Natural Materials for Sustainable Water Pollution Management

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"Once you eliminate the Impossible, whatever remains, no matter how improbable, must be the truth" (Sherlock Holmes. (Sir Arthur Conan, Doyle, 1859 - 1930))

1. Introduction

The world Health Organization has estimated that up to 80% of all diseases and sicknesses in the world are caused by inadequate sanitation, polluted water or unavailability of water. (Cheesbrough, 1984, Pritchard et al., 2009 and Yongabi et al., 2010) Faeces, garbage resulting from improper sewage disposal are an important source of pathogenic organisms in water, especially the causative agents of diarrhoeal and dysentery diseases. Faeces are attractive to flies which support the development of the larval stages (maggots) of filth flies. These hazards couple with the indiscriminate disposal of faeces can also constitute a grave nuisance from the offensive sight and smell. In many parts of rural Africa, toilets and garbage disposal pits and/or sites are cited close to wells. The leachates from these could contaminate ground and surface water (Yongabi et al., 2011) Diseases associated with water could be broadly categorised into five epidemiological groups viz: Waterborne infections e.g cholera, typhoid, infective hepatitis, Water shortage diseases e.g. skin infections, trachoma, Water-impounding diseases e.g schistosomiasis and guinea worm, Water-arthropod disease e.g. malaria onchocerciasis, Chemical constituents either excess or shortage e.g. fluoride - this may have an indirect effect in the body. The World population is currently growing at an unprecedented rate, and as of 1996 a 1.8% growth rate per annum which translates to over 80 million people a year was reported (UN, 1996). There is no doubt that most African countries are presently characterised by inexorable population explosion (Adegbola, 1987 and Yongabi et al., 2010) This has dire consequences to the food and environmental resources, more industries are springing up and the quest for survival is generating a lot of pollution. Inorder to meet the needs of this soaring population, the production capacity in all the sectors would have to be multiplied. All these sectors would need water as a raw material and apparently, there is water shortage in Sub Saharan Africa. For instance, in Nigeria, a lot of volumes of water is used and needed for irrigation especially in the dry season. The potential irrigation area of Nigeria stands at about 2.5 Million hectares which is capable of producing close to 40% of the current total annual crop production. The small scale fadama irrigation constitutes 90% of the country's irrigation potentials (Dada et al., 1990).
This picture is similar across sub Saharan Africa. Apart from agriculture, the health sector, hospitals and pharmaceutical companies in Nigeria uses a lot of water during manufacturing and cooling systems. The waste water generated is high and often discharged untreated. This ultimately pollutes surface water. Such waste water is difficult and expensive to treat and re-use as it contains a mix of chemical compounds. The major industries in Nigeria like food-based industries and breweries and refineries really use huge volumes of water and as such generate so much waste water.

Taking the brewing industry as a good first point of call, only 8% of the nutrients in the spent grain are used. The other 92% is waste and usually discharged into the environment. Imagine 18.5 hl of wastewater (alkaline is leached out to the environment to produce a bottle of beer (Ihl) with 1.2kg of BOD, equally, the water input is high. (20hl) Many breweries across sub saharan africa and the world over function along similar lines (Pauli, 1998) These sectors depend heavily on water which is scare and generates a lot of wastewater that is at best untreated and goes along way to generate ecological imbalances especially in the face of population explosion. As consequence of population explosion is heavy dependence on fresh water resources, fresh water gradually becoming impoverished in many parts of the world through a number of means; contamination, reclamation and exhaustion. For instance, in Jordan and Yemen 30% of their water from their ground water aquifers are depleted per annum than the aquifers are able to recharge (Engle/Manand eroy, 1993). This trend may be similar across many countries the world over.

1.1 Statement of problem / justification

Estimates reveal that water borne diseases contribute to the death of 4 million children in the developing countries each year. This estimate by UNICEF may be a far under estimation of the real situation on ground. In many African countries for instance, water is scarce in both the rural and urban settings. A local survey carried out in the states of Bauchi, Plateau and Benue states shows that most of the rural communities lack potable water and has to travel many miles to search for water in nearby polluted streams for domestic uses. (Table I) At Agakwe, in Tiv Land, Edoma land in Benue state of Nigeria, pipe borne water was found (Survey done by researchers with CARUDEP, JOS, 2005) while in Bauchi, an all the local Governments in the rural areas of Tafawa Balewa. Ganjuwa etc depend on wells that dry up in the dry season. The same holds true for many communities in the northern parts of Cameroon, Chad, Sudan, Central African Republic and Niger. The communities lack basic hygiene, sanitation and water (Table 2.) UNICEF (1993, 2009) acknowledged that 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 ( UNEP, 2002) Unfortunately, this obstacle remains and it is uncertain if the strategies on ground can generate sustainability in any way. This is because, poor people in semi-urban and some rural communities in subsharan africa still pay a disproportionate share of their meagre incomes for water services that is irregular, inconvenient and often suspicious in quality. A survey done in some villages in Cameroon shows just how potable water is rare. (Table1). Paradoxically, so much attention, the world over has been placed on water pollution and sanitation programmes with huge expenditures but the impact, however, remains questionable. Perhaps to attempt to explain this short falls could be that most of the strategies used to solve these problems are in themselves not sustainable.
**Table 1.** Baseline survey on water and sanitation facilities from communities in Bauchi state, Nigeria (The survey indicates limited safe water and sanitation among the rural area in Bauchi-Nigeria)

| Community Name | No. of adult male | No. of adult women | Children 1-12 | Youth 12-25 | No. Latrines | No. houses | Water point |
|----------------|-------------------|--------------------|---------------|-------------|--------------|------------|-------------|
| GarinAbare     | 400               | 800                | 1500          | 700         | 120          | 300        | 5           |
| GIKAR          | 1000              | 2250               | 4000          | 700         | 21           | 250        | 3           |
| KWABLANG       | 250               | 350                | 1000          |             | 32           | 100        | 19          |
| Barkaya        | 500               | 600                | 3500          | 1500        | 32           | 100        | 6           |
| DUKKUN Dindima | 25                | 36                 | 59            | 18          | 32           | 13         | 19          |
| Turiya         | 25                | 38                 | 88            | 15          | 15           | 11         | 9           |
| Nassarawa      | 35                | 42                 | 81            | 19          | 26           | 29         | 5           |
| Dindima        | 25                | 36                 | 56            | 19          | 38           | 13         | 2           |
| Fumbinare      | 42                | 59                 | 102           | 54          | 49           | 12         | 5           |
| G/Total        | 2302              | 4211               | 10386         | 1625        | 365          | 828        | 73          |

*Done in collaboration with Development Exchange Centre (DEC), an NGO which provides / assist these communities with development projects in 2005*

Table 2. Frequency distribution of common illnesses found in Bulli Village, 10km away from the university Community in Bauchi, Nigeria. (The results in this table indicates high frequency of water borne diseases in the study area)

| Illnesses            | Adults No% | Children No% |
|----------------------|------------|--------------|
| Scabies              | 24 (2.9%)  | 32 (3.8%)    |
| Skin sepsis          | 16 (1.9%)  | 20 (2.4%)    |
| Yaws                 | 4 (0.5%)   | 10 (1.2%)    |
| Lice                 | 152 (18.3%)| 250 (30.0%)  |
| Trachoma             | 16 (1.9%)  | 18 (2.2%)    |
| Conjunctivitis       | 65 (7.8%)  | 133 (16.0%)  |
| Bacillary dysentery  | 106 (12.7%)| 198 (23.8%)  |
| Salmonellosis        | 16 (1.9%)  | 28 (3.4%)    |
| Diarrhoea            | 14 (1.7%)  | 108 (13.0%)  |
| Ascariasis           | 10 (1.2%)  | 18 (2.2%)    |
| Paratyphoid Fever    | 20 (2.4%)  | 34 (4.1%)    |
| Worms                | 4 (0.5%)   | 24 (2.9%)    |
| Stomach ache         | -          | 103 (2.9%)   |
| Malaria              | 70 (8.4%)  | 6 (0.7%)     |
|                       | 517 (53.7%)| 986 (117.4%) |

