Efficiency studies with *Dolichos lablab* and solar disinfection for treating turbid waters

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Abstract

The aim of this study was to investigate the combined applicability of natural coagulant and solar disinfection for turbid water clarification and inactivation of bacteria. The coagulation ability of *Dolichos lablab* (Hyacinth Bean) extract was assessed by the use of standard jar test measurements in water with various turbidities. Investigation of *Dolichos lablab* as a natural coagulant was confirmed by its positive effective coagulation activity. An optimum dose of 200 mg of this coagulant resulted in 68% coagulation activity for clarification of water along with inactivation of bacteria in 60 min. Further clarified water with natural coagulant was exposed to sunlight, which showed 100% inactivation of both *Escherichia coli* and coliform counts within 2 h, with no subsequent reactivation of growth after 24 h. The disinfected water complied with prescribed World Health Organization guidelines for domestic use, in terms of bacteriological quality.

Keywords: Turbidity; natural coagulant; solar disinfection; *Dolichos lablab*; *E. coli*.

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1. Introduction

Contaminated water is a problem in many parts of the world. In many places, people have to use water from rivers, lakes, or rain, which they collect in containers. This water is often full of disease-creating microorganisms. The diseases are spread when people use the water for cleaning food or drinking. Waterborne diseases contribute to the death of about four million children in developing countries annually [1]. Access to water is a basic human necessity unavailable to hundreds of millions of the world's poorest people. The search for water and the effects of unsafe water and poor sanitation are enormous burdens on their daily lives, especially in the most rural communities [2].

The lack of universal access to health, education, and water services for the world's poorest people is a big obstacle to the global targets for sustainable development set by the Earth's Summit through Agenda 21. The high cost of water treatment makes potable water expensive. This situation is stigmatizing, in that, on average, most people in developing countries cannot boast 25 liters of clean water a day. Poor town planning, overpopulation, overcrowding, septic tanks located close to wells, and refuse dumps are all potential health risks [3].

Aluminum salts are the most widely used coagulants in water and wastewater treatment throughout the world [4]. However, several studies have raised concern about introducing aluminum into environment. Ferric salts and synthetic polymers have been used as an alternative but with limited success, because their influence on living organisms is not fully understood. Some studies have reported that aluminum remains in the water after coagulation, and may induce Alzheimer's disease [5]. It has also been reported that monomers of some synthetic organic polymers, such as acrylamide, are neurotoxic and have strong carcinogenic properties. In addition, many developing countries cannot afford the cost of imported chemicals for water and wastewater treatment.

On the other hand, naturally-occurring coagulants are biodegradable and are presumed safe for human health. Some studies on natural coagulants have been carried out, with various natural coagulants produced or extracted from microorganisms, animals, or plants. The use of natural materials of plant origin to clarify turbid raw waters is not new. Natural coagulants have been used to treat water for domestic household use for centuries in rural areas. Interest in the use of natural coagulants has increased over time, especially to reduce water and wastewater treatment problems in developing countries to avoid health risks [6].

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The aim of this study was to use natural treatment methods to obtain potable water, safe for human consumption. Plant materials and solar radiation were used as alternatives to conventional chemical treatment methods for turbidity removal and sterilization of water by inactivation of bacteria, respectively.

2. Materials and Methods

2.1. Preparation of raw water

A stock of synthetic turbid water samples from Pragathi Nagar Lake, Kukatpally, Hyderabad was prepared by suspending 10 g of clay in 1 liter of tap water. The suspension was stirred for 30 min and left to stand for 24 h to hydrate particles. The desired turbidities of 20 nephelometric turbidity units (NTUs) (low), 40 NTUs (medium), and 80 NTUs (high) were prepared by mixing a fraction of decanted clay suspension with tap water. The pH of the water samples was kept constant at 7.3 using 0.1 M HCl.

2.2. Plant material

The Hyacinth Bean (*Dolichos lablab*), also called Indian Bean, is a species of bean in the family Fabaceae that is widespread as a food crop throughout the tropics, especially in Africa, India, and Indonesia. The seeds of *Dolichos lablab* were obtained in pods and only seeds from dry pods were used. Seeds were separated from the pods and then pulverized using a clean pestle and mortar (shown in Figure 1).

