5% | 95.8% | 95.2% | 94.8% | 92.0% |
| Stems powder of <i>Tridax procumbens</i> at pH 2; Equilibration time: 5 hrs and sorbent concentration: 2.0 gms/lit | 89.1% | 88.0% | 86.0% | 85.8% | 84.2% |
| Stems powder of <i>Morinda tinctoria</i> at pH 2; Equilibration time: 5 hrs and sorbent concentration: 2.0 gm/lit | 87.3% | 89.2% | 88.1% | 87.1% | 87.2% |

CONCUSSIONS

1. Thermally activated powders of leaves or barks of *Phyllanthus niruri* , *Annona squamosa* , *Tridax procumbens* , *Morinda tinctoria* have been found to have strong affinity towards nitrates at low pH values.
2. % of removal of nitrite is pH sensitive and also depends on sorption concentration and time of equilibration
3. The minimum dosage needed for the maximum removal of nitrite is considerably less for the activated bark powders than for activated leaves powder.
4. Ten fold excess of common ions present in natural waters, viz., Cl^- , F^- , Sulphate, Carbonate, Ca^{2+} , Mg^{2+} , Cu^{2+} and Zn^{2+} has less affected the % of extraction of Nitrite at optimum conditions of pH , equilibration time and sorbent concentration.
5. We claim 98% ,96%,78% and 70% removal of nitrite with the activated leaves powders of *Phyllanthus niruri* , *Annona squamosa* , *Tridax procumbens* and *Morinda tinctoria* respectively at pH:2, 6 hrs of equilibration time and at optimum sorbent concentrations. The % of removal is enhanced to 100%, 98%,90% and 93% with the activated stem/bark powders of *Phyllanthus niruri* , *Annona squamosa* , *Tridax procumbens* and *Morinda tinctoria* respectively at pH 2 and at low equilibration times and sorbent concentrations.
6. The suitability of the developed sorbents are tested with respect to waste water samples collected from polluted waters of five lakes in different places in Bapatla Mandalam of Guntur Dt of Andhra Pradesh. The sorbents are found to be successful in the removal of considerable amounts of nitrites.

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