Providing pipe borne water to communities, is laudable but when the low income earning communities cannot cope with maintenance cost and the robustness of the technologies in place, then it becomes a major problem. UNICEF (1993) reported that in the 1980s, some 10 million dollars was spent yearly in the developing countries on high technology to improve services to people who already had water and sanitation predominantly in the cities. Only a fraction (20%) was reluctantly spared on low-cost appropriate technology.
for the underserved majority of people in peri-urban areas (UNICEF, 1993). The high cost of treating water and its attending high energy input is prohibitive to most industries and factories in developing countries and as such release untreated wastewater into neighbouring streams, thereby polluting many fresh water bodies. These sources of water pollution includes heavy metals, halogenated hydrocarbons, dioxins, organochlorines such as DDT which do not easily break down under natural processes and tend to accumulate in biological food chain. The popular treatment system of water in sub saharan africa is the sedimentation, coagulation, disinfection (chlorination), filtration. Undoubtedly, this has generated potable water but, however, the final water products remain unaffordable by 70% of their populace (Schultz et al., 1983; Yongabi et al., 2010) Reports also suggest chlorine resistant organisms such as, cryptosporidium oocysts, strains of salmonella sp, aeromonas, entamoeba cyst, mycobacterium sp, escherichia Coli. 0157:47 and host of others (Madore et al., 1987; Yongabi et al., 2011). Chlorine has been noted as a potential carcinogen forming compounds such as tetrachloromethane (TCM) which also produces hormonal analogue that may interfere with male fertility. Aluminium sulphate (Alum), the widely used water coagulant thus generate acidic water, unsafe for pregnant women and causes predementia in some people (loss of memory) While all these defects exists, mankind has been endowed with indigenous knowledge and has been using it to survive preceding the advent of all these technologies. There is a need to revisit our roots, study this indigenous system and improve on them. Exploring and exploiting the potentials of natural materials such as plants and sand to bring about cheap clean water in a more ecological friendly manner are the thrust of this work. This may have great lessons for ecological sustainability now and centuries to come.

1.2 Aims / objectives
The ultimate purpose of this chapter is to report results of our research on a water pollution management technology that is low-tech, cheap and above all ecologically friendly. The specific objectives of this study, therefore, are:to report the results of analysis of the pathogen level of polluted water from refinery, food and confectionery processing industry in Nigeria and Cameroon, stagnant pond water where people fetch water for household chores and for irrigation at. To carry out a survey / inventory on problems of clean water and indigenous knowledge on how communities treat their water in Nigeria and Cameroon. To use the collected knowledge and screen these plant materials and their extract for their coagulation/disinfection activities in vitro using polluted water samples. To test their potential antimicrobial activity on isolates from polluted water samples and, to generate clean water using a constructed integrated biocoagulant - sand filter system and other geological-materials.

2. Brief overview of interdisciplinary importance, dangers of water and existing gaps in water pollution management
2.1 The necessity of water as a consumable product in all the aspects of life
The role of water in life as a whole cannot be over emphasised as this universal solvent is the basis of life after air. What a life without water? Evolutionary, biologists hold strongly that life began in water and therefore explains why human use water at times for rituals. Water is a prime necessity for life, it forms the basis for a balanced diet without which digestion cannot function well. It is a lubricant for biological processes such as
excretion and major glands secretion are usually in water form. It acts as a cushion preventing crushing in internal structures, example synovial fluid.

To the agriculturalist, crops cannot grow without water, therefore, it is needed for germination, that is probably why in dry areas irrigation is used to shunt this adversity. Water is used for laundry, domestication such as cooking and washing of utensils. This vital community is the basis for electricity in which case more important than electricity. It is the source for hydroelectricity which is the backbone of all industries and factories. Additionally, water is used in agro-industry for washing, media for dissolution, production of dairy products, beverages etc. To the engineer, water is used as a cooling agent, lubricant and for building and construction. In addition, water is a useful transport source: Navigation. Water is a habitat for fish and minerals such as petroleum. Fish being used as sources of protein for man and petroleum and other minerals used as fuels.

Water serves as a touristic site, for example the Kribi beach in Cameroon and the Gubi dam in Bauchi state. In the wise, a source of revenue for the Government. Indeed, highlighting the use of water could only tantamount to an infinite list. Taking water as previously mentioned is a source of a balanced diet, it contains vital minerals such as Manessium, Ca, Fe, Cu, Zu, F, No₃, So₄ etc) for the International Standard for drinking water (WHO, 1984). Therefore, good water should actually possess these minerals. Good water should be colourless, odourless and free from any toxic elements. Some toxic substances in drinking water could include Pb, Se, As, Cr and CN. According to World Health Organization, 1958, showed that a 0.001 4 p.m is the maximum concentrations allowable, exceeding this level is pollution (see As cited above, toxic elements could be consumed from water that could lead to cancer.

Water, even though has many uses serves as a breeding ground for some vectors of man's parasitic diseases, for example. Malaria and schistosomiasis. Rain water in excess could cause flood and hence heavy economic losses. Besides, this could also lead to erosion which inflicts heavy pains to Agriculture. Finally, sea accidents lead to loss of lives too. The overwhelming indispensability of water as a primordial stuff for all the arms of Economy has become a hot topic for discussion by many state governors and their administrations in Africa. If the State governments are not supplying boreholes and other rural water Sources, she is either trying to solve flood problems or some other hazard cause by rain storm. From analysis of all budgets speeches since 1982 to date, it is interesting to note that water has been placed as a top priority amongst other projects yet little is achieved. Water which is safe for drinking must be free of pathogenic organisms, toxic substances and an excess of minerals and organic debris. It must be colourless, tasteless and odourless in order to be attractive to consumers and preferably cool. Water is the basis of life. About 75% of the body weight is made of water. In developing countries 15 million infants die every year due to contaminated drinking water, poor hygiene and malnutrition. About 80% of illness in developing countries are directly connected with contaminated drinking water (WHO). The Provision of water supply near by for consumers and sufficient for their daily needs will help greatly in decreasing the incidence of skin diseases and eye infections and also reduce diarrhea diseases and most worm infections, particularly if the water is of good quality bacteriological. However, major improvements in health conditions through provision of sufficient safe water can only be achieved through domestic hygiene and proper methods of water purification (Yongabi et al., 2010). Oyawaye et al (2000) in their study of water sources for three years in Bauchi, noted elevated levels of nitrates (33.3mgkg) in ground water.
sources in the dry season. Higher nitrate values for treated and untreated waters still remained high in the rainy season but within acceptable limits. Excess nitrates in water has been linked to methemoglobinemia (blue babies). Many infant deaths in Africa and particularly sub Saharan Africa are mainly attributed to dysentery and diarrhoea of undefined sources which may be due to nitrates in water. Similarly, literature elsewhere has evidence that implicates N-Nitrosamines in the incidence of carcinogenesis. The density of microbial isolates has been reported to be inversely proportional to the level of residual chlorine from 1.0mg/L to less than 0.2mg/L. Residual chlorine also reduces steadily from point of application to point of collection. Twenty three bacterial genera belonging to groups of coliform, faecal coliform and Staphylococcus spp were isolated at various stages (Yongabi et al., 2011).

2.2 General methods of water pollution management

2.2.1 Application of chlorine, halogens and alum in water treatment

Chlorine is widely applied to disinfect water. For instance, a well or a tank containing 1000 litres of relatively clean water 2g of chlorine is added and if organic matter is present or one is doubting the purity you add 4g of chlorine. Then thoroughly mix it into the water and allow standing for at least 30 minutes before using it. However if water is highly turbid i.e containing a lot of sediments, alum is first added to make the sediments settle at the base. The water is then drain into another tank before chlorinating. The amount of Alum required treating 1000 litres of relatively clean water is 56g while for sufficient safe water for a community but then it requires highly skilled technicians who can measure and control the chlorine and alum dosage. This knowledge is lacking in the rural areas. Other halogens such as bromine and iodine are also applied in water treatment. The set backs have been discussed in recent publications (Yongabi et al., 2010 and Yongabi et al., 2011).

2.2.2 Sand filter

This method of purifying water has been known right from time immemorial. Over thousands of years now clean water have been obtained from river beds when dug. As water falls or flows over the river bed it percolates through the sand grains where the disease-causing organism filter out. Clean Sharp River sand is obtained and thoroughly washed; gravels are also obtained and washed. Tow clean containers are used for the construction of the sand filter. The container for the filter and storage could be made out of metal plastic or traditional clay. A hole is made two-thirds of the way up the filter container and hose with blocked base and perforated will be fixed at the opening into the drum. The gravel is then placed over it to a height of 7.5cm and the sand is placed above it to a height just below the hose fitting. The filter is then thoroughly flushed out with clean water for a week to allow for formation of biofilm. The attributes and set backs of the sand filter in terms of cost, installation, management and efficacy and the need to intergrate it with plant coagulants have been reported (Yongabi et al., 2010).

