Bacteriological Findings Of Reverse Osmosis Water From Karachi City.

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Abstract

Water is one of the most abundant and essential element for all the living organisms. Safe drinking water is a need for human life. It is generally considered that bottled water is safe for usage by people. For long-distance travelers, it serves as the only source of reliable drinking water. But, several studies have reported that bottled water does not always meet the acceptability standards.

In this study reverse osmosis plant water is used because it is able to do so much good in a way that it removes all dissolved salts, chemicals, impurities and microorganisms as well and is considered as safe, clean and pure for human consumption in any way. Therefore, it is important to analyze the quality of Reverse Osmosis water. There are various categories of bottled water and these are mineral water, spring water and purified water

This study is based on the quantitative analyses of Gram negative bacteria in reverse osmosis purified water bottles because contaminated water poses great health risk, especially when it is used by infants, hospitalized patients and immuno-compromised ones. Gram negative causes some severe gastrointestinal diseases such as Cholera, Gastroenteritis, Typhoid fever, Salmonellosis, Shigellosis, Diarrhea, etc. For this purpose the samples of bottled reverse osmosis drinking water from different areas of Karachi were tested for the presence of Gram negative bacteria. The result of this study showed 14% of the samples, were free from Gram negatives and 86% samples were significantly positive for the presence of Gram negative. Thus water must undergo an effective treatment to reduce the bacterial count in order to prevent from infection.

Key Words: ***Reverse Osmosis, quality, bottled water, gram negative.***

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INTRODUCTION:

Water is transparent fluid which forms the world's stream, lakes, oceans, rivers and the major constituent of the fluids of living things (1). It is important to life on Planet Earth. It is important for food production, industrial development, and ecological sustainability. The total amount of water reserved by our thirsty world was estimated at 3906 km3 in 1995. Estimates for 2025 predict a 50% increase for local, livestock, and industrial uses, leaving less water for irrigation and therefore threatening food production(2) shows the total water withdrawal by regions around the world and the predictions for 2025.

Early on, the ancient people (Egyptians, Greeks, and the Romains) were already worried with water hygiene and sanitation (3). A major innovation was due to Antony van Leeuwenhoek who, in 1684, called "animalcules" in water, and later on, in the nineteenth century, Robert Koch and Joseph Lister, showed that the "animalcules" were definitely microorganisms (4, 5). Another major breakthrough is when John Snow, a British physician, recognized a link between microorganisms and water-related diseases like cholera.

The mission for safe drinking water dates back to at least 4000 years ago when a medical theorist promoted the use of boiling, sunlight, charcoal filter, and copper vessels to treat water (6) and now drinking water safety is a universal concern. Polluted drinking water has the greatest influence on human health worldwide, particularly in developing countries. It is projected that more than one billion of the world's population does not have access to safe clean water and about 2.6 billion lack improved sanitation (7, 8, 9)

TYPES OF WATER:.

Water and its significance have been highlighted upon since centuries. The colorless and tasteless mixture of life comes in many types.

Tap water:

Tap water is the water that you get straight from your faucet, it may or may not be suitable for drinking purposes. It is widely used for domestic routines such as cleaning, cooking, gardening and washing clothes. It must meet the guidelines set by the local urban bodies. Tap water is a type of water that you get directly from your tap.

Well water:

When it rains, water drops down and travels through the inner gaps of the soil, underneath the ground to form underground lakes. This occurs over a period of time. In rural areas, one of the prime sources of water is what is dug out from deep wells. Deep wells directly hit

groundwater and bring it to the exterior from which people can yield their water. (10)

Rainwater:

In areas where rainfall is plentiful and common, rainwater can be a noble source of water supply. Rainwater has several benefits. It is free, fairly clean and usually reliable, even if it rains only once or twice a year, and a rainwater collecting system can be easily built and conserved at low cost. Although mainly found in rural areas, rainwater collecting can also be useful in an urban state. (11)

Seawater:

Saltwater is plentiful in the surface of the planet. However, saltwater is currently not mainly useful when it comes to clean water supplies. Purification plants, while they do exist, are rare because the energy required for purification makes the process enormously expensive. (12)

Distilled or Demineralized Water:

Distilled water goes through a boiling procedure that strips away all of its impurities—and all of its healthy minerals. While it's clearly good to remove the pollutants in your diet, there are probable health concerns that come from eliminating the minerals from water. (13)

Waste Water:

Wastewater is any water that has been affected in feature by human actions. Wastewater can cultivate from agricultural activities, urban water use, and sewer invasion and storm water runoff just to name a few. Wastewater from a city is also called sewage. Most of us don't want to think about it, but at times the water that spins in the bowl ends up being cured and ends up in our taps. (14)

PROPERTIES OF WATER:

Chemical Properties of Water:

SThe chemical value of the aquatic location differs according to native geology, the climate, the distance from the ocean and the quantity of soil cover, etc. If superficial waters were totally unaffected by human actions, up to 90-99 per cent of worldwide freshwaters, dependent on the variable of interest, would have natural chemical deliberations appropriate for aquatic life and most human uses. Rare (between 1 and 10 per cent and between 90 and 99 per cent of the universal circulation) and very rare (< 1 per cent and > 99 per cent of the universal circulation) chemical circumstances in freshwaters, such as happen in salt lakes, hydrothermal waters, acid volcanic ponds, peat bogs, etc., usually make the water inappropriate for human. However, a variety of aquatic organisms have improved to these extreme situations. In many areas groundwater concentrations of total dissolved salts, fluoride, arsenic, etc., may also truly surpass maximum allowable concentrations (MAC) (15)

Physical Properties of Water:

Water is a minor solvent, occupying about 0.03 nm3 per molecule in the liquid form at room temperature and pressure, yet it is extremely cohesive because of the strong intermolecular connections (hydrogen bonds, or H-bonds) between the oxygen and hydrogen atoms. This is revealed in its high boiling point, the great amount of heat required to vaporize it, and its high surface tension. Replacement of one or both of the hydrogens dramatically fails these intermolecular interactions, decreasing the magnitude of these measures. The strong cohesive connections in water also result in:

(1) A high viscosity, since for a liquid to flow interactions between adjacent molecules must always be broken; and

(2) A high specific heat capacity – the capability to store a large quantity of potential energy for a given addition in kinetic energy (temperature). (16)

Biological Properties of Water:

The growth of biota (flora and fauna) in superficial waters is ruled by a diversity of ecological conditions which define the choice of species as well as the physiological presentation of individual organisms. The primary construction of organic matter, in the form of phytoplankton and macrophytes, is most concentrated in lakes and pools and usually more partial in rivers. The degradation of organic materials and the related bacterial invention can be a long-term procedure which can be important in groundwater and deep lake waters which are not directly visible to sunlight. In distinction to the chemical feature of water bodies, which can be measured by appropriate analytical means, the explanation of the biological quality of a water body is a mixture of qualitative and quantitative classification. Biological monitoring can usually be carried out at two dissimilar levels. (15)

WATER SOURCES:

Groundwater:

Groundwater is that water which is present beneath Earth's surface in soil pore spaces and in the fractures of rock formations. A unit of rock or an unconsolidated deposit is called an aquifer when it can yield a usable amount of water. The depth at which soil pore spaces or fractures and voids in rock become fully saturated with water is termed as water table. Groundwater is recharged from the surface. (17)

Groundwater is commonly cheaper, more convenient and less vulnerable to pollution than surface water. Therefore, it is used for public water supplies. For example, groundwater provides the largest source of usable water storage in the United States, and California annually withdraws the largest amount of groundwater of all the states.(18) Underground reservoirs contain far more water than the capacity of all surface reservoirs and lakes in the North America, including the Great Lakes. Many municipal water supplies are derived solely from groundwater. (19)

Upland lakes and reservoirs:

Typically present in the headwaters of river systems, upland reservoirs are usually sited up above any human habitation

and may be surrounded by a protective zone to restrict the opportunities for contamination. Bacteria and pathogen levels are usually low however some bacteria, protozoa or protoctist will be present. Where uplands are forested or peaty, humic acids may color the water. Many upland sources have low pH which needs adjustment.

Rivers, canals and low land reservoirs:

Low land surface waters will have a significant bacterial load and may also contain algae, suspended solids and a variety of dissolved constituents. (20)

Atmospheric water generation:

A device, named an atmospheric water generator (AWG) that extracts water from humid ambient air. Water vapor in the air can be extracted by condensation - cooling the air below its dew point, exposing the air to desiccants, or pressurizing the air. (21)

Rain water harvesting or fog collection:

Rainwater harvesting is a type of harvest in which the rain drops are collected and stored for the future use, rather than allowing it to run off. (22) The harvested water can also be used as drinking water, longer-term storage, and for other purposes such as groundwater recharge. Rainwater harvesting is one of the simplest and oldest methods of self-supply of water for households usually financed by the user. (23)

Desalination of seawater by distillation:

Desalination is a process that takes away mineral components from saline water. More generally, desalination refers to the removal of salts and minerals from a target substance (24) as in soil desalination, which is an issue for agriculture. (25).