Figure 1. Fine powder of *Dolichos lablab* seeds.

2.3. Coagulant solution preparation

The pulverized seed material of about 200 mg (optimum) was made into a paste using a small amount of water mixed into 50 ml of clean water, and shaken for 1 min to activate the coagulant properties of the seed to form a solution [7]. The solution was filtered through a muslin cloth into the 500 ml of turbid water to be treated.

2.4. Solar exposure time

Polyethylene terephthalate (PET) bottles made of transparent, clear plastic, cylinder shape with a surface area of 23 x 7.2 x 6.0 cm were used for experimental purposes, as they are good transmitters of light in the ultraviolet (UV) and visible range of the solar spectrum. The total exposure time of experiments varied from 2 to 8 h. Sunlight is strongest from 10 a.m. to 2 p.m., so initial experiments were conducted to encompass this time bracket by up to 1.5 h before and up to 3 h after (from 8:30 a.m. to 4:30 p.m.).

2.5. Physico-chemical and microbial analysis

Samples were analyzed for potability before and after treatment using [8] methods. The turbidity of water samples was measured using the turbidity meter (ELICO CL 52D; Elico Ltd., Sanathnagar, Hyderabad, Andhra Pradesh, India; http://www.elicoltd.com). pH was determined using a pH strip. The *Escherichia coli* (*E. coli*) and coliform bacteria counts were enumerated on eosin methylene blue and MacConkey and were performed hourly during the experimental period. The inverted Petri dishes were incubated for 24 h at 37 ^{o}C. Colonies with a gold metallic sheen on the eosin methylene blue were considered to be positive for *E. coli* growth and white colonies on the MacConkey agar as positive for coliform growth. All results were reported as log CFU (coliform units)/100 ml.

2.6. Methodology

The dried seeds of *Dolichos lablab* were pulverized into a fine powder. Preliminary coagulation experiments were conducted at rapid and slow mixing speeds of 100 and 40 rpm for durations of 1 and 10 min, respectively. Different coagulant dosages were added into beakers during rapid mixing. After various settling intervals, samples were drawn at 5 cm from the surface and residual turbidity was measured using nephlometer for optimum dose and settling time. The clarified water with natural coagulant was then poured into plastic PET bottles and placed on a black cloth in a sunny place, in order to absorb more sunlight. Transparent bottles containing samples placed indoors served as controls. After various exposure hours of solar disinfection, the bottles were put in a cool place. The water samples were analyzed for the water's potability before and after combined treatment (natural coagulant treatment followed by solar disinfection). All of the experiments were performed in duplicate and the average values were presented.

3. Results and Discussion

From the visual observation results, the turbidity of the water samples was high with a terrible odor, which decreased after treatment. Initial turbidities of 20 (low), 40 (medium), and 80 (high) NTUs mainly considerably decreased when the coagulant doses increased. Coagulation was the most effective at a dose of 200 mg/500 ml, when the coagulation activity of the *Dolichos lablab* seed extract was 65, 62, and 68% at a 60-min settling time (shown in Figure 2).

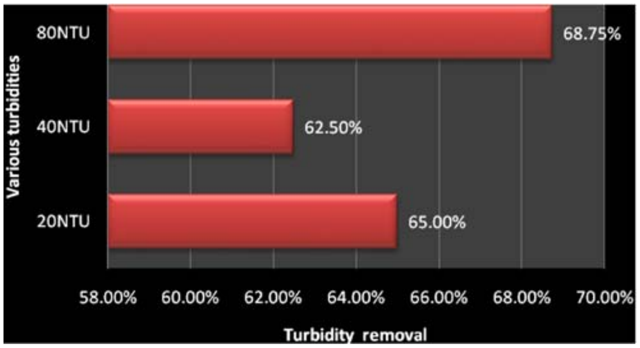

Figure 2. Percentage of turbidity removal at various turbidities with *Dolichos lablab* at dosage of 200 mg/500 ml at settling time of 60 min.