2.2.3 Water treatment with plants: The case of moringa oleifera and water hyacinth

The seed pods of Moringa oleifera have been used for water treatment. After shelving, the seeds are crushed, seized (3.5mm mesh) using traditional techniques employed in the production of maize flour. Approximately 50-150mg of the ground seed will be needed to
treat a litre of river water, depending on the quantity of suspended matter. Normally, a small amount of clean water is then mixed with the crushed seed to form a paste. The crushed seed powder when added to water, yields water soluble proteins that possess a net positive charge. The coagulant/flocculant characteristics of seed is linked to a series of low molecular weight cationic protein. Dose of Moringa oleifera seeds depends much on the turbidity of the water in question. Generally, 75-25mg/l (0.75 - 2.5g) has been employed. For a turbidity of 400NTU, 0.5g of Moringa oleifera powder is used for a litre of turbid water. Extensive studies have been done on the applications of Moringa oleifera in water treatment. Other plants used in water treatment include, cactus, water hyacinth and syntrochnus potatorum which are reported to remove turbidity and heavy metals from water (Bina, 1991; Yongabi et al., 2010). The need to catalogue such useful natural materials in Africa needs intensification.

3. Materials and methods

3.1 Study area
The study was conducted in Bauchi State (at Abubakar Tafawa Balewa University) Nigeria and Bamenda, Cameroon 2010 to 2011. These two countries are located in sub shaharan Africa with the same climatic conditions and similar traditions and problems. These Natural plant materials collected from these focused more on other plants rather than Moringa oleifera which has been extensively reported in literature.

3.2.1 Materials used
MacCathney and bijore bottles were purchased from supplies of Hospital and laboratory materials from Bauchi metropolis. They were washed repeatedly using detergent and rinsed in clean water and then sterilised by autoclaving alongside with all glasswares used for the study. Autoclaving was done at 121°C for 15 minutes.
A number of agars: Nutrient agar (Oxoid Ltd), MacConkey Eosine Methylene blue, potato Dextrose agars (Oxoid) Ltd) were obtained from the University Zeri Research laboratory and Phytobiotechnology Research laboratory and School of Chemical engineering, The University of Adelaide, South Australia.

3.2.2 Special equipment and apparatus
Some of the equipment and apparatuses used for the study include spectrophotometer (Phips) Vu/Vis, Pve, unicam sp 6-450 incubator (Jouan) bench. Centrifuge (Mistrail 1000) weighing balance (meter am100) and soxhlet apparatus (Galler kamp).

3.2.3 Chemicals and reagents
Nutrient Agar were obtained from biotech laboratories survey, UK, Ferric chloride, potassium hydroxide, copper acetate, lead acetate, bismuth nitrate sodium chloride, chloroform, diethylether, ethanol were purchased from Britiest Drug Houses (BDH). Chemicals Ltd, poole England.
Ammonia solution potassium tartrate picric acid, filing solutions were obtained from Mand B Ltd England.
All other regents an chemicals used were of analytical grade obtained from reputable scientific and chemical companies. All solutions were prepared in distilled water, redistilled from pyrex apparatus.
Carica Papaya Plant
seeds used as a phydisinfectant and coagulant in rural Cameroon

Clay
used as a geocoagulant

Pieces of Alum  Garcinia Kola seeds
Alum is a synthetic coagulant used as a phytocoagulant and phytodisinfectant, widely used, people have to buy. locally available in the communities in Africa
3.2.4 Polluted water sample collection
Refinery wastewater was collected from other Kaduna Petroleum oil refinery. This is located in Kaduna Town in Kaduna State in the northern parts of Nigeria and SONARA oil in Cameroon. The crude oil is fractionated and fuel produced amongst other, bye products. The refinery wastewater is usually discharged untreated into river Kaduna. Ten litres of the wastewater was collected with the assistance of students undertaking their internship at the refinery in 2005-2011. Wastewater from the NASCO Company Ltd in Jos, plateau State of Nigeria was also collected. The NASCO household in Jos produces a number of confectioneries including, biscuits, conflakes etc and then Nasco soaps, detergents etc. Jos is located in the North Central region of Nigeria and has a teeming population. The wastewater is usually discharged into the neighbouring streams and brooks. Ten litres of the wastewater was collected with the acid of students on internship. Lastly, dirty (turbid) pond water was collected from a stagnant pond located at the western part of the Abubakar Tafawa Balewa University Campus. The stagnant water is used by the local people around for irrigation of Crops within the vicinity, other activities include fishing and washing of clothes as well as at times swimming. Ten litres of the sample was collected for laboratory studies.

3.3 Microbiology analyses
One ml of each of the samples was diluted in 9ml of sterile distilled water and serially diluted up to 10^-5 dilution and plated in triplicates on Nutrient agar for total heterotrophic bacterial counts, MacConkeyagar and Eosine Methylene blue agars for Total Coliform and E. coli counts respectively while on potato dextrose agar for fungal counts. Incubation was done at 37°C for 24 hours for bacterial counts and at 25°C for fungal counts. Discret colonies on each plates were counted on each plate and average of three plates taken. The presence of colonies on Eosine methylene blue agar indicated probable identify for typical coliform colonies, Gram stained portions of the colonies showed gram negative roads with absence of spores as a further elucidation of the Micromorphology of coliform. Colonies on EMB that appeared as Metallic greenish sheen confirmed the presence of Escherichia Coli.

3.3.1 Collection and Identification of plants
The leaves and Abus precatorius were collected from Shere hills, Jos Nigeria. This plant sample was authenticated by plant taxonomists at the Federal College of Forestry Jos,
Nigeria. They were dried and pounded into well labeled, clean air tight. Containers and stored until required.

3.3.2 Sources of test organisms
Clinical isolated of Eseherica colı, staphyococcus aureus, Salmonella paratyphi and Candida albican were isolated from polluted streams in Bamenda, Cameroon.

3.3.3 Methods
Preparation of plants for biological test
The dried and pulverized samples were then extracted using ethanol, diethyl and water. This was done in increasing polarity. Most traditional healers sometimes use palm wine, which contains ethanil as their solvent. This gives additional reason for preferring ethanol to methanol. Deithyl ether is one of the best solvent for antimicrobial activities (nastro et al, 2000). Water is a universal solvent.

3.3.4 Aqueous extraction
10g of powdered sample was weighed on a Mettle balance. It was then put in separate clean and sterile conical flasks containing 200ml of cold sterile.

3.3.5 Biological assay
Preparation of dilutions of the extracts
The concentrations of various crude extracts were made in sterile distilled water and for dichtylether extrac) the concentrations prepared in those solvents were 50ml, 100mg/ml and 150mg/ml.

3.3.6 Purification of bacterial isolates
The stock cultures of the bacterial isolates were subculture unto nutrient agar, blood agar and agar and macconkey agar to produce discrete colonies and incubated for 24 hours at 37°C. The plates were examined for purity and specific biochemical test were carried out to confirm the identity of the different isolates according to methods described by (Baker et al, 1980).

3.3.7 Test for bacterial suspensions
Preparation of fresh plates of the test bacteria was made from isolated stocks stored on agar slants. By the use of a stride wire loop, colonies of fresh cultures were picked and suspended in 20ml of nutrient broth in different sterile universal bottles. The centrifugation bottles were done in MSE refrigerating centrifuge at 1000m rpm for 30 minutes in virology department of N.V.R.I Vom. The supernatant was discarded. The organisms were again resuspended using equal volumes of sterile normal saline. The concentrations of the organism were obtained by comparison with 10 standard opacity bottles (Macfarland's Naphelometry) method of opacity, which contained various amounts of barium sulphate in 1% sulpheric acid (N/36). Most of the tubes corresponded to 10-4 which was very turbid. The organisms were then diluted down to 10-6 s then one loopted equivalent of 0.02ml from each of the bottles 10-4, 10-5 and 10-6 was plated out on three different
petridishes containing nutrient agar and incubator over-right at 37°C to determine population density of the test organism. Member of colony forming units per millitre was obtained as follows, since for example 10^-6 deliration had 25 colonies. Colonies on the second day 25 x 50 x 10^-6 + 1.25 x 10^-9 C.FU/ml. 1ml of the 10-6 dilution of various bacteria was used in flooding nutrient agar plates in the agar diffusion method of invitro sensitivity test.

3.3.8 Preparation of media
2.3g of nutrient agar was dissolved in 100ml of distilled water and heated slowly while shaking until the solution become clear and yellow in colour. The nutrient agar was cool to about 47°C and become semi-solid state. This is to facilitate the diffusion of large molecules of the crude extract as compound or standard or processed and purified antibiotics with small and readily diffusible molecules.