Surface water:

Water on the surface of continents such as in a river, lake, or wetland is surface water. It can be contrasted with atmospheric water and groundwater. Non-saline surface water uses is replenished by precipitation and by recruitment from ground-water. (26)

WATER PURIFICATION:

Water purification is the process of removing undesirable substances like chemicals, biological contaminants, suspended solids, and gases from water. The aim is to produce water fit for specific purposes. Most water is purified and disinfected for human ingestion (drinking water), but water purification may also be carried out for a variety of other purposes such medical, pharmacological, chemical, and industrial applications. The WHO estimates that 94% of these diarrheal disease cases are preventable through modifications to the environment, including access to safe water. (27) Simple techniques for treating water at home, such as chlorination, filters, and solar disinfection and for storing it in safe containers could save a huge number of lives each year. (28) Reducing deaths from waterborne diseases is a major public health goal in developing countries. (20)

Screening:

A screen filter is a type of [filtration of water] employing a rigid or versatile screen to separate sand and other fine particles out of water for irrigation or industrial applications. These are typically not recommended for filtering out organic matter such as algae, since these types of contaminants may be extruded into spaghetti-like strings through the filter if enough pressure drops occurs across the filter surface. Typical screen materials include stainless steel (mesh), polypropylene, nylon and polyester. (29)

Pre chlorination and Primary Disinfection:

Many serious diseases, such as cholera, are caused by drinking water that contains parasitic microorganisms. We need to treat the drinking water that is suitable to us so we can stay healthy and so we don't develop diseases or health problems through our drinking water. Water disinfection is the removal, deactivation or killing of pathogenic microorganisms.

Microorganisms are deactivated or destroyed, resulting in termination of growth and reproduction. When microorganisms are not removed from drinking water, drinking water intake will cause people to fall ill. For chemical disinfection of water, the following disinfectants may be used:

- Chlorine (Cl2)
- Chlorine dioxide (ClO2)
- Hypo chlorite (OCl-)
- Ozone (O3)
- Halogens: bromine (Br2), iodine (I)
- Bromine chloride (BrCl)
- Metals: copper (Cu2+), silver (Ag+)
- Potassium permanganate (KMnO4)
- Phenols
- Alcohols
- Soaps and detergents
- Kwartair ammonium salts
- Hydrogen peroxide
- Several acids and bases

For physical disinfection of water, the listed disinfectants can be used:

- Ultraviolet light (UV)
- Electronic radiation
- Gamma rays
- Sounds
- Heat (30)

Coagulation and flocculation:

One of the primary steps in a conventional water purification

process is the addition of chemicals to assist in the removal of particles suspended in water. Particles can be inorganic such as clay and slit or organic such as algae, bacteria, viruses, protozoa and natural organic matter. Inorganic and organic particles contribute to the color and turbidity in water. (31) In coagulation, a liquid coagulant, such as aluminum sulfate, is added to the water to attract suspended particles. The water is then gently stirred to permit the particles to come together and form larger particles (flocculation), which may then be removed by sedimentation, settlement or filtration. The amount of coagulant needed will depend on the nature of the contaminating chemical compounds and solids. (32)

Sedimentation:

After the bulk of the suspended particles have settled, water exits the flocculation basin and enters a sedimentation basin. Sedimentation basins move treated waters along through the purification process while allowing remaining particles to settle. Sludge forms that appear on the floor of the tank are removed and treated. (33) Typical detention times for sedimentation vary from 1.5 to 4 hours and basin depths vary from 10 to 15 feet (3 to 4.5 meters).(31)

Filtration:

A rapid sand filter is the most common type of filter. Water moves vertically through sand which often has a layer of activated carbon or anthracite coal on the sand. The top layer removes organic compounds that contribute to taste and odor. The space between sand particles is larger than the tiny suspended particles therefore simple filtration is not enough. Most particles pass through surface layers but are trapped in pore spaces or adhere to sand particles. (34)

Slow sand filters can be used where there's sufficient land and space, as the water flows very slowly through the filters. These filters rely on biological treatment processes for their action instead of physical filtration. They are carefully constructed using graded layers of sand, with the coarsest sand, along with some gravel, at the bottom and finest sand at the top. (35)

Secondary Disinfection:

Disinfection is accomplished both by filtering out harmful micro-organisms and conjointly by adding disinfectant chemicals. Water is disinfected to kill pathogens which pass through the filters and to provide a residual dose of disinfectant to kill or inactivate potentially harmful micro-organisms in the storage and distribution systems. Possible pathogens include viruses, bacteria, including Salmonella, Cholera, Campylobacters and Shigella, and protozoa, including Giardia lamblia and other cryptosporidia. After the introduction of any chemical disinfecting agent, the water is usually held in temporary storage – often called a contact tank or clear well – to allow the disinfecting action to complete. (36)

Chlorine disinfection:

The most used disinfection method involves some form of chlorine or its compounds such as Chloramine or chlorine dioxide. Chlorine is a strong oxidant that rapidly kills many harmful micro-organisms and help keep the water clean until distribution.

Ozone disinfection:

Ozone is an unstable molecule which readily gives up one atom of oxygen providing a powerful oxidizing agent that is toxic to most waterborne organisms. It is a very strong, board spectrum disinfectant that is widely used in Europe. It is an efficient method to inactivate harmful protozoa that form cysts (29).

WATER POLLUTION:

Water pollution is the contamination of natural water bodies by radioactive, chemical, physical or pathogenic microbial substances. Adverse alteration of water quality presently produces large scale illness and deaths, accounting for approximately 50 million deaths per year worldwide, most of these deaths occurring in Africa and Asia (37). In China, as an example, about 75 percent of the population (or 1.1 billion people) are without access to pure drinking water, according to China's own standards (38). Widespread consequences of water pollution upon ecosystems include species mortality, biodiversity reduction and loss of ecosystem services.

TYPES OF WATER POLLUTION:

There are many types of water pollution because water comes from many sources. Here are few types of water pollution:

Chemical pollutants:

They are generally atoms or molecules, which have been discharged into natural water bodies, usually by activities of humans. Common examples of such chemical water pollutants are mercury emanating from mining activity, some nitrogen compounds utilizing in agriculture, chlorinated organic molecules arising from sewage or water treatment plants (39) or various acids which are the externalities of various manufacturing activities.

Physical pollutants :

Physical water pollutants are either (a) much more larger particles or (b) physical factors like temperature change, both of which while not typically toxic, cause a variety of harmful effects. The absolute physical pollutants are (a) excessive sediment load, mostly arising from over-intense land use practices and (b) rubbish discarded from anthropogenic activity (e.g. plastic bags, bottles). Whereas these materials are not so harmful to human health as chemicals or pathogens, they comprise the majority of visual impact of water pollution.

Radioactive substances

They are really merely a special sub-class of chemical pollutants, and by mass represent the smallest of the contributors to water pollution; however, their potential for harm allows recognition

as a separate class. In fact, most discharge of radiation is not from the negligible escape from nuclear power plants however but rather arises from agricultural practices like tobacco farming, where radioactive contamination of phosphate fertilizer is a common technique of introduction of radioactive materials into the environment.

Microbial Pollutants:

Common pathogenic microorganisms introduced into natural water bodies are pathogens from untreated sewage or surface runoff from intensive livestock grazing. One of the most common disease agents is a Giardia lamblia, a parasitic protozoan present in fecal material of many fauna including humans; this pathogen is particularly insidious, due to its resistance to conventional sewage treatment. Giardia and other protozoan and bacteria are important causes of illness and mortality in developing countries where population density, water insufficiency and inadequate sewerage treatment combine to occasion widespread parasitic and bacterial disease. (40)

Pesticide pollution of water

In terms of pesticide water pollution, each pesticide has unique properties and many variable factors affect its risk, such as the active ingredients, contaminants and additives as well as any degradate formed during chemical, photochemical, microbial degradation of active ingredients. The increased use of pesticides has been accompanied by the growing presence in soil of a large number of transformation products (TPs) for a wide variety of pesticides. (41)

Salt loads to water:

The quantity of dissolved salts in water is called salinity. Freshwater bodies can receive salt through distinct pathways, for example through direct surface runoff from saline lands, subsurface drainage of saline waters to fresh water bodies, or the interception of saline stores because of elevation of the ground water table that also may recharge surface waters. (42)

Agricultural erosion and sediment loads to water:

Agricultural activities contribute to enhanced soil erosion and sediment loads in river systems. Pollutants may be released from sediments when the environment (e.g. redox potential or pH) changes. Therefore, additionally to the dissolved pollutants that reach water bodies, sediments may carry more pollutants to aquatic ecosystems, and are effectively the most important pathway for some sorts of pollutants with low solubility such as phosphates (43), some metals (44) and pesticides (45).

Organic Matter:

Pollution by organic matter is increasing because of increasing municipal and industrial wastewater discharge, the intensification of agriculture (including animal farming) and reduction in river dilution capacity due to weather change and water extractions. Water pollution by organic matter from intensive livestock farming is now significantly more widespread (46)

BOTTLED WATER:

Today, bottled water is primarily used for drinking but has also been considered for the preparation of baby formula, cleaning contact lens or used in humidifiers (47). Bottled water is perceived by consumers as being of better quality and better taste than usual tap water. However, the higher quality of bottled water is sometimes a thought. Several researchers reported that bottled water is of no better quality than municipal drinking water. Furthermore, it is also hundred times more expensive than tap water.

In 2011, the highest consumers were Italy (179 L/person per year) and Germany (171 L/ person per year) while the lowest was Finland (18 L/person per year) (48)

TYPES OF BOTTLED WATER:

There are various kinds of bottled water and these are not directly comparable to one another (49).

Spring water:

Water is derived from an underground formation that flows naturally to the earth surface. In 2011, spring water accounted for 14% of total sales in Europe, as compared to 83% for mineral water. In United States, approximately 25% of the bulk water is spring water. This water might be disinfected with ozone.

Sparkling Bottled Water:

It is “naturally” carbonated with carbon dioxide. It can also be created bubbly by adding dissolved carbon dioxide. Some brands of bottled water (e.g., Perrier) are made bubbly by harvesting the “natural” carbon dioxide at the source and re injecting it into the water during the bottling operation.