The variance of reduction in different settling times might have occurred due to insufficient time for the reaction to complete (30-min settling time) and due to disassociation of proteins (90-min settling time). The 60-min settling time can be opted as the optimum settling time for treatment. In the water with higher turbidity, coagulation activities were almost independent of the coagulant dose. The coagulation process is usually dependent on a multitude of factors, such as pH value, initial turbidity, temperature, dose, and mixing time. On the basis of the presented results, the extract of *Dolichos lablab* seeds is proper for treatment of water with different initial turbidities, in a dose of 200 mg/500 ml at a 60-min settling time.

Clarified water samples of different NTUs after treatment with a natural coagulant were immediately exposed to solar disinfection. Solar disinfection efficiency in *E. coli* and coliform reduction was determined to 100% at 2 h radiation time for turbidities from low-medium-high (depicted in Figure 3 and 4). After a 24-h lag period, the solar disinfection water showed no growth on the respective inoculated agar plates. This indicates that the bacterial cells were irreversibly damaged or killed by the disinfection process. However, increased concentrations of *E. coli* and coliform were observed in controls after the 48-h lag period.

For the present experiments, normal plastic bottles with 0.1% UV transmittance (at the wavelength of 254 nm) were used. The average temperature was 31 ^{o}C at the beginning of the experiments, and the average water temperature was 48 ^{o}C after a 2-h radiation time. From the data presented above, it can be seen that the disinfection efficiency of the solar process is higher when turbidity is <30 NTUs in raw water where radiation penetrates sufficiently. The degree of disinfection can be inferred from the bacteria counts before and after application of solar radiation. When the number of colonies falls appreciably at 1 h, some amount of disinfection has occurred. When the number of cells is zero, adequate disinfection has occurred.

Our study indicates that clarification of turbid water with natural coagulants from plant material (seeds) combined with use of solar radiation could provide a simple low-cost water treatment method for rural communities in India. Laboratory studies using artificial water samples have shown that *Dolichos lablab* seeds are highly effective in coagulating and removing suspended solids from water containing low-medium-high turbidity.

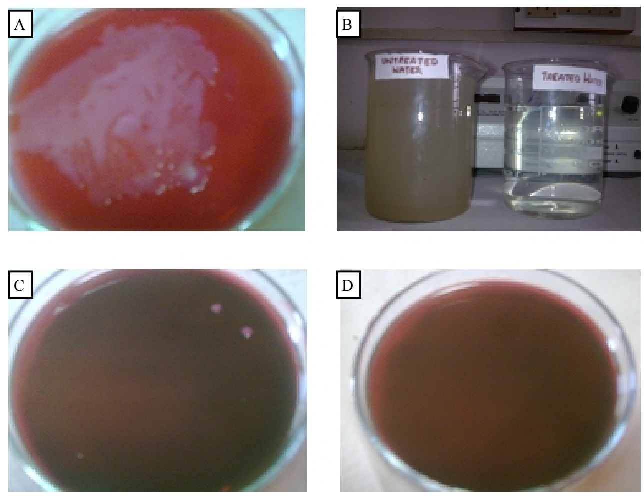

Figure 3. *E. coli* inactivation by water clarified with *Dolichos lablab* and solar disinfection. A) *E. coli* count in turbid water, B) Treatment with natural coagulant (NC), C) *E. coli* count after NC treatment, and D) *E. coli* count after solar disinfection.

At its optimum concentration, *Dolichos lablab* seed powder does not affect the pH or conductivity of the water. Alkalinity and total hardness remained almost constant and were within acceptable levels according to World Health Organization standards for drinking water. Moreover, coagulation of medium to high turbidity water with *Dolichos lablab* seed powder with the finest grain size reduced turbidity further. The best performance of the finest seed powder could be due to its large total surface area, whereby most of the water-soluble proteins are at the solid-liquid interface during the extraction process [9]. This might have increased the concentration of active coagulation polymer in the extract, which improved the coagulation process. The coagulant extract from seeds is believed to have shown antimicrobial activity in the comparative culture test [10].