3.4.1 Agar gel diffusion test (punch-hole method)
The plates of nutrient agar were seeded in duplicates with 1.0ml of 10^-6 dilution of the test bacteria. The plates were then swirled to allow the inoculum to spread on the excess was discarded in a disinfectant jar. The plates were allowed on the bench for 5 minute is and they were dired in the incubator for 1 hour at 37°C.
Using a sterile cork borer four well were bored at equal distances around for plate. The 5th well was made in the middle. The bottoms of the wells were sealed with one drop each of sterile nutrient agar before the extracts were puts.
The prepared concentrations the extracts were put into the wells. Sterile distilled water was put in the 5th well to serve as negative control for aqueous and ethanolic extracts while dimethysulfoxide ws used as negative control for diethylether. Gentamycin was used as a positive control in the 4th well. After allowing on the the bench for 1 hour, for diffusion of the extracts, the plates were incubated at 37°C for one day. The plates were examined the next day to concentrations of the extracts on the test bacteria.
The zones of inhibitions were measured using a ruler in millimeters and the average of the two readings was taken to be the zone of inhibition of the bacterial species in a particular concentration.

3.4.2 Minimum Inhibitory Concentration (MIC)
This was determined using broth dilution technique (Puyelde, 1956). Freshly prepared broth in sterile Bijou bottles was used. Two sets of six Bijou bottles were used for each test. 1 ml of sterile nutrient broth was put in Bijou bottle number 1 to 6.1ml 200mg/ml was added to Bijou bottle number one. The extract in the bottle on was therefore diluted 1:2. It was properly mixed and 1ml was transferred to bottle number two which was diluted 1:4 and this was continued until the 5th bottle from which one ml was discarded. Bottle number six contained only sterile nutrient broth to serve as negative control.A loopful of 10^-6 dilution of bacteria suspension with microbial load of 1.25 x 10^9 C.F.U/ml was then added to all the six bottles. This entire procedure was done for all the organisms that were susceptible to the various extracts. The bottles were thoroughly mixed by gentle shaking and incubation for 24 hours at 37°C. The bottles were observed for turbidity after incubation visually by comparing with the control.Cultures from incubated bottles were subcultured onto fresh nutrient agar plates. The inoculated were incubated at 37°C for 24 hours. The plates were
examined for growth indicated bacteriocidal effect of the concentration of the extract used. Plates showing light growth were taken to have bacteriostatic effect, while those showing moderate or heavy growth were taken to have no inhibitory effect on the bacteria (Puyuelde, 1986).

3.4.3 PH analysis
The PH of the raw and treated wastewater samples was tested using a combi-9 test strip (a standard strip for routine urinary biochemical analysis). A fresh strip each was dipped into each of the samples and after sixty seconds, the colour change noticed was compared with a range of colour standards and when the colour of the strip Matched any of the colour standards, the PH label was directly read off.
(Photo field solar weighing balance and Combi-9 Ph strip).

3.4.4 Turbidity evaluation
A subjective visual observation was done. The presence of colloidal suspended matter was noted in the untreated samples while their absence noted in the treated samples Floc formation and lack of floc formation was also observed as a distinct evidence of coagulation for the treated samples. The presence of odour and absences was also noted by suing the nose. The use of the sight and small senses were highly exploited.

3.4.5 Plant sample selection and collection
The plant coagulants used in this study were selected based on a survey of their local use in water purification by the indigenous people in sub-saharan africa (Yongabi K. A, 2004, www.biotech.kth.se/iobb/new/kenneth04.doc) Moringa Oleifera (Lam) seeds have been used by a rural Nigeria for water treatment and Literature elsewhere abound (Fuglie, 1999, Folkland et al, 2000, ) The dried seed of Moringa Oleifera were harvested from Bauchi State, Nigeria. Seeds of Garcinia Kola, Hibiscus sabdariffa and Carica papaya were collected from Enugu in Nigeria and Bamenda, Cameroon (Photo).

3.4.6 Plant processing
The seed pods were harvested and stored in Khaki envelopes, They were deshelled (specifically M Oleifera Garcinia Kola) while the seed of Carica papaya were scoped out from riped fruits as well as Hibiscus sbadafrifa seeds were purchased from the market at Mdulawal Market in Bauchi Metropolis.

3.4.7 Coagulation studies
Graded weight (0.5g to 5g) of the pulverized plant Materials each and Alum, Hydrogen peroxide, were each added to 200mls of each of the wastewater samples in 250ml capacity beakers.
Increased weights in grams from 0.5g to 5.0g of each of the plant material was mixed in a small quantity of turbid water for form a paste and then mixed carefully with the water samples in the beakers.
The same procedure was done for Alum and a turbid water sample in a beaker (200mls. was allowed to stand in a beaker for 24 hours as controls). The Coagulative effects and change in total bacterial counts, PH, visual clarity amongts other parameters were evaluated.
3.5.1 Cold extraction (Buck extraction)
A cold Methanol and aqueous Extraction was then carried out on 50 grams each of Hibiscus sabdarigga seed and *Carica papaya* seed powders except for Moringa Oleifera. 50 grams of each of the powders was steeped in 250mls each of methanol and water for 24 hours. Gravity filtration was carried out using whatman filter paper N° 13 and solvent evaporated at room temperature.

3.5.2 A cold sequential extraction of *Moringa oleifera* and *Garcina kola* seeds
A cold Sequential solvent Extraction was carried out on Moringa oleifera seed powder using n-hexane, Dichloromethane Methanol and water in that order. The purpose of this was to exploit the polarity effect of the solvent on the possible isolation of the active portion from the plant material 50grams of the pulverized seed (pulverised using a pestle and mortar) was steeped in 250ml of n-hexane left for 24hours, filter off using gravity filtration using whatman filter paper No 13, The plant residue was dried in the sun and used for the next solvent and the order maintained for all the other solvents.
The extracts were left in the open for 2 weeks for the solvent to evaporate. The extracts were now used for antibacterial bioassay.

3.5.3 Antibacterial assay (agar diffusion method)
The bacterial isolates were re-cultured in peptone water for 18 hours and 0.3ml of each of the bacterial suspension was mixed aseptically with 15ml nutrient agar (oxoid) in sterile petri plates and allowed to solidify. A stainless steel borer of 6mm diameter was used to punch wells into the agar and each well was filled with 0.1ml of 2% extract, and with oil and of sterile distilled water, H2O2 and Alum as controls.

3.5.4 Phytochemical screening
The phytochemical screening of the powdered extracts obtained from the leaves of Abrus precatorius were carried out using standard qualitative procedures (Trease and Evans 1989, Sofowora 1986).

3.5.5 Test for alkaloids
Two grams of plants materials thoroughly grounded was treated in a test tube with 25ml of 1% Ad for 15min in a water bath. The suspension was filtrated in a test tube and the filtration was divided in two pants A and b.
To filtrate A, five drops of Dragendorff reagent were added. The formation of a precipitate indicated the presence of alkaloids.

3.5.6 Test for flavonoids
i. Well ground plant material (1g) was extracted with water (10ml) and methanol (5ml) and filtered. Few magnesium turnings were added to 3ml of filtrate and concentrated added dropwise (cyanidine reaction). Developments of colour indicate the presence of flavonoids a red colour and flavonones give a pink colour.
ii. To 1ml of the extract 1ml of Naoh was added. The formation of a golden yellow precipitate indicated the presence of flavonoids.
3.5.7 Test for cardiac glycosides (salkowski test)
0.5g of extract was added to 2ml of chloroform and after mixing, 2ml of H2 so were carefully added to from a lower layer. Reddish brown colour at the interface indicates the presence of a steroidal ring i.e. glycogen portion of the cardiac glycoside.

3.5.8 Test for anthraquinones
Anthraquinones are a subset of anthranoids. For the specific test an ether chloroform maceration (1g in 5ml of CHd3 and 5ml of ethered was filtered and 1ml of 10% NaOH solution. A red quinones. A weak coloration was assigned a +, while a strong coloration a +++.

3.5.9 Test for steroids
Powdered plant material (1g) was covered with either and shaken occasionally for 2 hours. The solution was filtered and decanted. 1ml of the solution was put on porcelain plate to evaporate. A drop of conc. H2So4 was added and stirred orange colorition was positive indication.

3.6.1 Test for saponins
Well-grounded plant material (1g) in water (15ml) in a test tube was heated on water bath for 5 minutes. The solution was filtered and left to cool to room temperature. The filtrate (10m) in 16 x 160mm test tube was shaken for 10 second and the height of honeycomb troth, which persisted, was measured. Froth higher than 1cm confirms the presence of saponins.

3.6.2 Test for tannins
10ml of water were added to 5g of extract ad the mixture was stirred and filtered. To 2ml of the filtered. To 2ml of the filtrate few drops of 0.1% fecl3 solution and the development of precipitate was observed. A blue-black, green precipitate indicates the presence of tannins.