Mineral water:

It contains dissolved cat ions and anions (e.g., Ca2+, Mg2+, Na+, HCO3−). The mineral content varies from one supply to another and may be sometimes not up to the mark of community water supplies. Mineral water is drawn from underground supply that contains at least 250mg/L of total dissolved solids. Minerals and trace elements must come from the supply of the underground water and cannot be supplemented later. Natural mineral water is one that is drawn from a protected supply. It becomes enriched with minerals because it flows through underground formations. According to European Union regulations, manufacturers are not allowed to alter the mineral content of this type of water nor disinfect it to alter the microbial flora of this natural resource. The sole treatments allowed are filtration to get rid of particles or iron, manganese, sulfur or arsenic along with the addition or removal of CO2. Furthermore, it must be monitored to prevent contamination from allochthonous microorganisms together with pathogens and parasites. (50)

Artesian Water:

Water from a well tapping a confined aquifer in which the water

level stands at some height above the top of the aquifer. (51)

Purified Water:

Water that has been resulted by distillation, deionization, reverse osmosis or other different suitable processes may be labeled as purified bottled water. Other suitable product names for bottled water treated by one of the above processes can include "distilled water" if it is produced by distillation," deionized water "if it is produced by deionization or "reverse osmosis water" if the process used is reverse osmosis. Alternatively, " drinking water" may be used with the blank being filled in with one of the terms defined in this paragraph (e.g., "purified drinking water" or "distilled drinking water") (52).

Reverse Osmosis (RO) is a method that employs the use of a semi-permeable spiral wound membranes to separate and eliminate dissolved solids, organic, pyrogens, submicron mixture matter, color, nitrate and bacteria from water. Feed water is conveyed under pressure through the semi porous membrane; where water permeates the minute pores of the membrane and is delivered as purified water referred to as permeate water. Impurities within the water are concentrated in the reject stream and flushed out of the drain is referred as reject water. These membranes are semi-permeable and reject the salt ions whereas allowing the water molecules to pass. The materials used for RO membranes are manufactured from cellulose acetate, polyamides and different polymers. The membrane consists of hollow-fiber, spiral-wound used for treatment; rely on the feed water composition and the operation parameters of the plant. Reverse Osmosis (RO) is a membrane based method technology used for desalination. Membrane-based seawater desalination and wastewater reuse are widely considered as promising solutions to augment water supply and alleviate water scarcity (53). The most common membrane processes used are the reverse osmosis (RO) and the electro dialysis (ED) used for brackish water desalination, but only RO competes with distillation processes in seawater desalination (54).

Scope of Reverse Osmosis:

The process has additionally been applied to treat municipal wastewater. Since standard municipal treatment processes do not remove dissolved solids, however RO process is used for the elimination of dissolved solids. RO is widely used as a separation technique in chemical and environmental engineering to get rid of organics and organic pollutants present in wastewater. It is seen from literature review that Reverse Osmosis (RO) processes have been widely used for separation and concentration (recovery) of solutes in many fields. (55)

Reverse Osmosis Process Description:

The RO process is easy in design consisting of feed, permeate and reject stream. For feed water it is necessary to perform pretreatment in order to get rid of inorganic solids and suspended solid and using high pressure pump given feed through semi permeable membrane. Relying upon the permeate where ever it is used necessary post treatment is given.

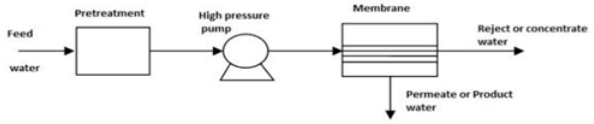


Figure 1. *The schematic diagram is as follow (56):*

Reverse Osmosis Requisites:

An RO desalination plant basically consists of 4 major systems: (a) Pretreatment system, (b) High pressure pumps, (c) Membrane systems, and (d) Post-treatment. Pre-treatment system is provided to get rid of all suspended solids in order that salt precipitation or microbial growth does not occur on the membranes. Pre-treatment could involve typical strategies sort of a chemical feed followed by coagulation/ flocculation/ sedimentation, and sand filtration or membrane processes i.e. micro filtration (MF) and ultra filtration (UF). High-pressure pumps supply the pressure required to change the water to pass through the membrane and have the salt rejected. The pressure ranges from 17 bar to 27 bar for brackish water, and from 52 bar to 69 bar for seawater. Membrane systems accommodate a pressure vessel and a semi-permeable membrane within that permits the feed water to pass through it. RO membranes for desalination usually available in 2 types: Spiral wound and Hollow fiber. Relying upon water quality of permeate and use of permeate; post treatment may consists of adjusting the pH and disinfection (56).

Membrane Characteristics:

The membrane ought to be cheap, have longer and stable life. Membrane ought to be easily manufactured with good salt rejection i.e. slightly permeable to salt. They ought to have high water flux i.e. highly permeable to water and fewer at risk for fouling. They ought to allow the flow of huge amount of water through the membrane relative to the degree they occupy. The membrane ought to be chemically, physically and thermally stable in saline waters. They have to robust enough to withstand high pressures and variable feed water quality. (57)

Membrane Fouling:

Membrane fouling is that the accumulation of materials at the surface or within the pores of a membrane, which decreases the permeate flux of the membrane (58). It is one of the serious problems faced by the membrane process. Fouling is that method leading to loss of the performance of a membrane because of deposition of suspended or dissolved substance on its pore (59).

RO Membrane Fouling

Membrane processes are frequently chosen since these applications achieve high removals of constituents like dissolved

solids, organic carbon, inorganic ions, and regulated and unregulated organic compounds. However, membrane fouling is a major obstacle for most applications in the drinking water industry (water treatment and desalination), especially when high concentrations of natural organic matter and inorganic constituents present. The fouling is classified as inorganic, organic, and biofouling (60). Therefore, membrane fouling is caused by dissolved inorganic (BaSO4, CaCO3, and CaSO4) or organic components (humic acid), collides (suspended particles), suspended solids or bacteria. The fouling deposits mainly consisted of a mixture of microorganism, organic matter, iron and phosphorous, in addition to the chemical constituents usually found in seawater or surface water. Despite its potential in water treatment, certain boundaries prohibit membrane process from large scale and continuous operation. RO membrane process achieves high removal of dissolved solids. (61)

Energy Requirement and Recovery:

The well-established seawater and brackish groundwater desalination techniques, no doubt, can be engaged to produce large amounts of good-quality water at a cost that as of today appears to be rationally quite competitive, but the main drawback of all such processes still remaining to be resolved is the high energy consumption. The energy for the desalination plant is generally supplied in the form of either electricity or steam. The electrical energy required is only for pumping the water to a relatively high operating pressure (62).

RO Plant

In RO process for episode of reverse osmosis, a very high pressure is to be applied on the concentrated solution. The primary energy use in RO structure is the power required to pump the feed water and is directly related to the feed pressure and flow rate. The high salt concentrations present in seawater require elevated hydrostatic pressures (up to 7000 kPa); the higher the salt concentration, the greater the pressure and pumping power required to produce a desired permeate flux (63).

Hydro turbines and impulse turbines are the two types of machines for recovering the residual energy available from the high-pressure feed stream. They have been used for several years in wide ranging applications, particularly in chemical industries (64).

MAJOR PATHOGENS AND PARASITES OF HEALTH CONCERN IN DRINKING WATER:

Bacterial Pathogens:

Vibrio cholerae. Vibrio cholerae is a gram-negative, coma or spiral shaped with a single polar flagellum, facultatively anaerobic bacterium which is a member of the aquatic microbial community and causes a disease called Cholera. This pathogen releases an enterotoxin that causes a mild to profuse diarrhea, vomiting, and a very rapid loss of body fluids. This pathogen is transmitted primarily via contaminated food and water. In aquatic environments, this pathogen generally attaches to solids and plankton such as cyanobacteria, zooplankton and algae and persists under the viable but non-culturable (VBNC) state (65).

Salmonella specie: An estimated 2–4 million human Salmonella infections occur every year in the United States. Salmonella typhi, an etiological agent of typhoid fever, produces an endotoxin. Salmonella is transmitted via contaminated food (e.g., chicken, eggs and milk) or drinking water. Typhoidal Salmonella species (e.g., S. typhi, S. paratyphi) are coupled with waterborne transmission whereas the non-typhoidal species (e.g., S. typhimurium) are associated with person-to-person contact and food-borne transmission (66).

Shigella specie: Shigella is a gram-negative, non-motile, rod shaped and a member of the Enterobacteriaceae. It is an etiologic agent of bacillary dysentery or shigellosis which affects the large intestine. There present four pathogenic species of Shigella: S. dysenteriae, S. flexneri, S. boydii, and S. sonnei (67). Shigella is transmitted via person-to-person contact and by the foodborne (e.g., salads or raw vegetables) and waterborne routes.

Escherichia coli. Escherichia coli or E.coli is a gram-negative facultative anaerobe which colonizes the gastrointestinal tract of humans and warm-blooded animals. While many its strains are harmless, some bear virulence factors and cause health problems (68). E. coli O157:H7 produces shiga-like toxins 1 and 2 encoded by stx1 and stx2 genes, respectively. It is the infectious agent of several waterborne outbreaks. (69)

Viral Pathogens

Hepatitis A virus (HAV) is a 27-nm RNA hepatovirus with a relatively short incubation period i.e. 2–6 weeks and displaying a fecal–oral transmission route, either by person-to-person contact, waterborne or foodborne transmission (70). It is an etiological agent of hepatitis. Waterborne and foodborne transmission of an infectious hepatitis have been documented worldwide (71).

Hepatitis E virus (HEV) is a single-stranded RNA virus. This virus is not well characterized due to the lack of a tissue culture cell line for assay but is now have been detected using molecular techniques (e.g., reverse transcriptase-polymerase chain reaction (RT-PCR) which show that HEV sequences are classified as four genotypes. Unlike HAV, it is mostly transmitted via fecally contaminated water while the person-to-person transmission is very low (70)

Rotaviruses: They are 70-nm particles in size containing double-stranded RNA surrounded with a double-shelled capsid. They are the main cause of infantile acute gastroenteritis in children younger than 2 years of an age and contribute significantly to childhood mortality in developing countries. It spreads mainly by the fecal–oral route, but a respiratory route has also been suggested (72).

