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Research Article

Preliminary Studies on Evaluation of Sapwood as Nanopore Filter: A Promising Low Cost Water Treatment Technique

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Abstract: In the present study an attempt was made to evaluate the efficiency of plant xylem in removing pollutants from drinking water. A total five locally available plant species (flowering plants) were randomly selected from the Ambo university campus and they were evaluated for their pollutant removal capabilities in terms of Bacterial, Solids and Color removal. All the plant species used in the study have shown varying degrees of filtration in terms of bacterial, total solids, and color removal. Filtration studies carried in both dry and wet xylem conditions. In all the studies, xylem in wet condition proved to be highly efficient in removing bacteria, solids and color than in dry condition. *Podocarpus falcatus* (Birbirs) and *Juniperus procera* showed highest percent removal of bacteria at the tune of 84 and 82% respectively. *Juniperus procera* has exhibited highest percent removal of solids (94%) followed by *Cordia africana* with a percent removal of 93. Least percent removal was observed with *Podocarpus falcatus* (86%) and moderate removal efficiencies were observed with other species. Most of the species were able remove color with varying degrees of efficiencies. In dry xylem studies, efficiency of plant species in removing bacteria, solids and color was found to be very poor. Filtration rates ranges from a minimum of 4L/D to a maximum of 7L/D. Highest filtration was observed with

Juniperus procera (Gaatiraa) and lowest filtration was associated with *Cordia Africana*.

Keywords: Plant xylem, Nano-pore filter, Bacteria & Color removal, Filtration rates.

INTRODUCTION

The lack of safe water creates a tremendous burden of diarrheal disease and other debilitating, life-threatening illnesses for people in the developing world. Point-of-use (POU) water treatment technology has emerged as an approach that empowers people and communities without access to safe water to improve water quality by treating it in the home. Several POU technologies are available, but, except for boiling, none have achieved sustained, large-scale use. Sustained use is essential if household water treatment technology (HWT) is to provide continued protection, but it is difficult to achieve. Ceramic and biosand household water filters are identified as most effective according to the evaluation criteria applied and as having the greatest potential to become widely used and sustainable for improving household water quality to reduce waterborne disease and death. This disease burden falls disproportionately on those in developing countries, where children experience multiple episodes of diarrheal disease each year¹. Recent systematic reviews of water, sanitation, and hygiene interventions suggest that the beneficial effects of improving household drinking water quality at the point of use (POU) to reduce diarrheal disease risks had been previously underestimated. Contemporary reviews estimate 30–40% reductions in diarrheal disease by improving household drinking water quality at the POU, making such treatment more effective than improvements at the source². Although a variety of POU technologies have been suggested, tested, and disseminated, not all have an evidence base of effectiveness and sustained use³. Peter-Varbanets *et al.*⁴ have outlined the key requirements for point-of-use devices for water disinfection: a) performance (ability to effectively remove pathogens), b) ease of use (no time-consuming maintenance or operation steps), c) sustainability (produced locally with limited use of chemicals and non-renewable energy), and d) social acceptability. Meeting all of these requirements has proved to be challenging, but point-of-use methods that have been successfully used for low-cost water treatment in developing countries include free-chlorine/solar disinfections, combined coagulant-chlorine disinfection, and biosand/ceramic filtrations⁵. Common technologies for water treatment/disinfection include chlorination, filtration, UV-disinfection, pasteurization/boiling, and ozone treatment. All these technologies, suffer from either with high costs, use of chemicals, and other barriers for the wide implementation in the developing countries especially Ethiopia. New approaches that can improve upon current technologies are urgently required. Specifically, Nanopore materials that are inexpensive, readily and naturally available, disposable and effective at pathogen removal could greatly impact our ability to provide safe drinking water to needy.

In this context, a potential solution exists in the form of plant xylem- a porous material that conducts fluid in the plants. In this context, a potential solution exists in the form of plant xylem- a porous material that conducts fluid in the plants. Plants have evolved specialized xylem tissues to conduct sap from their roots to shoots. Xylem has evolved under the competing pressures of offering minimal resistance to the ascent of sap fluid while maintaining small nanoscale pores to prevent cavitation. The size distribution of these pores typically a few nanometers to a maximum of around 500nm depending on the plant species also happens to be ideal for filtering out pathogens most commonly found in drinking water. Although scientists have extensively studied plant xylem for various purposes, no studies were conducted on use of plant xylem for water filtration especially for the removal of pathogenic water borne bacteria. The simple construction of xylem filters, combined with

their fabrication from an inexpensive, biodegradable, and disposable material suggests that further research and development of xylem filters could potentially lead to their widespread use and greatly reduce the incidence of waterborne infectious disease in the world. Hence in this study an attempt was made to use the plant xylem as an effective Nanopore filter.