3.6.3 Test for carbohydrate
5g of the powder sample was boiled in 1oml-distilled water on hot plate for 5 minutes and filtered whole hot. The filtrated was used for the following tests.
  i. Molisih test
To 3.0ml of the 2 filtrate was added 3 drops of molisch reagents then carefully run 3.0ml conc, H2SO4 without shaking. The interphase formed was then observed for purple.
  ii. Benedicts test
3 drops of the filtrate was added to 2.0ml of benedict reagent and placed on a hot plate for 5 minutes to observe the formation of brick red precipitate

3.6.4 Balsam test
To 3 drops of alcoholic ferric chloride was added to 2.0ml of extract then warm a dark green coloration if formed with balsam. To 2.0ml of the extract were added few drops of potassium permanganate. The solutions was then warmed on hot plate and observe for benzaldehyde or almond adour.
This was carried out in duplicate, and each set up was incubated at 37°C for 24 hours and the diameter of zone of inhibition in mm was recorded using a vernier caliper.
3.6.5 Phytochemical screening
3.6.6 Test for alkaloid
Twenty (20) mg of each of the extract was placed into a test tube, 1 ml of distilled water and 2 drops of 1% HCL were added and the solution was warmed gently in a waterbath to effect complete dissolution of the extract. A stream of dragendorff's reagent was added to the solution from a test tube.

3.6.7 Test for glycosides
A ml (10 of each of the extract solution was placed in a test tube and a drop each of 2% 3.5 dinitrobenzois acid in Methanol and 5% OH in water was added.

3.6.8 Test for tannins
A ml (1) of each of the extract was placed in a test tube and a stream of 5% FeCl₃ solution was added.

3.6.9 Test for flavonoids
Twenty (20) mg of each of the extract was dissolved in 2 ml ethanol in a test tube, a small size spatula full of zinc powder was added and a few drops of HCl was then added.

3.7.1 Test for soluble carbohydrates
Twenty (20) mg of each of the extracts was dissolved in 1 ml distilled water and 2 drops of 5% L-nahthol solution in methanol added in a test tube. While holding the tube at an angle, a stream of cone' H₂SO₄ was added to it.

3.7.2 Test for saponin
Twenty (20) mg of each of the extracts was dissolved in 1 ml of distilled water and 2 drops of 1% HCl was then heated gently on a water bath.

3.8 Construction of a sand filter
The design of a sand filter using two 200 litres plastic drums. The drum is cleaned out and hole is made two thirds of the way up so that an outlet pipe can be filtered. Depending on the size of the nipple, the hole is made. The water-collecting pipe is made with of hose piping. This is connected to the outlet pipe by a short hose piping. A number of saw cuts of drilled hole are made in hose piping ring and this is laid down on the bottom of the drum. The second drum is constructed for a storage drum. First a hole is made at the same level as that on the first drum and an appropriate nipple is fitted. A connecting hose is fix from the filter to the hole on the storage drum. Another hole is made at the other side and at the bottom of the drum at a height of about 7 cm from the base and a water collecting pipe if fitted such that it is long enough to be dipped at the top. In other cases a tap could be fitted for collecting water, but this can easily become loose as a result of constant opening and closing, so the hose is more preferable.

For setting up of the filter, clean sharp river sand of different sizes are obtained from a riverbed and sieved out. Gravels and coal of the correct sizes are also obtained and thoroughly washed with clean water; the sand is also thoroughly washed too kept in a place safe from dirt and dust.
3.8.1 Sand/gravel filter
This was constructed using sand and gravel only as media. The gravels are first placed at the bottom to a height of 75mm (7.5cm), this is followed by a layer of coarse sand to a height about 100mm. The last layer of fine sand is placed to a height just below the level of outlet pipe. This arrangement is made so that even if the tap is left on, the water drains out of the filter, a small layer of water remains above the sand. The sand must never be allowed to go dry, otherwise the biologically active ingredients in the sand which are important to the purification process, will die out. Both drums should have a lid to cover the drum, and this is made with a sieve or strainer for water with allot of sediments. The sieve should be covered too. When the filter has been completed, it must be thoroughly flushed through with clean water to further remove any dirt present. Once this is completed, a daily routine of adding raw water is maintained for a week or more so that the filter skin can form before usage begins. The design of the working components of the filter using a 200 litre drum should provide at least 624 litre of water per day. The yield rate will be controlled or regulated to 0.4 litres/minutes because the rate of flow of the filtered water to be slow to ensure satisfactory performance. However, if the raw water to be filtered has a bad odour, taste or colour, player of coal can be introduced between the sand and the gravel layers to control the situation. The procedure outlined about is for a 200 litre drum but the same technique can be used for a brick built container, metal drum, or clay pots. Below are some models of sand filter using different media and each had the calculation of the yield rate as a guide for other types of containers.

4. Results highlights
The picture below shows the turbidity clearance level with various treatments including untreated storm water left as control.

![Turbidity Clearance Level with Various Treatments](image)

Storm water being treated with Alum in 15 minutes, water is appears clear. 1 storm water treated with garcinia kola seeds, particles settles comparable to Alum. The third treatment container from left is storm water treated with Hibiscus seeds, particles settle but not as clear as Alum and Garcinia. The fourth treatment container from left is untreated storm water left as control, less settlement of particles.
The findings are presented in the following tables. Data in table 1a shows the pH and bacterial counts of foul wastewater from refinery, in the untreated wastewater, the total bacterial and fungal counts were high with a strog foul odour. Pseudomonas was also isolated in the untreated wastewater. In Table 1b, after treatment with plant materials, the total microbial counts dropped significantly to tolerable levels, the pH was stabilized while odour was no longer perceived. Pseudomonas spp was no more isolated. However, the various degree of treatment varies with the different plant materials applied. Moringa oleifera, Garcinia kola and Carica papaya exhibited the best results.

| Type of Treatment | Colour       | PH   | Smell         | Appearance | THBC Cfu/ml | Coliforms Cfu/ml | E. Coli Cfu/ml | TFC |
|-------------------|--------------|------|---------------|------------|-------------|------------------|----------------|-----|
| Untreated OVH     | Colourless   | 6.6  | Engine Oil, crude Oil smell | Clear       | 560         | Nil              | Nil            | 315 |
| Untreated DFH     | Brownish     | 7.05 | strong Engine oil Smell | Turbid      | 300         | nil              | nil            | 6140|

* OVH --> Overhead fraction or foul wastewater * Pseudomonas spp isolated
* DFH --> Desalter foul water

Table 3. (a) Effect of plant seed powders and Alum on oil Refinery wastewater from Kaduna State, Nigeria (Significant reduction in turbidity and microbial load using plant coagulants and disinfectants as indicated in the table)

| Type of Treatment | Colour       | PH   | Smell         | Appearance       | THBC Cfu/ml | Coliforms Cfu/ml | Coli Cfu/ml | TFC |
|-------------------|--------------|------|---------------|------------------|-------------|------------------|-------------|-----|
| Untreated OVH     | Colourless   | 6.6  | Engine oil, crude oil smell | Clear         | 560         | Nil              | Nil         | 315 |
| Treatment with Moringa Oleifera seed | Very colourless | 7.0  | odour absent | very clear | 36          | Nil              | Nil         | 100 |
| Treatment with Garcinia Kola seed | Very Colourless | 7.0  | odour absent | Clear        | 70          | Nil              | Nil         | 173 |
| Treatment with Carica Papaya seeds | Colourless   | 7.0  | odour absent (papaya odour) | Clear | 62          | Nil              | Nil         | 87  |
In table 4 below, the results indicated that plant materials exhibited great disinfection potentials on grey water (detergent based water) when compared to alum.

| Types of treatments                                 | THBC (Cfu/ml) | Coliforms (Cfu/ml) | E Coli (Cfu/ml) |
|-----------------------------------------------------|---------------|--------------------|-----------------|
| Untreated waste water sample                        | 2,200         | 2,300              | 1,900           |
| Untreated waste water sample left on bench and analysed | 2,120         | 2,224              | 1,892           |
| Alum treated sample                                 | 600           | 1,070              |                 |
| Moringa Oleifera treated sample                     | 320           | 520                | 343             |
| Jatropha Curcas treated sample                      | 770           | 890                | 729             |
| Garcinia Kola treated sample                        | 700           | 675                | 521             |
| Carica papaya treated sample                        | 697           | 682                | 575             |
| Persea americana treated sample                     | 800           | 760                | 690             |
| Hibiscus sabdariffa treated sample                  | 600           | 800                |                 |

* Wastewater normally stored for a week and then disposed 5g of powders of plant seeds used.