Astroviruses. They are 27- to 34-nm in size, single-stranded RNA viruses with a characteristic star-like appearance. They spread

via person-to-person contacts and via contaminated drinking water or food. Patients have mild, watery diarrhea lasting from 3– 4 days or more for immunocompromised patients (73)

Protozoan Parasites:

Giardia: Giardia is a protozoan parasite which persists in an environment as ovoid cysts. Upon ingestion, the cysts develope into trophozoites which attach to the epithelial cells of the upper small intestine where they multiply and interfere with the absorption of fats and important other nutrients. Then they encyst as they reach the colon (74)

Cryptosporidium specie: Cryptosporidium parvum (genotype 2) is a coccidian protozoan parasite that infects both, the humans and animals while C. hominis (genotype 1) is only specific to humans. This parasite persists in the environment as a thick-walled oocyst which after ingestion, undergoes excystation to release infective sporozoites in the gastrointestinal tract (GIT). The infective dose of oocysts is relatively low, as 1–10 oocysts can initiate infection.

Cyclospora specie: Cyclospora cayetanensisis is a coccidian parasite which was earliest reported in the 1980s, and has been often mentioned in the literature as a "cyanobacterium-like body" (75). It infects the duodenum and jejunum parts in the gastrointestinal tract. It causes Cyclosporiasis which is an endemic in countries such as Nepal, Haiti, Indonesia, Guatemala, and Peru. (76)

Naegleria. Naegleria is a free-living protozoan that is commonly found in thermal spring waters, thermally polluted effluents, and drinking water supply wells with about 10% of the wells being positive for it. Naegleria fowleri is the causative agent of primary amoebic meningoencephalitis (PAME), a fatal disease first reported in the 1960s. (77).

WATERBORNE DISEASES

In Asia and Pacific region, about 7000 million natives are living without proper water supply system especially in rural areas. About 70 % of the population in Pakistan relies on ground water and high rate abstraction is causing decline in water table. Water contamination is caused by sewerage and industrial effluents, surface run-off and many human activities that alter the physical (color, taste, and odor) and chemical characteristics of water. In the reports, it is mentioned that in developing countries, water and sanitation are responsible for high illnesses. Frequent waterborne diseases and illness has increased the cost of medicinal facilities and poverty in rural areas. Reduced water quality is responsible for disease outbreaks in many parts of the developing world (78).

CLASSIFICATION OF WATER-RELATED DISEASES

•Those due to micro-organisms and chemicals in drinking water;

•Diseases like schistosomiasis which have part of their lifecycle in water;

•Diseases like malaria with water-related vectors;

•Drowning and some injuries;

•Diseases such as legionellosis, carried by aerosols containing certain micro-organisms (79, 80).

Cholera:

Cholera disease is primarily caused due to water pollution. In polluted, dirty and hard water, different bacteria are present which contaminate it and cause different diseases like cholera (81). Its symptoms include the stomach ulcer, severe dehydration, rapid diarrhea and sometimes, it leads to death. Main causes of the cholera are the bacteria available in polluted water. Sometimes, hard water contains cholera causing germs.

Dracunculiasis:

Dracunculiasisis guinea-worm crippling parasitic disease caused by Dracunculusmedinensis. It is transmitted absolutely when people drink stagnant water contaminated with parasite-infected water fleas. Dracunculiasis is not often fatal, but infected people become nonfunctional for weeks. It affects people in rural areas, deprived and isolated communities who depend mainly on open surface water sources. (82)

Typhoid:

Typhoid fever is a sort of enteric fever along with paratyphoid fever. The cause is the bacterium, named Salmonella Typhi, also known as Salmonella enterica serotype Typhi, growing in the stomach intestines and blood. Typhoid is spread by drinking or eating water or food contaminated with the feces of an infected person. Other symptoms are headache, stomach pain, loss of appetite, weakness, weight loss, constipation (83, 84)

Diarrhea:

Diarrhea is an increase in the rate of bowel movements or a decrease in the form of stool (greater looseness of stool). Although, the change in frequency of bowel movements and looseness of stools can vary independently of each other (83). The main cause for Diarrhea is drinking polluted and contaminated water with bacteria and chemicals.

Ulcers:

Peptic ulcers are open sores that develop on the inside lining of stomach and the upper portion of small intestine. The most general symptom of a peptic ulcer is stomach pain. Peptic ulcers include: Gastric ulcers that occur on the inside of the stomach. Duodenal ulcers that occur on the inside of the upper portion of small intestine i.e. duodenum (85).

Hepatitis:

Hepatitis is an inflammation of the liver tissue. Some people have no symptoms whereas others may develop yellow discoloration of the skin and whites of the eyes, poor appetite, vomiting, tiredness, abdominal pain, headache or diarrhea. Hepatitis can be temporary (acute) or long term (chronic). Hepatitis A and E are spread by contaminated food and water. (86)

Arsenicosis:

Arsenicosis is a chronic illness due to drinking water with high levels of arsenic over a long period of time (such as from 5 to 20 years). It is called an 'arsenic poisoning'. The WHO has recommended a limit of 0.01 mg/l of arsenic in drinking water. It results in various health effects including skin problems, skin cancer, cancers of the bladder, kidney and lung, and some diseases of the blood vessels. (87)

WATER QUALITY TESTS:

Temperature:

Water and air temperatures are measured by a shielded Celsius thermometer.

pH:

A relative measure of alkanity or acidity. In this test, a liquid reagent and a color comparator is used. (88)

Bacteriological tests:

These tests are performed generally to check for indicator bacteria (for example, total coliform, fecal coliform or Escherichia coli) and can indicate the presence or absence of disease-causing bacteria. However, there are many types of bacteriological test methods that cover a variety of bacteria. These tests are costly and are conducted only if they are absolutely essential.

Mineral tests:

Mineral tests can determine if the mineral content of the water is high enough to affect either health or the aesthetic and cleaning capacities of water. Mineral tests include calcium, magnesium, manganese, iron, copper, zinc and some others. An abundance of these minerals can cause water to be hard, plumbing and laundry stains, or bad odors.

Organic chemicals:

These tests are usually performed only if there is a reason to believe that a specific contaminant has infiltrated the water system (such as pesticides entering the water supply). Industrial and petroleum contamination can also be found through organic chemical testing.

Other tests:

Other tests may be conducted on radiological contaminants (radium and radon) or heavy metals like arsenic, mercury, lead or cadmium based on the suspected natural and anthropogenic (man-made) sources of such contaminants. (89)

RO bottled water with different names are consumed by people all over the world. After all the purification and filtration processes, still some bacteria are present therefore study is designed to check the quality of RO water in aspect of bacteria. The work was done by collecting couple of samples and which were analyzed by Spread Plate Technique.

INTRODUCTION:

Water is transparent fluid which forms the world's stream, lakes, oceans, rivers and the major constituent of the fluids of living things (1). It is important to life on Planet Earth. It is important for food production, industrial development, and ecological sustainability. The total amount of water reserved by our thirsty world was estimated at 3906 km3 in 1995. Estimates for 2025 predict a 50% increase for local, livestock, and industrial uses, leaving less water for irrigation and therefore threatening food production(2) shows the total water withdrawal by regions around the world and the predictions for 2025.

Early on, the ancient people (Egyptians, Greeks, and the Romains) were already worried with water hygiene and sanitation (3). A major innovation was due to Antony van Leeuwenhoek who, in 1684, called "animalcules" in water, and later on, in the nineteenth century, Robert Koch and Joseph Lister, showed that the "animalcules" were definitely microorganisms (4, 5). Another major breakthrough is when John Snow, a British physician, recognized a link between microorganisms and water-related diseases like cholera.

The mission for safe drinking water dates back to at least 4000 years ago when a medical theorist promoted the use of boiling, sunlight, charcoal filter, and copper vessels to treat water (6) and now drinking water safety is a universal concern. Polluted drinking water has the greatest influence on human health worldwide, particularly in developing countries. It is projected that more than one billion of the world's population does not have access to safe clean water and about 2.6 billion lack improved sanitation (7, 8, 9).

TYPES OF WATER:

Water and its significance have been highlighted upon since centuries. The colorless and tasteless mixture of life comes in many types.

Tap water:

Tap water is the water that you get straight from your faucet, it may or may not be suitable for drinking purposes. It is widely used for domestic routines such as cleaning, cooking, gardening and washing clothes. It must meet the guidelines set by the local urban bodies. Tap water is a type of water that you get directly from your tap.