MATERIALS AND METHODS

Selection of Plant Species: Ten plant species (Flowering plants) were randomly selected from Ambo University Campus for the present study. A stem of having dimensions of nearly 1 inch long, ½ inch girth excised from each plant, and bark was removed from them. Care was taken so that the xylem vessels are not dried until the commencement of experiment.

Construction of The Xylem Filter: 1 inch-long wood section were cut from the branch of selected plant species with approximately 1 cm diameter. The bark and cambium were peeled off, and the piece was inserted into the end of 3/8 inch internal diameter PVC tubing, sealed with 5 Minute Epoxy, secured with hose clamps, and allowed to cure for ten minutes before conducting flow rate experiments. . Care was taken to avoid drying of the filter. The whole set up will be kept ready for further studies.

Collection of Samples: For evaluation of efficiency of selected plant species (xylem) in removing bacteria, solids and color water samples were collected using standard collection methods and brought to the lab and preserved at 4°C until further analysis. Deionized water (Millipore) was used throughout the experiments unless specified otherwise. Saffranine pigment dissolved in deionized water was used for color removal experiments.

Estimation of Total Bacterial Count in the Raw Water: Raw water was collected and brought to the lab for further analysis. The total bacteria/mL in the sample was estimated as per standard microbiological procedures. Proper care was taken to maintain aseptic conditions throughout the study. The whole bacterial analysis was done under the laminar unit.

Initially the number of bacteria/mL was estimated from the raw water. The same water was filtered through prepared xylem filter and bacterial count/mL was estimated after 24hrs incubation. The efficiency was calculated by the difference in bacterial count before and after the filtration and expressed as percent removal. The same procedure was adopted for all the xylem filters prepared from selected plant species.

Estimation of bacterial count: Bacteria/mL = number of colonies developed/petri plate x 18 gives total number of bacteria per mL of water. [1mL of water contains 18 drops]

Estimation of Total Solids in the Raw Water: Raw water was collected and brought to the lab for further analysis. Initially a 500mL glass beaker was taken and its weight was measured and recorded as W1gm. A 100mL water sample was taken into pre-weighed 500mL glass beaker and all the water was completely evaporated on a water bath. After complete evaporation, again weight of glass beaker was recorded as W2gm. The difference in the weight gives amount of solids and expressed as ppm.

$$\text{Total solids} = \frac{W2 - W1}{W1} \times 1000$$

The Efficiency of solids removal was measured as difference in amount of solids before and after filtration using plant xylem.

Studies with Plant Xylem as a Nanopore Filter:

Bacterial Removal Efficiency Studies: Initially before the experiment, plant xylem in the tube was flushed with 5mL of distilled water to make the xylem damp. A 5 mL of raw water was slowly released into the tube over the plant xylem using aseptic pipette. The filtered water was collected from the bottom of the tube and the filtrate was immediately analyzed for total bacterial count under the lamina hood using method mentioned aforesaid. Whole experiment was conducted in complete aseptic conditions using laminar hood. Same method was repeated with all the plant xylems. Results of the experiments were analyzed and tabulated.

Total Solids Removal Efficiency Studies: Initially before the experiment, plant xylem in the tube was flushed with 5mL of distilled water to make the xylem damp. A 5 mL of raw water was slowly released into the tube over the plant xylem using aseptic pipette. The filtered water was collected from the bottom of the tube and the filtrate was for total solids using method mentioned aforesaid. Results of the experiments were analyzed and tabulated. Same procedure will be repeated with all the selected plant species.

Color Removal Efficiency Studies: Initially before the experiment, plant xylem in the tube was flushed with 5mL of distilled water to make the xylem damp. Before the experiment, a colored dye (saffranine) was dissolved in distilled water and was slowly released into the tube over the plant xylem using aseptic pipette. The filtered water was collected from the bottom of the tube and the filtrate was analyzed for color removal efficiency. Since there are no any direct devices available for estimating color intensity a visual perception method was employed for estimating color removal efficiencies. Color removal efficiency was classified into Excellent, Good and bad based on the visual interpretation. Results of the experiments were tabulated according to it. Same procedure was repeated with all the selected plant xylems.

RESULTS & DISCUSSIONS

Results of the present study were represented in the following tables. **Table-1** shows the name of the plant species selected for the study. All the plant species were randomly selected from the Ambo University campus. All the species were identified by their both local and scientific name. All the plant species identified as angiosperms.