Table 4. Effects of Plant seed powders and alum on grey water detergent based water from Nasco Factory Jos, Nigeria
Table 5, the data shows that the physicochemical properties of the detergent based water such as turbidity and pH was significantly reduced when treated with the plant based coagulants when compared with the untreated wastewater sample.

| Treatment Material | Turbidity assessment                  | pH  | Remarks                               |
|--------------------|---------------------------------------|-----|---------------------------------------|
| Untreated wastewater sample | Very turbid, foamy bluish            | 6.5 | Odour intense turbidity remains the same |
| Untreated wastewater sample left on bench and analysed | Remains turbid, foamy.               | 6.5 | Odour intense turbidity the same      |
| Alum treated sample | Fast precipitation, very clear, slight odour | 5.0 | —                                     |
| Moringa Oleifera treated sample | Flocs formed, odour totally removed  | 7.0 | Odour removal colour removal protein positive. |
| Jatropha Curcas treated sample | Flocs formed settled at bottom       | 6.0 | —                                     |
| Garcinia Kola treated sample | Clear, flocs formed with a suspended pellicle | 6.0 | Second stage treatment clearer, odour off, after proper filtration |
| Carica papaya treated sample | —                                     | 6.0 | —                                     |
| Persea americana treated sample | clear, no odour flocs settled at the bottom | 6.0 | a second stage treatment was better.  |
| Hibiscus sabdarifera seeds treated sample | Flocs settled at the bottom must slight odour | 6.0 | Protein positive                      |

Table 5. Physicochemical properties of treated and untreated detergent based wastewater from food/detergent factory

In table 6, the data shows that the plant seed powders demonstrated a significant disinfection properties on stagnant water frequently used for irrigation, more than Alum. This observation has been extensively reported for Moringa (Yongabi et al., 2010) but not with the other plant materials used in this study.

| Type of Treatment                                      | THBC CFU/ml | Coliform counts CFU/ml | E Coli counts CFU/ml |
|--------------------------------------------------------|-------------|------------------------|----------------------|
| Untreated water sample initially collected             | TN TC       | TN TC                  | 8,900                |
| Untreated water Sample left to settle and supernatant analysed | TN TC       | 1,380                  | 980                  |
| Alum treated                                           | 3,598       | 298                    | 125                  |
| Moringa Oleifera seed treated                          | 485         | 298                    | 125                  |
| Jatropha seeds treated                                 | 2,212       | 598                    | 386                  |
| Garcinia Kola treated                                  | 387         | 452                    | 294                  |
| Carica papaya seed treated                             | 868         | 483                    | 223                  |
| Persea americana seed treated                          | 1,201       | 822                    | 429                  |
| Hibiscus sabdarifera seeds treated                     | 258         | 205                    | 110                  |

* TN TC = CFU/ml > 10,000

Table 6. Effect of plant seed powders and Alum on stagnant water from a dirty pond and used for irrigation of crops at Abubakar Tafawa Balawa University.

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In table 7, data indicates significant changes in pH and turbidity when plant materials are applied in a 24-72 hours retention time.

| Treatment materials | Turbidity assessment | PH range | Other Remarks |
|----------------------|----------------------|----------|---------------|
| Retention time 24 - 72 hours | | | |
| Untreated water sample initially collected | No floc formed at all | 7.0 | bad odour perceived |
| Untreated water sample left to settle and supernatant analysed | Few particles stuck to the wall of the container | 7.0 | |
| Alum treated | Flocs formed and settled | 5.0 | Odour (faint) |
| Moringa Oleifera treated | Flocs settled and good settlement | 7.0 | Very clear and second stage treatment very good, odour no trace. |
| Jatropha seed treated | Flocs formed particles settle | 7.0 | Odour of Jatropha |
| Garcinia Kola treated | Flocs settled with suspended pellicle. | 7.0 | Water clear and a second stage treatment clearer no odour. |
| Carica papaya seed treated | flocs formed slowly | 7.0 | papaya odour |
| Persea americana seed treated | flocs settlement seen slowly | 7.0 | no dour |
| Hibiscus sabdarrifa seed treated | flocs settled at the bottom | 5.0 | Little odour |

Table 7. Physicochemical properties of Treated and Untreated stagnant water used for irrigation of crops at Abubakar Tafawa Balewa University.

In table 8, the results show a significant level of disinfection of storm water by the plant materials in comparison with Alum. This findings is in tandem with a similar findings using Hibiscus, Moringa and Jatropha by Yongabi et al., 2011

| Types of Treatment | THBC CFU/ml | Coliform CFU/ml | E Coli CFU/ml |
|--------------------|-------------|-----------------|---------------|
| Untreated storm water sample initially collected | TNTC | TNTC | TNTC |
| Untreated storm water sample left to settle and supernatant analysed (24hours) | 9,280 | 212 | 36 |
| Alum treated storm water | 9,200 | 60 | 0 |
| Jatropha seeds treated water | 6,930 | 180 | 20 |
| Moringa Oleifera seeds treated storm water | 120 | 40 | 11 |
| Garcinia Kola seeds treated water | 6,33 | 160 | 18 |
| Carica Papaya seeds | 398 | 29 | 5 |
| Persea americana seeds | 5,360 | 64 | 0 |
| Hibiscus sabdarrifa | 4,024 | 50 | 32 |

* 1. Strom water harvested in flowing through the dirty streets of Yelwa after heavy rains.
  2. 5g of each the seed powder Alum into 100mls of the wastewater and left on bench for 24 hours.

Table 8. Effect of plant seed powders and Alum on Storm water collected from Bauchi Metropolis.
Not only did the microbial content changed after treatment with the natural materials but the pH and turbidity also changed considerably as shown in the data in table 9.

| Treatment Materials Retention time 24 - 72 hours | Turbidity assessment                                      | PH range  | Other remarks                      |
|--------------------------------------------------|-----------------------------------------------------------|-----------|-----------------------------------|
| Untreated storm water sample initially collected | No floc formed no settlement of particles, brownish and dusty smell | 6.9 - 7.0 | -                                 |
| Untreated storm water sample left on bench for 24 hours | No floc formed, few particles settle at the bottom, supernatant still has suspended particles. Some stuck to the walls of the container. | 6.9 - 7.0 | -                                 |
| Alum treated storm water                          | Floc formed and fast, water clear supernatant clear       | 5.0       | Clean and clear standard coagulant (1st)* |
| Moringa Oleifera seeds treated storm water        | Floc formed when seeds dispersed in water flocs settled slowly supernatant | 7.0       | Moringa mildly extracts in water. Good Coagulant (2nd)* |
| Jatropha Curcas seeds treated                      | Floc formed gently and settle                             | 7.0       | Good Coagulant (4th)*              |
| Garcinia Kola seed                                 | Floc formed, particles settled, very good coagulant at the bottom Excellent | 6.9-7.0   | Good Coagulant (3rd)*              |
| Carica papaya seed                                | Floc formed, particles settled                             | 6.9-7.0   | Good Coagulant (6th)*              |
| Persea americana Hibiscus sabdariffa               | Not very clear excellent, particles settled                | 6.9-7.0   | not a good coagulant (5th)*        |
|                                                   | Excellent, particles settled                               | 5.0-5.0   | Good Coagulant (5th)*              |

Table 9. Physicochemical properties of treated and untreated storm water with Alum and plant seed powders.

In the table 10 below, combined plant material with clay was applied in the treatment of refinery wastewater. This hybrid plant and geological material significantly improved the water quality bacteriologically and physicochemically than with the application of just either of the materials alone.

| Type of Treatment untreated OVH | Colour | PH Smell | Appearance | THBC | Coliform | Ecoli | TFC |
|--------------------------------|--------|----------|------------|------|----------|-------|-----|
| Untreated OVH                  | colourless | 6.6      | Engine oil, crude oil smell | Clear | 560 | Nil | Nil | 315 |
| Treatment with Moringa Oleifera seeds | Very clear no odor | 7.0      | no Odour | clear | 36 | Nil | Nil | 100 |
| Treatment with clay            | clear  | 7.0      | no odour  | clear | 32 | nil | nil | 96  |
| Untreated DFH                  | Brownish | 7.05     | Strong engine oil smell | Turbid | 300 | nil | nil | 6140 |
| Treatment with Moringa Oleifera seeds | very clear no odor | 7.0 | Odour absent | clear | 96 | nil | nil | 89  |
| Treatment with clay powder     | Clear  | 7.0      | Odour absent | clear | 93 | Nil | Nil | 84  |