Well water:

When it rains, water drops down and travels through the inner gaps of the soil, underneath the ground to form underground lakes. This occurs over a period of time. In rural areas, one of the prime sources of water is what is dug out from deep wells. Deep wells directly hit groundwater and bring it to the exterior from which people can yield their water. (10)

Rainwater:

In areas where rainfall is plentiful and common, rainwater can be a noble source of water supply. Rainwater has several benefits. It is free, fairly clean and usually reliable, even if it rains only once or twice a year, and a rainwater collecting system can be easily built and conserved at low cost. Although mainly found in rural areas, rainwater collecting can also be

useful in an urban state. (11)

Seawater:

Saltwater is plentiful in the surface of the planet. However, saltwater is currently not mainly useful when it comes to clean water supplies. Purification plants, while they do exist, are rare because the energy required for purification makes the process enormously expensive. (12)

Distilled or Demineralized Water:

Distilled water goes through a boiling procedure that strips away all of its impurities—and all of its healthy minerals. While it's clearly good to remove the pollutants in your diet, there are probable health concerns that come from eliminating the minerals from water. (13)

Waste Water:

Wastewater is any water that has been affected in feature by human actions. Wastewater can cultivate from agricultural activities, urban water use, and sewer invasion and storm water runoff just to name a few. Wastewater from a city is also called sewage. Most of us don't want to think about it, but at times the water that spins in the bowl ends up being cured and ends up in our taps. (14)

PROPERTIES OF WATER:

Chemical Properties of Water:

The chemical value of the aquatic location differs according to native geology, the climate, the distance from the ocean and the quantity of soil cover, etc. If superficial waters were totally unaffected by human actions, up to 90-99 per cent of worldwide freshwaters, dependent on the variable of interest, would have natural chemical deliberations appropriate for aquatic life and most human uses. Rare (between 1 and 10 per cent and between 90 and 99 per cent of the universal circulation) and very rare (< 1 per cent and > 99 per cent of the universal circulation) chemical circumstances in freshwaters, such as happen in salt lakes, hydrothermal waters, acid volcanic ponds, peat bogs, etc., usually make the water inappropriate for human. However, a variety of aquatic organisms have improved to these extreme situations. In many areas groundwater concentrations of total dissolved salts, fluoride, arsenic, etc., may also truly surpass maximum allowable concentrations (MAC) (15)

Physical Properties of Water:

Water is a minor solvent, occupying about 0.03 nm3 per molecule in the liquid form at room temperature and pressure, yet it is extremely cohesive because of the strong intermolecular connections (hydrogen bonds, or H-bonds) between the oxygen and hydrogen atoms. This is revealed in its high boiling point, the great amount of heat required to vaporize it, and its high surface tension. Replacement of one or both of the hydrogens dramatically fails these intermolecular interactions, decreasing the magnitude of these measures. The strong cohesive connections in water also result in:

(1) A high viscosity, since for a liquid to flow interactions between adjacent molecules must always be broken; and

(2) A high specific heat capacity – the capability to store a large quantity of potential energy for a given addition in kinetic energy (temperature). (16)

Biological Properties of Water:

The growth of biota (flora and fauna) in superficial waters is ruled by a diversity of ecological conditions which define the choice of species as well as the physiological presentation of individual organisms. The primary construction of organic matter, in the form of phytoplankton and macrophytes, is most concentrated in lakes and pools and usually more partial in rivers. The degradation of organic materials and the related bacterial invention can be a long-term procedure which can be important in groundwater and deep lake waters which are not directly visible to sunlight. In distinction to the chemical feature of water bodies, which can be measured by appropriate analytical means, the explanation of the biological quality of a water body is a mixture of qualitative and quantitative classification. Biological monitoring can usually be carried out at two dissimilar levels. (15)

WATER SOURCES:

Groundwater:

Groundwater is that water which is present beneath Earth's surface in soil pore spaces and in the fractures of rock formations. A unit of rock or an unconsolidated deposit is called an aquifer when it can yield a usable amount of water. The depth at which soil pore spaces or fractures and voids in rock become fully saturated with water is termed as water table. Groundwater is recharged from the surface. (17)

Groundwater is commonly cheaper, more convenient and less vulnerable to pollution than surface water. Therefore, it is used for public water supplies. For example, groundwater provides the largest source of usable water storage in the United States, and California annually withdraws the largest amount of groundwater of all the states.(18) Underground reservoirs contain far more water than the capacity of all surface reservoirs and lakes in the North America, including the Great Lakes. Many municipal water supplies are derived solely from groundwater. (19)

Upland lakes and reservoirs:

Typically present in the headwaters of river systems, upland reservoirs are usually sited up above any human habitation and may be surrounded by a protective zone to restrict the opportunities for contamination. Bacteria and pathogen levels are usually low however some bacteria, protozoa or protoctist will be present. Where uplands are forested or peaty, humic acids may color the water. Many upland sources have low pH which needs adjustment.

Rivers, canals and low land reservoirs:

Low land surface waters will have a significant bacterial load and may also contain algae, suspended solids and a variety of dissolved constituents. (20)

Atmospheric water generation:

A device, named an atmospheric water generator (AWG) that extracts water from humid ambient air. Water vapor in the air can be extracted by condensation - cooling the air below its dew point, exposing the air to desiccants, or pressurizing the air. (21)

Rain water harvesting or fog collection:

Rainwater harvesting is a type of harvest in which the rain drops are collected and stored for the future use, rather than allowing it to run off. (22) The harvested water can also be used as drinking water, longer-term storage, and for other purposes such as groundwater recharge. Rainwater harvesting is one of the simplest and oldest methods of self-supply of water for households usually financed by the user. (23)

Desalination of seawater by distillation:

Desalination is a process that takes away mineral components from saline water. More generally, desalination refers to the removal of salts and minerals from a target substance (24) as in soil desalination, which is an issue for agriculture. (25).

Surface water:

Water on the surface of continents such as in a river, lake, or wetland is surface water. It can be contrasted with atmospheric water and groundwater.

Non-saline surface water uses is replenished by precipitation and by recruitment from ground-water. (26)

WATER PURIFICATION:

Water purification is the process of removing undesirable substances like chemicals, biological contaminants, suspended solids, and gases from water. The aim is to produce water fit for specific purposes. Most water is purified and disinfected for human ingestion (drinking water), but water purification may also be carried out for a variety of other purposes such medical, pharmacological, chemical, and industrial applications. The WHO estimates that 94% of these diarrheal disease cases are preventable through modifications to the environment, including access to safe water. (27) Simple techniques for treating water at home, such as chlorination, filters, and solar disinfection and for storing it in safe containers could save a huge number of lives each year. (28) Reducing deaths from waterborne diseases is a major public health goal in developing countries. (20)

Screening:

A screen filter is a type of [filtration of water] employing a rigid or versatile screen to separate sand and other fine particles out of water for irrigation or industrial applications. These are typically not recommended for filtering out organic matter such as algae, since these types of contaminants may be extruded into spaghetti-like strings through the filter if enough pressure drops occurs across the filter surface. Typical screen materials include stainless steel (mesh), polypropylene, nylon and polyester. (29)

Pre chlorination and Primary Disinfection:

Many serious diseases, such as cholera, are caused by drinking water that contains parasitic microorganisms. We need to treat the drinking water that is suitable to us so we can stay healthy and so we don't develop diseases or health problems through our drinking water. Water disinfection is the removal, deactivation or killing of pathogenic microorganisms.

Microorganisms are deactivated or destroyed, resulting in termination of growth and reproduction. When microorganisms are not removed from drinking water, drinking water intake will cause people to fall ill. For chemical disinfection of water, the following disinfectants may be used:

- Chlorine (Cl2)
- Chlorine dioxide (ClO2)
- Hypo chlorite (OCl-)
- Ozone (O3)
- Halogens: bromine (Br2), iodine (I)
- Bromine chloride (BrCl)
- Metals: copper (Cu2+), silver (Ag+)
- Potassium permanganate (KMnO4)
- Phenols
- Alcohols
- Soaps and detergents
- Kwartair ammonium salts
- Hydrogen peroxide
- Several acids and bases

For physical disinfection of water, the listed disinfectants can be used:

- Ultraviolet light (UV)
- Electronic radiation
- Gamma rays
- Sounds
- Heat (30)

Coagulation and flocculation:

One of the primary steps in a conventional water purification process is the addition of chemicals to assist in the removal of particles suspended in water. Particles can be inorganic such as clay and slit or organic such as algae, bacteria, viruses, protozoa and natural organic matter. Inorganic and organic particles contribute to the color and turbidity in water. (31) In coagulation, a liquid coagulant, such as aluminum sulfate, is added to the water to attract suspended particles. The water is then gently

stirred to permit the particles to come together and form larger particles (flocculation), which may then be removed by sedimentation, settlement or filtration. The amount of coagulant needed will depend on the nature of the contaminating chemical compounds and solids. (32)

Sedimentation:

After the bulk of the suspended particles have settled, water exits the flocculation basin and enters a sedimentation basin. Sedimentation basins move treated waters along through the purification process while allowing remaining particles to settle. Sludge forms that appear on the floor of the tank are removed and treated. (33) Typical detention times for sedimentation vary from 1.5 to 4 hours and basin depths vary from 10 to 15 feet (3 to 4.5 meters).(31)

Filtration:

A rapid sand filter is the most common type of filter. Water moves vertically through sand which often has a layer of activated carbon or anthracite coal on the sand. The top layer removes organic compounds that contribute to taste and odor. The space between sand particles is larger than the tiny suspended particles therefore simple filtration is not enough. Most particles pass through surface layers but are trapped in pore spaces or adhere to sand particles. (34)

Slow sand filters can be used where there's sufficient land and space, as the water flows very slowly through the filters. These filters rely on biological treatment processes for their action instead of physical filtration. They are carefully constructed using graded layers of sand, with the coarsest sand, along with some gravel, at the bottom and finest sand at the top. (35)

Secondary Disinfection:

Disinfection is accomplished both by filtering out harmful micro-organisms and conjointly by adding disinfectant chemicals. Water is disinfected to kill pathogens which pass through the filters and to provide a residual dose of disinfectant to kill or inactivate potentially harmful micro-organisms in the storage and distribution systems. Possible pathogens include viruses, bacteria, including Salmonella, Cholera, Campylobacters and Shigella, and protozoa, including Giardia lamblia and other cryptosporidia. After the introduction of any chemical disinfecting agent, the water is usually held in temporary storage – often called a contact tank or clear well – to allow the disinfecting action to complete. (36)

Chlorine disinfection:

The most used disinfection method involves some form of chlorine or its compounds such as Chloramine or chlorine dioxide. Chlorine is a strong oxidant that rapidly kills many harmful micro-organisms and help keep the water clean until distribution.

Ozone disinfection:

Ozone is an unstable molecule which readily gives up one atom of oxygen providing a powerful oxidizing agent that is toxic to most waterborne organisms. It is a very strong, board spectrum disinfectant that is widely used in Europe. It is an efficient method to inactivate harmful protozoa that form cysts (29).