Table-1: Name of the Local Plant Species Selected

S. No	Name of the Plant Species [Local Name]	Scientific Name	Family	F/NF
1	Gaatiraa	Juniperus procera	Cupressaceae	F
2	Buna	Coffee Arabica	Rubiaceae	F
3	Wadesa	Cordia Africana	Boraginaceae	F
4	Birbirs	Podocarpus falcatus	Podocarpaceae	F
5	Hodheesa	Germinalia brownie	Combretaceae	F

Key: F: Flowering plant; NF: Non- flowering plant

Bacterial removal efficiency of selected plant species with wet xylem condition were depicted in the **Table-2** and results have shown that all the selected plant species were capable of removing bacteria with different degree of efficiencies. Out of ten species, both Podocarpus falcatus (Birbirs) and Juniperus procera showed highest percent removal of bacteria at the tune of 84 and 82% respectively. The least percent removal was observed with Cordia Africana species (62%). Rest of the plant species showed moderate efficiencies of bacterial removal.

The varying degrees of bacterial removal efficiencies shown by the selected plant species may be attributed to the pore size of xylem vessels. The xylem filter could effectively filter out bacteria from water with rejection exceeding 80%. Pit membranes were identified as the functional unit where actual filtration of the bacteria occurred. The studies conducted by Karnik *et al.*⁶, using pine trees achieved bacterial removal efficiencies of almost 99% and these high removal efficiencies were attributed to the nano-pore size of xylem vessels.

Table-2: Bacterial removal efficiency of selected plant species (with wet xylem)

S. No	Name of the Plant Species [Local Name]	Scientific Name	Initial count (No. of bacteria/mL)	% Removal (Final count)
1	Gaatiraa	Juniperus procera	1080	82(195)
2	Buna	Coffee Arabica	1080	73(292)
3	Wadesa	Cordia Africana	1080	62(411)
4	Birbirsa	Podocarpus falcatus	1080	84(173)
5	Hodheesa	Germinalia brownie	1080	81(206)

Table-3 represents the solid removal efficiencies of selected plant species. Juniperus procera has exhibited highest percent removal of solids (94%) followed by Cordia africana with a percent removal of 93. Least percent removal was observed with Podocarpus falcatus (86%) and moderate removal efficiencies were observed with other species.

Table-3: Solids removal efficiency of selected plant species (with wet xylem)

S. No	Name of the Plant Species [Local Name]	Scientific Name	Initial TS (ppm)	% Removal (Final TS)
1	Gaatiraa	Juniperus procera	876.54	94(78.89)
2	Buna	Coffee Arabica	876.54	89(96.42)
3	Wadesa	Cordia Africana	876.54	93(61.36)
4	Birbirsa	Podocarpus falcatus	876.54	86(122.72)
5	Hodheesa	Germinalia brownie	876.54	91(78.89)

A saffranine dye was used for the present study and results of color removal efficiency of selected plant species in wet condition were represented in **Table-4**. Mostly these results were based on the visual interpretation. Removal efficiencies of Juniperus procera (Gaatiraa), and Podocarpus falcatus (Birbirsa) were classified as Excellent. Cordia Africana (Wadesa) exhibited very poor abilities in removing color. Other species were considered as 'good' in color removal capabilities. The varying degrees of color removal efficiencies can be attributed to the pore size and size of dye particles used for the experiment.

Table-4: Color removal efficiency of selected plant species (with wet xylem)

S. No	Name of the Plant Species [Local Name]	Scientific Name	Remarks
1	Gaatiraa	Juniperus procera	E
2	Buna	Coffee Arabica	G
3	Wadesa	Cordia Africana	NC
4	Birbirsa	Podocarpus falcatus	E
5	Hodheesa	Germinalia brownie	G

*Color removal studies are based on visual perception E: Excellent; G: Good; NC: No change

Above results with wet xylem were in excellent agreement with the studies conducted by Karnik *et al.*⁶ using pine tree xylem. In pine tree studies, they have reported higher percentage removal of bacteria and solids removal when compared to the present study. This variation can be attributed to the factors like length of xylem vessel, the pore size of xylem vessel.

In general, researchers have identified that when compared to flowering plants, the non- flowering plants will have shorter xylem lengths and larger pores which imparts higher filtration abilities. The same trend was observed in the present study also.

Results of the studies using plant xylem in absolute Dry condition were represented in **Table-5** through 7. Experiments with dry xylem as indicated in the **Table-5** revealed that the xylems in dry condition when compared to wet condition, exhibited very poor capabilities of bacterial removal and this may be attributed to the complete drying of xylem vessels which obstructs the free flow of materials through it. The removal efficiencies of dry xylems were almost 60-70% less than that of wet xylems.

Table-5: Bacterial removal efficiency of selected plant species (with dry xylem)

S. No	Name of the Plant Species [Local Name]	Scientific Name	Initial count (No. of bacteria/mL)	% Removal (Final count)
1	Gaatiraa	Juniperus procera	1008	23(776)
2	Buna	Coffee Arabica	1008	26(745)
3	Wadesa	Cordia Africana	1008	19(816)
4	Birbirs	Podocarpus falcatus	1008	27(735)
5	Hodheesa	Germinalia brownie	1008	16(846)

Results of solid removal efficiencies were shown in **Table-6**. The same trend was observed as with the bacterial studies. The removal efficiencies were in the range of 22 to 33%. The low efficiency removal can be attributed to damage of the xylem vessels due to complete dehydration.