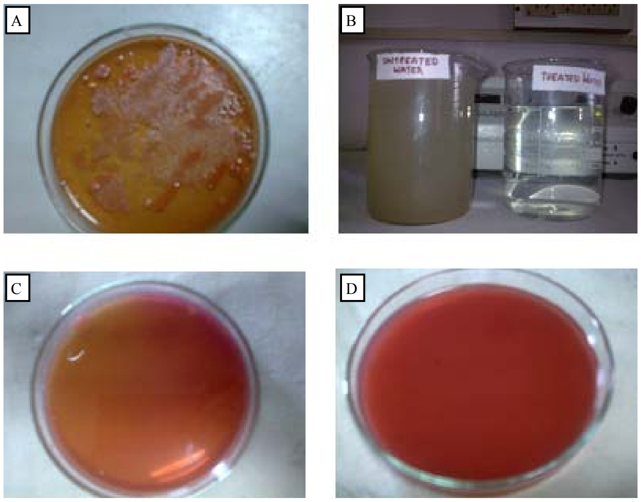

Figure 4. Coliforms inactivation by water clarified with *Dolichos lablab* and solar disinfection. A) Coliform count in turbid water, B) Treatment with natural coagulant (NC), C) Coliform count after NC treatment, and D) Coliform count after solar disinfection.

Dolichos lablab demonstrated the best performance with high turbidity water, in which a turbidity removal efficiency of 68% was observed. The restabilization of destabilized colloidal particles, which was associated with higher residual turbidities, occurred at dosages above the optimal. It is commonly observed that particles are destabilized by small amounts of hydrolyzing metal salts, and that optimum destabilization corresponds with neutralization of particles' charge. Larger amounts of coagulants cause charge reversal so that the particles become positively charged and thus restabilization occurs, which results in elevated turbidity levels [11]. It has also been observed that the reduction in turbidity is associated with significant improvements in bacteriological quality. The effect of natural coagulants on turbidity removal and the antimicrobial properties against microorganisms may render them applicable for simultaneous coagulation and disinfection of water for rural and peri-urban people in developing countries [12].

The advantages of using solar radiation are numerous, no toxic or hazardous by-products are produced, and no smell and/or taste is imparted to the water. It is economical and is easy to apply. The UV component of sunlight is, however, filtered out by ozone. For example, by water droplets and smoke, so that the UV light that actually reaches the earth's surface is restricted to a wavelength range between 295 and 400 nm [13].

For over 4,000 years, sunlight has been used as an effective disinfectant [14]. When organisms are exposed to sunlight, photosensitizers absorb photons of light in the UV-A and early visible wavelength regions of 320 to 450 nm. The photosensitizers react with oxygen molecules to produce highly reactive oxygen species. In turn, these species react with DNA; this leads to strand breakage, which is fatal, and base changes, which result in mutagenic effects such as blocks to replication. The biocide effect of sunlight is due to optical and thermal processes and a strong synergistic effect occurs for water temperatures exceeding 45 ^{o}C [15]. Most of the published investigation to date has been made using *E. coli* as a model microorganism, because it is a very well-known bacteria from all points of view (DNA, metabolism, structure and composition, morphology, behavior under different nutrient media, pathogenicity, types, strains, *etc*.).

The viability of the bacteria *E. coli* depends to a great extent on its temperature of incubation [16]. In general, the chemical composition of water, as well as its content in suspended solid particles, their turbidity, *etc*. affect in a very important way the disinfection processes [17].

This finding is particularly significant as there has been some limited success in the use of natural coagulants alone as a household water disinfectant. It has been shown that neither aluminum nor natural coagulants can yield water free from *E. coli* [18]. The results from the combination of natural treatment methods using plant seeds and solar radiation indicate that this method can effectively clarify and disinfect household drinking water.

4. Conclusion

Access to clean and safe drinking water is difficult in rural areas of India. Water is generally available during the rainy season, but it is muddy and full of sediments. Because of a lack of purifying agents, communities drink water that is no doubt contaminated by sediment and human feces. So the use of natural coagulants that are locally available in combination with solar radiation, which is abundant and inexhaustible, provides a solution to the need for clean and safe drinking water in the rural communities of India. Use of this technology can reduce poverty, decrease excess morbidity and mortality from waterborne diseases, and improve overall quality of life in rural areas.