WATER POLLUTION:

Water pollution is the contamination of natural water bodies by radioactive, chemical, physical or pathogenic microbial substances. Adverse alteration of water quality presently produces large scale illness and deaths, accounting for approximately 50 million deaths per year worldwide, most of these deaths occurring in Africa and Asia (37). In China, as an example, about 75 percent of the population (or 1.1 billion people) are without access to pure drinking water, according to China's own standards (38). Widespread consequences of water pollution upon ecosystems include species mortality, biodiversity reduction and loss of ecosystem services.

TYPES OF WATER POLLUTION:

There are many types of water pollution because water comes from many sources. Here are few types of water pollution:

Chemical pollutants:

They are generally atoms or molecules, which have been discharged into natural water bodies, usually by activities of humans. Common examples of such chemical water pollutants are mercury emanating from mining activity, some nitrogen compounds utilizing in agriculture, chlorinated organic molecules arising from sewage or water treatment plants (39) or various acids which are the externalities of various manufacturing activities.

Physical pollutants :

Physical water pollutants are either (a) much more larger particles or (b) physical factors like temperature change, both of which while not typically toxic, cause a variety of harmful effects. The absolute physical pollutants are (a) excessive sediment load, mostly arising from over-intense land use practices and (b) rubbish discarded from anthropogenic activity (e.g. plastic bags, bottles). Whereas these materials are not so harmful to human health as chemicals or pathogens, they comprise the majority of visual impact of water pollution.

Radioactive substances

They are really merely a special sub-class of chemical pollutants, and by mass represent the smallest of the contributors to water pollution; however, their potential for harm allows recognition as a separate class. In fact, most discharge of radiation is not from the negligible escape from nuclear power plants however but rather arises from agricultural practices like tobacco farming, where radioactive contamination of phosphate fertilizer is a common technique of introduction of radioactive materials into the environment.

Microbial Pollutants:

Common pathogenic microorganisms introduced into natural

water bodies are pathogens from untreated sewage or surface runoff from intensive livestock grazing. One of the most common disease agents is a Giardia lamblia, a parasitic protozoan present in fecal material of many fauna including humans; this pathogen is particularly insidious, due to its resistance to conventional sewage treatment. Giardia and other protozoan and bacteria are important causes of illness and mortality in developing countries where population density, water insufficiency and inadequate sewerage treatment combine to occasion widespread parasitic and bacterial disease. (40)

Pesticide pollution of water

In terms of pesticide water pollution, each pesticide has unique properties and many variable factors affect its risk, such as the active ingredients, contaminants and additives as well as any degradate formed during chemical, photochemical, microbial degradation of active ingredients. The increased use of pesticides has been accompanied by the growing presence in soil of a large number of transformation products (TPs) for a wide variety of pesticides. (41)

Salt loads to water:

The quantity of dissolved salts in water is called salinity. Freshwater bodies can receive salt through distinct pathways, for example through direct surface runoff from saline lands, subsurface drainage of saline waters to fresh water bodies, or the interception of saline stores because of elevation of the ground water table that also may recharge surface waters. (42)

Agricultural erosion and sediment loads to water:

Agricultural activities contribute to enhanced soil erosion and sediment loads in river systems. Pollutants may be released from sediments when the environment (e.g. redox potential or pH) changes. Therefore, additionally to the dissolved pollutants that reach water bodies, sediments may carry more pollutants to aquatic ecosystems, and are effectively the most important pathway for some sorts of pollutants with low solubility such as phosphates (43), some metals (44) and pesticides (45).

Organic Matter:

Pollution by organic matter is increasing because of increasing municipal and industrial wastewater discharge, the intensification of agriculture (including animal farming) and reduction in river dilution capacity due to weather change and water extractions. Water pollution by organic matter from intensive livestock farming is now significantly more widespread (46)

BOTTLED WATER:

Today, bottled water is primarily used for drinking but has also been considered for the preparation of baby formula, cleaning contact lens or used in humidifiers (47). Bottled water is perceived by consumers as being of better quality and better taste than usual tap water. However, the higher quality of bottled water is sometimes a thought. Several researchers reported that bottled water is of no better quality than municipal drinking water. Furthermore, it is also hundred times more expensive than tap water.

In 2011, the highest consumers were Italy (179 L/person per year) and Germany (171 L/ person per year) while the lowest was Finland (18 L/person per year) (48)

TYPES OF BOTTLED WATER:

There are various kinds of bottled water and these are not directly comparable to one another (49).

Spring water:

Water is derived from an underground formation that flows naturally to the earth surface. In 2011, spring water accounted for 14% of total sales in Europe, as compared to 83% for mineral water. In United States, approximately 25% of the bulk water is spring water. This water might be disinfected with ozone.

Sparkling Bottled Water:

It is “naturally” carbonated with carbon dioxide. It can also be created bubbly by adding dissolved carbon dioxide. Some brands of bottled water (e.g., Perrier) are made bubbly by harvesting the “natural” carbon dioxide at the source and re injecting it into the water during the bottling operation.

Mineral water:

It contains dissolved cat ions and anions (e.g., Ca2+, Mg2+, Na+, HCO3−). The mineral content varies from one supply to another and may be sometimes not up to the mark of community water supplies. Mineral water is drawn from underground supply that contains at least 250mg/L of total dissolved solids. Minerals and trace elements must come from the supply of the underground water and cannot be supplemented later. Natural mineral water is one that is drawn from a protected supply. It becomes enriched with minerals because it flows through underground formations. According to European Union regulations, manufacturers are not allowed to alter the mineral content of this type of water nor disinfect it to alter the microbial flora of this natural resource. The sole treatments allowed are filtration to get rid of particles or iron, manganese, sulfur or arsenic along with the addition or removal of CO2. Furthermore, it must be monitored to prevent contamination from allochthonous microorganisms together with pathogens and parasites. (50)

Artesian Water:

Water from a well tapping a confined aquifer in which the water level stands at some height above the top of the aquifer. (51)

Purified Water:

Water that has been resulted by distillation, deionization, reverse osmosis or other different suitable processes may be labeled as purified bottled water. Other suitable product names for bottled water treated by one of the above processes can include “distilled water” if it is produced by distillation," deionized water “if it is produced by deionization or “reverse osmosis water” if the process used is reverse osmosis. Alternatively, “ drinking water” may be used with the blank being filled in with one of the

terms defined in this paragraph (e.g., "purified drinking water" or "distilled drinking water") (52).

Reverse Osmosis (RO) is a method that employs the use of a semi-permeable spiral wound membranes to separate and eliminate dissolved solids, organic, pyrogens, submicron mixture matter, color, nitrate and bacteria from water. Feed water is conveyed under pressure through the semi porous membrane; where water permeates the minute pores of the membrane and is delivered as purified water referred to as permeate water. Impurities within the water are concentrated in the reject stream and flushed out of the drain is referred as reject water. These membranes are semi-permeable and reject the salt ions whereas allowing the water molecules to pass. The materials used for RO membranes are manufactured from cellulose acetate, polyamides and different polymers. The membrane consists of hollow-fiber, spiral-wound used for treatment; rely on the feed water composition and the operation parameters of the plant. Reverse Osmosis (RO) is a membrane based method technology used for desalination. Membrane-based seawater desalination and wastewater reuse are widely considered as promising solutions to augment water supply and alleviate water scarcity (53). The most common membrane processes used are the reverse osmosis (RO) and the electro dialysis (ED) used for brackish water desalination, but only RO competes with distillation processes in seawater desalination (54).

Scope of Reverse Osmosis:

The process has additionally been applied to treat municipal wastewater. Since standard municipal treatment processes do not remove dissolved solids, however RO process is used for the elimination of dissolved solids. RO is widely used as a separation technique in chemical and environmental engineering to get rid of organics and organic pollutants present in wastewater. It is seen from literature review that Reverse Osmosis (RO) processes have been widely used for separation and concentration (recovery) of solutes in many fields. (55)

Reverse Osmosis Process Description:

The RO process is easy in design consisting of feed, permeate and reject stream. For feed water it is necessary to perform pretreatment in order to get rid of inorganic solids and suspended solid and using high pressure pump given feed through semi permeable membrane. Relying upon the permeate where ever it is used necessary post treatment is given.

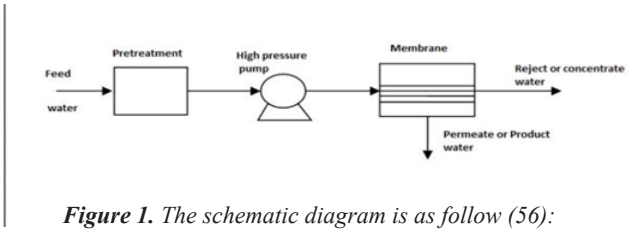


Figure 1. *The schematic diagram is as follow (56):*

Reverse Osmosis Requisites:

An RO desalination plant basically consists of 4 major systems: (a) Pretreatment system, (b) High pressure pumps, (c) Membrane systems, and (d) Post-treatment. Pre-treatment system is provided to get rid of all suspended solids in order that salt precipitation or microbial growth does not occur on the membranes. Pre-treatment could involve typical strategies sort of a chemical feed followed by coagulation/ flocculation/ sedimentation, and sand filtration or membrane processes i.e. micro filtration (MF) and ultra filtration (UF). High-pressure pumps supply the pressure required to change the water to pass through the membrane and have the salt rejected. The pressure ranges from 17 bar to 27 bar for brackish water, and from 52 bar to 69 bar for seawater. Membrane systems accommodate a pressure vessel and a semi-permeable membrane within that permits the feed water to pass through it. RO membranes for desalination usually available in 2 types: Spiral wound and Hollow fiber. Relying upon water quality of permeate and use of permeate; post treatment may consists of adjusting the pH and disinfection (56).