Table-6: Solids removal efficiency of selected plant species (with dry xylem)

S. No.	Name of the Plant Species [Local Name]	Scientific Name	Initial TS (ppm)	% Removal (Final TS)
1	Gaatiraa	Juniperus procera	876.54	31(736.28)
2	Buna	Coffee Arabica	876.54	28(631.11)
3	Wadesa	Cordia Africana	876.54	33(587.29)
4	Birbirs	Podocarpus falcatus	876.54	31(604.82)
5	Hodheesa	Germinalia brownie	876.54	22(683.71)

Table-7 indicates the results of color removal efficiency of selected plant species in dry condition. Only Germinalia brownie could remove color and other species, failed in removing the color. This may be attributed to the closure of the xylem vessels which in turn could not filter the dye particles responsible for the color.

Table-7: Color removal efficiency of selected plant species (with dry xylem)

S. No	Name of the Plant Species [Local Name]	Scientific Name	Remarks
1	Gaatiraa	Juniperus procera	NC
2	Buna	Coffee Arabica	NC
3	Wadesa	Cordia Africana	NC
4	Birbirsaa	Podocarpus falcatus	NC
5	Hodheesa	Germinalia brownie	G

*Color removal studies are based on visual perception E: Excellent; G: Good; NC: No change

Results of filtration rates of selected plant species were represented in the **Table-8** and results have revealed appreciable rates of filtration. Filtration rates ranges from a minimum of 4L/D to a maximum of 7L/D. Highest filtration was observed with Juniperus procera (Gaatiraa) and lowest filtration was associated with Cordia Africana. Other species have shown moderate rate of filtration. The hierarchical arrangement of the membranes in the xylem tissue effectively amplifies the available membrane area for filtration, providing high flow rates. Flow rates of about 4 L/d were obtained through filter which is sufficient to meet the drinking water needs of one person. The above results on filtration rates were in agreement with the studies conducted on pines by Karnik *et al.*⁶.

Table-8: FITRATION RATES OF SELECTED PLANT SPECIES

S. No	Name of the Plant Species [Local Name]	Scientific Name	Filtration Rate (Min/50mL)	Filtration Rate (L/Day)
1	Gaatiraa	Juniperus procera	10	7L/D
2	Buna	Coffee Arabica	14	5.2L/D
3	Wadesa	Cordia Africana	18	4L/D
4	Birbirsaa	Podocarpus falcatus	16	4.5L/D
5	Hodheesa	Germinalia brownie	12	6L/D

CONCLUSIONS

Results of the present study revealed that plant xylem can be used as an effective filter for drinking water treatment. All the plant species used in the study have shown varying degrees of filtration in terms of bacterial, total solids, and color removal. Filtration studies carried in both dry and wet xylem conditions. In all the studies, xylem in wet condition proved to be highly efficient in removing bacteria, solids and color than dry condition. Bacterial removal efficiency of selected plant species with wet xylem condition have shown that all the selected plant species were capable of removing bacteria with different degree of efficiencies. Out of five species, both Podocarpus falcatus (Birbirsaa) and Juniperus procera showed highest percent removal of bacteria at the tune of 84 and 82% respectively. The least percent removal was observed with Cordia Africana species (62%). Rest of the plant species showed moderate efficiencies of bacterial removal.

Solid removal efficiencies of selected plant species have revealed that Juniperus procera has exhibited highest percent removal of solids (94%) followed by Cordia africana with a percent removal of 93. Least percent removal was observed with Podocarpus falcatus (86%) and moderate removal efficiencies were observed with other species.

Color removal efficiencies of *Juniperus procera* (Gaatiraa) and *Podocarpus falcatus* (Birbirsa) were classified as Excellent. *Cordia Africana* (Wadesa) exhibited very poor abilities in removing color. Other species were considered as 'good' in color removal capabilities. The varying degrees of color removal efficiencies can be attributed to the pore size and size of dye particles used for the experiment.

Results of filtration rates of selected plant species have revealed appreciable rates of filtration. Filtration rates ranges from a minimum of 4L/D to a maximum of 7L/D. Highest filtration was observed with *Juniperus procera* (Gaatiraa) and lowest filtration was associated with *Cordia Africana*. Other species have shown moderate rate of filtration. The hierarchical arrangement of the membranes in the xylem tissue effectively amplifies the available membrane area for filtration, providing high flow rates. Flow rates of about 4 L/d were obtained through filter which is sufficient to meet the drinking water needs of one person.

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