5. References

[1] WHO/UNICEF (2005). Water for life: making it happen. WHO/UNICEF Joint Monitoring Programme for Water Supply and Sanitation. Accessed 12 April 2010 at http://www.wssinfo.org/resources/documents.html.

[2] Mahvi AH (2007). Feasibility of solar energy in disinfection of drinking water in Iran. *American-Eurasian J Agric Environ Sci* 2 (4): 407 – 410.

[3] Babu R, Chaudhuri M (2005). Home water treatment by direct filtration with natural coagulant. *J Water Health* 3 (1): 27 – 30.

[4] Lye DJ (2002). Health risks associated with consumption of untreated water from household roof catchment systems. *JAWRA* 38 (5): 1301 – 1306.

[5] Sobsey MD (2006). Drinking water and health research: a look to the future in the United States and globally. *J Water Health* 4 (Suppl 1): 17 – 21.

[6] Diaz A, Rincon N, Escorihuela A, Fernandez N, Chacin E, Forster CF (1999). A preliminary evaluation of turbidity removal by natural coagulants indigenous to Venezuela. *Process Biochem* 35 (3-4): 391 – 395.

[7] Muyibi SA, Evison LM (1995). Optimizing physical parameters affecting coagulation of turbid water with *Morninga oleifera* seeds. *Water Res* 29 (12): 2689 – 2695.

[8] APHA (1998). Standard Methods for the Examination of Water and Wastewater. 20th ed., Clesceri LS, Greenberg AE, Eaton AD (Eds.). A joint publication of the American Public Health Association (APHA), the American Water Works Association, and the Water Environment Federation. Washington, DC, USA: APHA.

[9] Gassenschmidt U, Jany KD, Tauscher B, Niebergall H (1995). Isolation and characterization of a flocculating protein from *Moringa oleifera* Lam. *Biochem Biophys Acta* 1243 (3): 477 – 481.

[10] Tandon P, Chhibber S, Reed RH (2005). Inactivation of *Escherichia coli* and coliform bacteria in traditional brass and earthenware water storage vessels. *Ant van Leeuw* 88: 35 – 48.

[11] Ghebremichael KA, Gunaratna KR, Henriksson H, Brumer H, Dalhammar G (2005). A simple purification and activity assay of the coagulant protein from *Moringa oleifera* seed. *Water Res* 39 (11): 2338 – 2344.

[12] Özacar M, Şengil A (2003). Evaluation of tannin biopolymer as a coagulant aid for coagulation of colloidal particles. *Colloids and Surfaces A: Physicochem Eng Aspects* 229 (1-3): 85 – 96.

[13] Reed RH, Mani SK, Meyer V (2000). Solar photo-oxidative disinfection of drinking water: preliminary field observations. *Lett Appl Microbiol* 30 (6): 432 – 436.

[14] Fujioka RS, Yoneyama BS (2002). Sunlight inactivation of human enteric viruses and fecal bacteria. *Water Sci Technol* 46 (11-12): 291 – 295.

[15] Kehoe SC, Joyce TM, Ibrahim P, Gillespie JB, Shahar RA, McGuigan KG (2001). Effect of agitation, turbidity, aluminium foil reflectors and container volume on the inactivation efficiency of bactch-process solar disinfectors. *Water Res* 35 (4): 1061 – 1065.

[16] Berney M, Weilenmann H-U, Simonetti A, Egli T (2006). Efficacy of solar disinfection of *Escherichia coli*, *Shigella flexneri*, *Salmonella* Typhimurium and *Vibrio cholerae*. *J Appl Microbiol* 101 (4): 828 – 836.

[17] Conroy RM, Meegan ME, Joyce T, McGuigan K, Barnes, J (2001). Solar disinfection of drinking water protects against cholera in children under 6 years of age. *Arch Dis Child* 85 (4): 293 – 295.

[18] Kehoe SC, Barer MR, Devlin LO, Mcguigan KG (2004). Batch process solar disinfection is an efficient means of disinfecting drinking water contaminated with *Shigella dysenteriae* type I. *Lett Appl Microbiol* 38 (5): 410 – 414.