Table 10. Effects of Combined Moringa Oleifera seed powder and clay in the treatment of oil refinery wastewater.
In the table 11 below, a combined plant material comprising plants (moringa oleifera seed powder) and sand filter media was applied to treat refinery wastewater and the results indicated a significant improvement in water quality both bacteriologically and phyicochemically better than with either of the materials alone. This corroborates a similar observation using surface water in Cameroon (Yongabi et al., 2010 and yongabi et al., 2011)

| Type of Treatment | Colour    | PH | Smell                              | Appearance | THBC | Coliform | Ecoli | TFC |
|-------------------|-----------|----|------------------------------------|------------|------|----------|-------|-----|
| untreated OVH     | colourles | 6.6| Engine oil, crude oil smell        | Clear      | 560  | Nil      | Nil   | 315 |
| Treatment with Moringa Oleifera seeds powder | Very Colourless | 7.0 | Odour absent                       | very clear | 36   | Nil      | Nil   | 100 |
| Final treatment with sand filter | Colourless | 7.0 | Odour absent                       | Very clear  | 3    | nil      | nil   | 111 |
| Untreated DFH     | Brownish  | 7.05| strong engine oil smell           | Turbid     | 300  | nil      | nil   | 6140|
| Treatment with Moringa oleifera seed powder | Very clear no colour | 7.0 | Odour absent                       | clear      | 96   | nil      | nil   | 89  |
| Final treatment with sand filter | Very clear no colour | 7.0 | Odour absent                       | clear      | 10   | nil      | nil   | 6   |

Table 11. Effect of combined Moringa Oleifera seed Powder and sand filter media on oil refinery waste water

The data in table 12 below also shows similar findings using Garcinia kola sand filter media as with moringa oleifera sand filter media.

| Type of Treatment | Colour    | PH  | Smell                              | Appearance | THBC | Coliform | Ecoli | TFC |
|-------------------|-----------|-----|------------------------------------|------------|------|----------|-------|-----|
| untreated OVH     | colourles | 6.6 | Engine oil, crude oil smell        | Clear      | 560  | Nil      | Nil   | 315 |
| Treatment with Garcinia Kola seeds powder | Very Colourless | 7.0 | Odour absent                       | Clear      | 70   | Nil      | Nil   | 173 |
| Final treatment with sand filter media | Very colourless | 7.0 | Odour absent                       | clear      | 15   | nil      | nil   | 8   |
| Untreated DFH     | Brownish  | 7.05| Strong engine oil smell           | Turbid     | 300  | nil      | nil   | 6140|
| Treatment with Garcinia Kola seed powder | Clear very little odour | 7.0 | no odour                          | clear      | 89   | nil      | nil   | 125 |
| Final treatment with sand filter media | Clear no colour | 7.0 | no odour                          | clear      | 13   | nil      | nil   | 29  |

Table 12. Effects of Combined Garcinia Kola seed powder and sand filter media on oil refinery wastewater
In tables 13, 14, 15, 16 and 17 the combined performance of the plant materials and clay, and cobined plant materials and sand filtered on various polluted water samples was tested bacteriologically and physicochemically. The results generally indicated strongly that these natural materials have strong ability to purify any type of water. The materials alone have the ability to treat water and wastewater but the combined effect of these materials have an added advantage in treating all kinds of polluted water as demonstrated by the data in the following tables 13, 14, 15, 16 and 17.

Table 13. Effects of Moringa Oleifera seed powder and clay in the treatment of detergent based waste

| Type of Treatment | Colour          | PH | Smell         | Appearance   | THBC | Coliform | Ecoli  | TFC |
|-------------------|-----------------|----|---------------|--------------|------|----------|--------|-----|
| untreated waste water sample (detergent) | Bluish, dirty   | 6.5 | Acid smell    | Turbid and foamy | 2,200 | 2,300 | 1,900 | -   |
| Treatments with Moringa seed powder | Blue, colour fades away | 7.0 | Odour absent  | Flocs formed | 320  | 520     | 343    | -   |
| Treatment with clay | Clear          | 7.0 | Odour absent  | Clear needs filtration | 309  | 511     | 338    | -   |

Table 14. Effects of Combined Hibiscus sabdariffa seed powder and sand filter media on oil refinery wastewater

| Type of Treatment | Colour          | PH | Smell         | Appearance   | THBC | Coliform | Ecoli  | TFC |
|-------------------|-----------------|----|---------------|--------------|------|----------|--------|-----|
| untreated OVH | Colourless      | 6.6 | Engine oil, crude oil smell | Clear | 560  | Nil      | Nil    | 315 |
| Treatment with Hibiscus Sabdariffa seed powder | Colourless   | 5.0 | Odour faint   | Clear | 133  | nil      | nil    | 113 |
| Treatment with sand filter media | Very colourless | 5.0 | Total odour removal | Very clear | 5    | nil      | nil    | 15  |
| Untreated DFH | Brownish        | 7.05 | Strong engine oil smell | Turbid | 300  | nil      | nil    | 6140|
| Treatment with Hibiscus Sabdariffa seeds | Clear       | 5.0 | Little odour   | Clear | 118  | nil      | nil    | 595 |
| Treatment with sand filter media | Clear       | 5.0 | No odour      | Clear | 3    | nil      | nil    | 20  |
| Type of Treatment | Colour          | pH  | Smell          | Appearance | THBC | Coliform | Ecoli | TFC |
|-------------------|-----------------|-----|----------------|------------|------|----------|-------|-----|
| untreated OVH     | colourless      | 6.6 | Engine oil, crude oil smell | Clear      | 560  | Nil      | Nil   | 315 |
| Treatment with carica papaya seeds | Colourless      | 7.0 | odour absent (papaya scent) | clear      | 62   | nil      | nil   | 87  |
| Treatment with sand filter media | very colourless | 7.0 | odour absent | clear      | 2    | nil      | nil   | 5   |
| Untreated DFH     | Brownish        | 7.05| Strong engine oil smell | Turbid     | 300  | nil      | nil   | 6140|
| Treatment with Carica papaya seeds | Clear very little odour | 7.0 | Little odour a bit of papaya scent | clear      | 90   | nil      | nil   | 95  |
| Treatment with sand filter media | Clear          | 7.0 | no odour       | clear      | nil  | nil      | nil   | 10  |

Table 15. Effects of Combined Carica papaya seed powder and sand filter media on oil refinery

| Type of Treatment | Colour          | pH  | Smell | Appearance | THBC | Coliform | Ecoli | TFC |
|-------------------|-----------------|-----|-------|------------|------|----------|-------|-----|
| untreated waste water sample (detergent) | bluish dirty    | 6.5 | acrid smell | Turbid and foamy | 2200 | 2300     | 1900  | -   |
| Treatment with Moringa Oleifera powder seed | flocs formed colour removed | 7.0 | odour removed totally | clear, no foams | 320  | 1070     | 343   | -   |
| Treatment with sand filter media | clear            | 7.0 | no odour | clear      | -    | -        | -     | -   |

Table 16. Effects of Combined Moringa Oleifera seed powder and sand filter media on detergent based

| Type of Treatment | Colour          | pH  | Smell          | Appearance | THBC | Coliform | Ecoli | TFC |
|-------------------|-----------------|-----|----------------|------------|------|----------|-------|-----|
| untreated wastewater sample (detergent) | very foamy blush | 6.5 | acrid smell | Turbid foamy | 2200 | 2300     | 1900  | -   |
| Treatment with Garcinia Kola seed powder | flocs formed with a suspended pellicle | 7.0 | smell reduced | becoming clear | 700  | 675      | 521   | -   |
| Final treatment with sand filter media | clear            | 7.0 | odour absent  | clear      | 100  | 5        | 1     | -   |

Table 17. Effects of Combined Garcinia Kola seed powder and sand filter media on detergent based water
In table 18 below, one of the plants: Moringa oleifera was used to study its effect on unicellular organisms in water. The results indicated that moringa oleifera seed powder gets rid of unicellular organisms such as amoeba, microalgae such as spirogyra from water. The need to study the application of plant materials in the removal of microalage from water systems could be rewarding.

| Types of Organisms | approximate number per field |
|--------------------|-----------------------------|
| Diatoms            | up to 15 per field          |
| Cercaria           | a few                       |
| Euglena            | 35 cells per field, actively motiles |
| Cyclops            | a few                       |
| Amoeba             | More than 15 per field      |
| Debris             | a lot of debris             |
| Spirogyra          | a lot                       |

a) Microscopy of pond/stagnant water before treatment

| Types of Organism                  | approximate number per field     |
|------------------------------------|----------------------------------|
| Euglena                            | totally absent, water clean      |
| Diatoms                            | absent                           |
| spirogyra (blue/green algae)      | absent                           |
| Cyclops and cercaria               | absent                           |
| Amoeba                             | absent                           |

b) Microscopy of Pond/Stagnant water after treatment with Moringa Oleifera seed powder, and after filtration