Membrane Characteristics:

The membrane ought to be cheap, have longer and stable life. Membrane ought to be easily manufactured with good salt rejection i.e. slightly permeable to salt. They ought to have high water flux i.e. highly permeable to water and fewer at risk for fouling. They ought to allow the flow of huge amount of water through the membrane relative to the degree they occupy. The membrane ought to be chemically, physically and thermally stable in saline waters. They have to robust enough to withstand high pressures and variable feed water quality. (57)

Membrane Fouling:

Membrane fouling is that the accumulation of materials at the surface or within the pores of a membrane, which decreases the permeate flux of the membrane (58). It is one of the serious problems faced by the membrane process. Fouling is that method leading to loss of the performance of a membrane because of deposition of suspended or dissolved substance on its pore (59).

RO Membrane Fouling

Membrane processes are frequently chosen since these applications achieve high removals of constituents like dissolved solids, organic carbon, inorganic ions, and regulated and unregulated organic compounds. However, membrane fouling is a major obstacle for most applications in the drinking water industry (water treatment and desalination), especially when high concentrations of natural organic matter and inorganic constituents present. The fouling is classified as inorganic, organic, and biofouling (60). Therefore, membrane fouling is caused by dissolved inorganic (BaSO4, CaCO3, and CaSO4) or organic components (humic acid), collides (suspended particles), suspended solids or bacteria. The fouling deposits mainly consisted of a mixture of microorganism, organic matter, iron and phosphorous, in addition to the chemical constituents usually found in seawater or surface water. Despite its potential in water treatment, certain boundaries prohibit membrane process from large scale and continuous operation. RO membrane

process achieves high removal of dissolved solids. (61)

Energy Requirement and Recovery:

The well-established seawater and brackish groundwater desalination techniques, no doubt, can be engaged to produce large amounts of good-quality water at a cost that as of today appears to be rationally quite competitive, but the main drawback of all such processes still remaining to be resolved is the high energy consumption. The energy for the desalination plant is generally supplied in the form of either electricity or steam. The electrical energy required is only for pumping the water to a relatively high operating pressure (62).

RO Plant

In RO process for episode of reverse osmosis, a very high pressure is to be applied on the concentrated solution. The primary energy use in RO structure is the power required to pump the feed water and is directly related to the feed pressure and flow rate. The high salt concentrations present in seawater require elevated hydrostatic pressures (up to 7000 kPa); the higher the salt concentration, the greater the pressure and pumping power required to produce a desired permeate flux (63).

Hydro turbines and impulse turbines are the two types of machines for recovering the residual energy available from the high-pressure feed stream. They have been used for several years in wide ranging applications, particularly in chemical industries (64).

MAJOR PATHOGENS AND PARASITES OF HEALTH CONCERN IN DRINKING WATER:

Bacterial Pathogens:

Vibrio cholerae. Vibrio cholerae is a gram-negative, coma or spiral shaped with a single polar flagellum, facultatively anaerobic bacterium which is a member of the aquatic microbial community and causes a disease called Cholera. This pathogen releases an enterotoxin that causes a mild to profuse diarrhea, vomiting, and a very rapid loss of body fluids. This pathogen is transmitted primarily via contaminated food and water. In aquatic environments, this pathogen generally attaches to solids and plankton such as cyanobacteria, zooplankton and algae and persists under the viable but non-culturable (VBNC) state (65).

Salmonella specie: An estimated 2–4 million human Salmonella infections occur every year in the United States. Salmonella typhi, an etiological agent of typhoid fever, produces an endotoxin. Salmonella is transmitted via contaminated food (e.g., chicken, eggs and milk) or drinking water. Typhoidal Salmonella species (e.g., S. typhi, S. paratyphi) are coupled with waterborne transmission whereas the non-typhoidal species (e.g., S. typhimurium) are associated with person-to-person contact and food-borne transmission (66).

Shigella specie: Shigella is a gram-negative, non-motile, rod shaped and a member of the Enterobacteriaceae. It is an etiologic agent of bacillary dysentery or shigellosis which affects the large intestine. There present four pathogenic species of Shigella: S. dysenteriae, S. flexneri, S. boydii, and S. sonnei (67). Shigella is transmitted via person-to-person contact and by the foodborne (e.g., salads or raw vegetables) and waterborne routes.

Escherichia coli. Escherichia coli or E.coli is a gram-negative facultative anaerobe which colonizes the gastrointestinal tract of humans and warm-blooded animals. While many its strains are harmless, some bear virulence factors and cause health problems (68). E. coli O157:H7 produces shiga-like toxins 1 and 2 encoded by stx1 and stx2 genes, respectively. It is the infectious agent of several waterborne outbreaks. (69)

Viral Pathogens

Hepatitis A virus (HAV) is a 27-nm RNA hepatovirus with a relatively short incubation period i.e. 2–6 weeks and displaying a fecal–oral transmission route, either by person-to-person contact, waterborne or foodborne transmission (70). It is an etiological agent of hepatitis. Waterborne and foodborne transmission of an infectious hepatitis have been documented worldwide (71).

Hepatitis E virus (HEV) is a single-stranded RNA virus. This virus is not well characterized due to the lack of a tissue culture cell line for assay but is now have been detected using molecular techniques (e.g., reverse transcriptase-polymerase chain reaction (RT-PCR) which show that HEV sequences are classified as four genotypes. Unlike HAV, it is mostly transmitted via fecally contaminated water while the person-to-person transmission is very low (70)

Rotaviruses: They are 70-nm particles in size containing double-stranded RNA surrounded with a double-shelled capsid. They are the main cause of infantile acute gastroenteritis in children younger than 2 years of an age and contribute significantly to childhood mortality in developing countries. It spreads mainly by the fecal–oral route, but a respiratory route has also been suggested (72).

Astroviruses. They are 27- to 34-nm in size, single-stranded RNA viruses with a characteristic star-like appearance. They spread via person-to-person contacts and via contaminated drinking water or food. Patients have mild, watery diarrhea lasting from 3– 4 days or more for immunocompromised patients (73)

Protozoan Parasites:

Giardia: Giardia is a protozoan parasite which persists in an environment as ovoid cysts. Upon ingestion, the cysts develope into trophozoites which attach to the epithelial cells of the upper small intestine where they multiply and interfere with the absorption of fats and important other nutrients. Then they encyst as they reach the colon (74)

Cryptosporidium specie: Cryptosporidium parvum (genotype 2) is a coccidian protozoan parasite that infects both, the humans and animals while C. hominis (genotype 1) is only specific to humans. This parasite persists in the environment as a thick-walled oocyst which after ingestion, undergoes excystation to

release infective sporozoites in the gastrointestinal tract (GIT). The infective dose of oocysts is relatively low, as 1–10 oocysts can initiate infection.

Cyclospora specie: Cyclospora cayetanensisis is a coccidian parasite which was earliest reported in the 1980s, and has been often mentioned in the literature as a "cyanobacterium-like body" (75). It infects the duodenum and jejunum parts in the gastrointestinal tract. It causes Cyclosporiasis which is an endemic in countries such as Nepal, Haiti, Indonesia, Guatemala, and Peru. (76)

Naegleria. Naegleria is a free-living protozoan that is commonly found in thermal spring waters, thermally polluted effluents, and drinking water supply wells with about 10% of the wells being positive for it. Naegleria fowleri is the causative agent of primary amoebic meningoencephalitis (PAME), a fatal disease first reported in the 1960s. (77).

WATERBORNE DISEASES

In Asia and Pacific region, about 7000 million natives are living without proper water supply system especially in rural areas. About 70 % of the population in Pakistan relies on ground water and high rate abstraction is causing decline in water table. Water contamination is caused by sewerage and industrial effluents, surface run-off and many human activities that alter the physical (color, taste, and odor) and chemical characteristics of water. In the reports, it is mentioned that in developing countries, water and sanitation are responsible for high illnesses. Frequent waterborne diseases and illness has increased the cost of medicinal facilities and poverty in rural areas. Reduced water quality is responsible for disease outbreaks in many parts of the developing world (78).

CLASSIFICATION OF WATER-RELATED DISEASES

•Those due to micro-organisms and chemicals in drinking water;

•Diseases like schistosomiasis which have part of their lifecycle in water;

•Diseases like malaria with water-related vectors;

•Drowning and some injuries;

•Diseases such as legionellosis, carried by aerosols containing certain micro-organisms (79, 80).

Cholera:

Cholera disease is primarily caused due to water pollution. In polluted, dirty and hard water, different bacteria are present which contaminate it and cause different diseases like cholera (81). Its symptoms include the stomach ulcer, severe dehydration, rapid diarrhea and sometimes, it leads to death. Main causes of the cholera are the bacteria available in polluted water. Sometimes, hard water contains cholera causing germs.

Dracunculiasis:

Dracunculiasisis guinea-worm crippling parasitic disease caused by Dracunculusmedinensis. It is transmitted absolutely when people drink stagnant water contaminated with parasite-infected water fleas. Dracunculiasis is not often fatal, but infected people become nonfunctional for weeks. It affects people in rural areas, deprived and isolated communities who depend mainly on open surface water sources. (82)

Typhoid:

Typhoid fever is a sort of enteric fever along with paratyphoid fever. The cause is the bacterium, named Salmonella Typhi, also known as Salmonella enterica serotype Typhi, growing in the stomach intestines and blood. Typhoid is spread by drinking or eating water or food contaminated with the feces of an infected person. Other symptoms are headache, stomach pain, loss of appetite, weakness, weight loss, constipation (83, 84)

Diarrhea:

Diarrhea is an increase in the rate of bowel movements or a decrease in the form of stool (greater looseness of stool). Although, the change in frequency of bowel movements and looseness of stools can vary independently of each other (83). The main cause for Diarrhea is drinking polluted and contaminated water with bacteria and chemicals.