Table 18. Effects of Moringa Oleifera seed powder on free living organisms in pond water used for irrigation

An novel attempt was made to classify materials that can be applied in water pollution management and shown in table 19 below. More studies for a detail classification are underway.
Table 19. Survey and classification of Natural materials for water pollution management in local communities

In table 20, the nature of extracts from Garcinia kola was described. The water extract is a black solid. The coagulant and disinfection activity of Garcinia kola observed in this study may be soluble in water. More studies are needed in this dimension.

| Botanical name of plants | Common name/ Hausa name | Part used | Types of wastewater | Types of Coagulant | Sources                      |
|-------------------------|-------------------------|-----------|---------------------|------------------|------------------------------|
| Jatrohops Curcas         | Physic not Benin Zugo   | Seeds     | Industrial effluents domestic wastewater | Phyto Coagulant   | Yongabi, K.A (2004)          |
| Sychonos Potatotum      | -                       | Seeds     | Domestic water      | Phyto-Coagulant   | -----                        |
| Moringa Oleifera        | Horse- raddish Zogale   | Seeds     | Domestic water      | Phyto-coagulant   | Pers.Comm. Yongabi, K. A     |
| Calotropsis proceria    | tumafiya                | latex     | Wastewater          | "                | Pers. comm.                  |
| Citrus aurantifolia     | Limes, lemu             | seeds     | Domestic water      | "                | -----                        |
| Punice                  | Rock                    | -         | domestic and industrial wastewater | Geocoagulant:Internet |
| Bentonite               | Rock                    | -         | "                   | "                | "                            |
| Immansil                | "                       | -         | "                   | "                | "                            |

Table 20. Analysis of Phytochemical tests on solvent extracts of Garcinia Kola

The results in table 19 gives an attempt to classify some of the phytoconstituents in this plant materials. More phytonutrients were detected in the aqueous extract suggesting an easy and cheap means of extracting water treatment chemicals from Garcinia kola.
Table 21. Result of Preliminary phytochemical analysis of Solvent Extracts of Garcinia Kola

| Solvent Extract | Cardiac Glycosides | Saponin | C6H12O6 | Tannins | Flavonoids | Alkaloids |
|-----------------|---------------------|---------|---------|---------|------------|-----------|
| Dichloromethane | -                   | -       | -       | -       | -          | -         |
| n-Hexane        | +                   | -       | -       | -       | -          | -         |
| Toluene         | -                   | -       | -       | -       | -          | -         |
| Acetone         | -                   | -       | -       | -       | -          | -         |
| Methanol        | -                   | +       | -       | +       | -          | -         |
| Water           | +                   | +       | +       | +       | -          | -         |

Table 22. PH Content of various waste water/water treated with Alum and plant seed powders

| Types of water     | PH (Normal) | PH Alum (treated) | PH Moringa (treated) | PH Garcinia (treated) | PH Hibiscus (treated) | PH Carica (treated) | PH Jatropha |
|--------------------|-------------|-------------------|-----------------------|-----------------------|-----------------------|---------------------|-------------|
| Dirty tap water    | 6.62        | 5.0               | 7.0                   | 6.99                  | 5.0                   | 7.0                 | 7.0         |
| YeIwa tap Water    | 7.36        | 5.0               | 5.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |
| University tap water | 7.25        | 5.0               | 7.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |
| YeIwa well water   | 7.37        | 5.0               | 7.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |
| Asbestos water (well) | 7.46        | 5.0               | 7.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |
| Asbestos tap water | 7.53        | 5.0               | 7.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |
| Cement waste water | 8.01        | 5.0               | 7.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |
| Cement waste water | 8.52        | 5.0               | 7.0                   | 7.0                   | 5.0                   | 7.0                 | 7.0         |

Table 22. PH Content of various waste water/water treated with Alum and plant seed powders

To further demonstrate the disinfection potential of the plant materials on the wastewater samples, a methanol extract of the plant materials was conceivable. The resulting extracts were tested on various bacterial isolates from all the polluted water samples. The data in table 21 below demonstrates a significant level of antibacterial activity comparable to Alum...
Table 23. Effect of cold Methanol and Aqueous Extract of Garcinia Kola, Carica papaya and Hibiscus Sabdariffa seeds on Bacterial isolates from waste water (Diameter zone of inhibition in mm)

| Extracts                                          | E. coli | Pseudomonas Sp | Klebsiella Sp | Staphylococcus |
|---------------------------------------------------|---------|----------------|--------------|---------------|
| Garcinia Kola seeds Aquenous Extract Methanol     | 60mm    | 6mm            | 15mm         | 18mm          |
| Hibiscus sabdariffa seeds Aqueous Extract Methanol| 10.8mm  | 12.0mm         | 12mm         | 15mm          |
| Carica papaya Seeds Aqueous Extract Methanol      | 9mm     | 12mm           | 14mm         | 16mm          |
| Aluminium Sulphate                                | 12mm    | 13mm           | 10mm         | 10.5mm        |
| Water                                             | 0mm     | 0mm            | 0mm          | 0mm           |
| Methanol                                          | 5mm     | 9mm            | 11mm         | 8mm           |

CFU Colony Forming Units
COD Chemical Oxygen Demand
CWE Crude Water Extract
IFX Ion Exchange
MIC Minimum Inhibitory Concentration
MO Moringa Oleifera
MOCP Moringa Oleifera Coagulant Protein
UC Uniformity Coefficient
OD Optical Density
WHO World Health Organization
WPC Water Production per cycle
TNTC Too numurous to count.
Mi Mililitre
OVH Overhead Fraction or Foul Water
DFH Desalter Foul Water

5. A pilot water treatment plant using natural materials at government technical College, Njinikom, Bamenda, Cameroon

The Phytobiotechnology Research Foundation (PRF), Cameroon, in collaboration with the School of Chemical Engineering, The University of Adelaide, South Australia, is proposing to carry out a capacity building training on: A simple Moringa- sand based water filtration technology for clean potable water supply in the rural schools and villages in Boyo Division, Cameroon. This is part of a doctoral research in chemical engineering, The University of Adelaide, south Australia. Three undergraduate students in chemical engineering are undertaking their honours thesis on the water quality, management and training, safety and ethical issues associated with the implementation of Integrated biocoagulant-sand filter system for drinking water purification at Government technical college,
Njinkom, Cameroon. A well with an approximate water volume of 2500 litres has been dug, and a filtration system using Moringa oleifera seeds and sand filter is being constructed expected to purify 2000 litres of water in 24 hours retention time to serve more than 7000 students.

5.1 Anticipated benefits

- Clean potable water will be available for rural people.
- Decimation of incidence of infectious/waterborne diseases
- Improved health

5.2 Training method

The training shall be conducted in conjunction with local NGOs in Bamenda, Cameroon. PRF is an NGO based in Bamenda and has a track record on community development projects in Cameroon and Nigeria. PRF has entry points to communities and has over the years worked with a number of Research institutes in the country. PRF has facilitated a number of training for local groups in Bamenda water quality in rural areas. Similarly, PRF has participated at training on water filtration technology at the ZERI Centre in Nigeria. Five (5) selected people from the local schools shall be trained and then the school authority shall provide them the resources to mount the outfits. The students will be encouraged to set up a household filter unit in their homes during holidays.
These trainees shall function in union with the PRF and the school authority who will in turn monitor and supervise effective functioning of the filter units.

6. Conclusion and recommendation

The research work has shown that there are many natural materials available in many communities in the world that can be used to treat water for drinking. Additionally, this research has demonstrated that these plant and geological materials can be applied in the treatment of any type of polluted water. These materials are ecological, low cost when compared to the application of synthetic chemicals currently used in water pollution management. The ongoing pilot system applying natural materials in water treatment in Cameroon could be replicated elsewhere. More research into the use of natural materials in water pollution management should be studied.

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Water pollution is a major global problem that requires ongoing evaluation and revision of water resource policy at all levels (from international down to individual aquifers and wells). It has been suggested that it is the leading worldwide cause of deaths and diseases, and that it accounts for the deaths of more than 14,000 people daily. In addition to the acute problems of water pollution in developing countries, industrialized countries continue to struggle with pollution problems as well. Water is typically referred to as polluted when it is impaired by anthropogenic contaminants and either does not support a human use, such as drinking water, and/or undergoes a marked shift in its ability to support its constituent biotic communities, such as fish. Natural phenomena such as volcanoes, algae blooms, storms, and earthquakes also cause major changes in water quality and the ecological status of water. Most water pollutants are eventually carried by rivers into the oceans.

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