Ulcers:

Peptic ulcers are open sores that develop on the inside lining of stomach and the upper portion of small intestine. The most general symptom of a peptic ulcer is stomach pain. Peptic ulcers include: Gastric ulcers that occur on the inside of the stomach. Duodenal ulcers that occur on the inside of the upper portion of small intestine i.e. duodenum (85).

Hepatitis:

Hepatitis is an inflammation of the liver tissue. Some people have no symptoms whereas others may develop yellow discoloration of the skin and whites of the eyes, poor appetite, vomiting, tiredness, abdominal pain, headache or diarrhea. Hepatitis can be temporary (acute) or long term (chronic). Hepatitis A and E are spread by contaminated food and water. (86)

Arsenicosis:

Arsenicosis is a chronic illness due to drinking water with high levels of arsenic over a long period of time (such as from 5 to 20 years). It is called an 'arsenic poisoning'. The WHO has recommended a limit of 0.01 mg/l of arsenic in drinking water. It results in various health effects including skin problems, skin cancer, cancers of the bladder, kidney and lung, and some diseases of the blood vessels. (87)

WATER QUALITY TESTS:

Temperature:

Water and air temperatures are measured by a shielded Celsius thermometer.

pH:

A relative measure of alkanity or acidity. In this test, a liquid

reagent and a color comparator is used. (88)

1.9.3 Bacteriological tests:

These tests are performed generally to check for indicator bacteria (for example, total coliform, fecal coliform or Escherichia coli) and can indicate the presence or absence of disease-causing bacteria. However, there are many types of bacteriological test methods that cover a variety of bacteria. These tests are costly and are conducted only if they are absolutely essential.

Mineral tests:

Mineral tests can determine if the mineral content of the water is high enough to affect either health or the aesthetic and cleaning capacities of water. Mineral tests include calcium, magnesium, manganese, iron, copper, zinc and some others. An abundance of these minerals can cause water to be hard, plumbing and laundry stains, or bad odors.

Organic chemicals:

These tests are usually performed only if there is a reason to believe that a specific contaminant has infiltrated the water system (such as pesticides entering the water supply). Industrial and petroleum contamination can also be found through organic chemical testing.

Other tests:

Other tests may be conducted on radiological contaminants (radium and radon) or heavy metals like arsenic, mercury, lead or cadmium based on the suspected natural and anthropogenic (man-made) sources of such contaminants. (89)

RO bottled water with different names are consumed by people all over the world. After all the purification and filtration processes, still some bacteria are present therefore study is designed to check the quality of RO water in aspect of bacteria. The work was done by collecting couple of samples and which were analyzed by Spread Plate Technique.

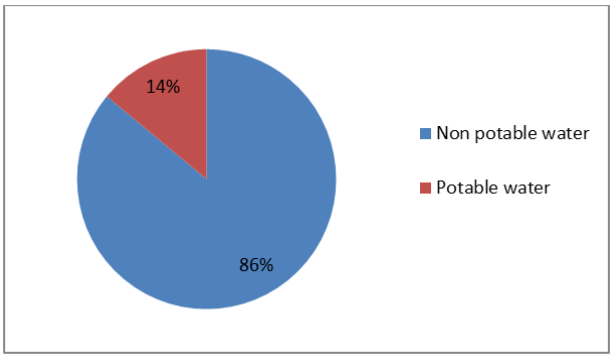
SAMPLE NO.	MACCONKEY (cfu/ml)
01	$2x10^4$
02	No growth
03	$3x10^4$
04	No growth
05	$2x10^4$
06	$1.88x10^6$
07	TFTC
08	$3.48x10^6$
09	$2.88x10^6$
10	TMTC
11	$2x10^6$
12	$4.56x10^6$
13	$3.92x10^6$
14	$3.36x10^6$
15	1.68x106

Table 3.1. *Determination of Gram Negatives in water samples.*

SERIAL NO.	NON POTABLE WATER	TOTAL SAMPLE	SAMPLE POSITIVE FOR GRAM -VE	% RATIO
01		15	13	86%
	POTABLE WATER		SAMPLE NEGATIVE FOR GRAM -VE	
02		15	2	13%

k)
Count

Table 3.2. *Representing % ratio of potable and non potable water.*



Graph 1. *Graphical representation showing percentage of potable water.*

RESULT:

Bacteriological safety of Reverse Osmosis treated drinking water was determined by the presence of Gram negative bacteria. They are pink or red in color, rod shaped, some are lactose fermenter organisms. It includes Klebsiella, E.coli., Vibrio Cholerae, etc.

13 out of total 15 samples show negative results on MacConkey. 86% water samples contain gram negative organisms in high amount which is not suitable for drinking purpose because ram negatives cause serious gastrointestinal problems if come from the fecal matter such as pathogenic strain of E.coli 0157:H7. On the other hand, 2 out of 15 samples show positive results on MacConkey agar. In sample 2 and sample 4 no gram negative organisms were isolate. 13% samples have no CFU count thus they are potable, safe for human consumption.

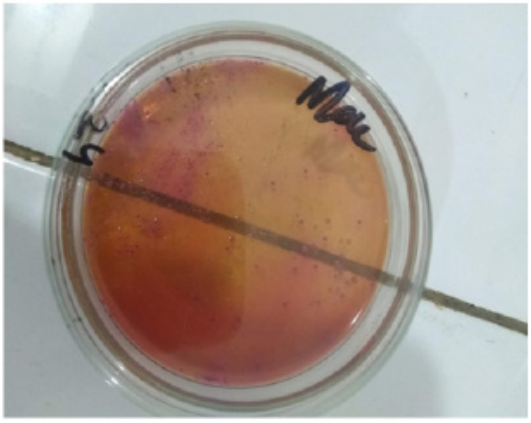


Fig.2: *Bacterial colonies on MaConkey agar*

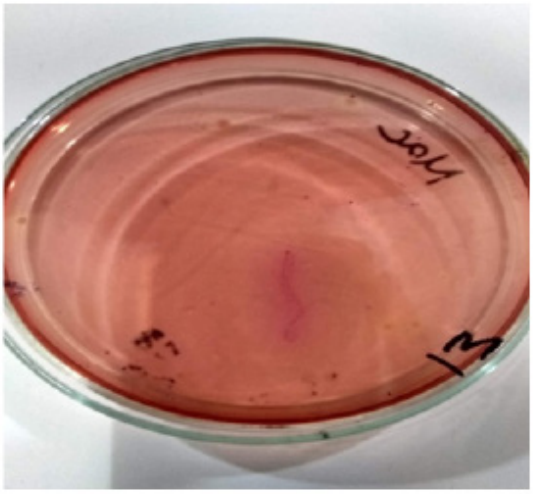


Fig.3: *Bacterial colonies on MaConkey agar*

DISCUSSION:

Water is an essential element of all life forms and this study is conducted from the RO water bottles in Karachi. Karachi city is a highly urbanized area of Pakistan, where different brands of bottled water are marketed. Bottled water should be of better microbiological quality for the consumption of drinking purposes. One of the greatest concerns of the water consumers with respect to the quality of drinking water is contaminated with pathogenic microorganisms. Therefore, it is a matter of concern that only about one-fifth of the bottled water tested was suitable for drinking in the present study. In other more studies done in India, the acceptability of bottled water ranged from 60% (90), 83% (91), 90% (92), and even 100% (93). In researches done in other parts of Asia, the acceptability of drinking water samples ranged from 50% (94), 64.2% (95), 97.1% (96) and a study done in Iran even reported 100% (97). In studies done in Africa, it was 67.4% (98), 70% (99), 71.4% (100), 75% (101), 85% (102), 88.9% (103), 90% (104), 94% (105) and a work done in Uganda (106) and Nigeria (107) reporting 100%.

A study done in Pakistan, showed the TVC in CFU/ml was <1 in 40%, 15–20 in 24%, 20–200 in 10%, 200–300 in 13%, and >300 in 13% (95). In a study done in different parts of North India, around 2% of the samples analyzed had bacterial counts of more than 1000 CFU/ml (91). The contamination level of drinking water samples reported in these above-mentioned studies was therefore much more than our observations. However, another study done in Chennai, India, reported that bacterial counts ranged from 0 to 41 CFU/ml among all the samples tested, which was much lesser than that observed in the present study in few samples (93). The presence of heterotrophic bacteria in the bottled water causes significant health risk especially for children, elderly, and immunocompromised individuals (108). Its presence in bottled water is also an indicator of poor practices performed in the manufacturing processes.

The kind of bacteria found in the bottled water has previously been reported were multiple drug resistant in samples collected from different parts of India. Safety of bottled water for consumption can be ensured with sealed caps on bottles, hygienic filling systems, the minimal time between production and sale, and use of nonreturnable plastic containers (90). It was experienced in a Nigerian study that contamination of packaged water aggravates as the product moves down the distribution chain (109). Evaluation of water quality is therefore required not only at various stages of production but also in postproduction stage (93). This will ensure advancement in transportation and storage practices in the supply chain. Government and other authorities need to intensify surveillance activities of water treatment processes at packaged water industries. This will ensure that strict hygienic measures are followed, resulting in safe and quality bottled water being available at various retail outlets for public use.

Among the physical parameters, turbidity was absent in all the water samples tested in this study. This was comparable to the observations of a study done in Ghana (102). Turbidity of water depends on the amount of particulate matter present in it. This interferes with the disinfection process of water. It also affects the taste, odor, and the color of the water (109).

CONCLUSION:

Water is a need of every person, animal and plant even the constituents by which Earth is made, water is one of them. For the drinking purpose, domestic use or for manufacturing products in industries, water free from microorganisms is require. Now, the use of bottled RO water is increasing among people so its quality check is necessary. Gram negatives shouldn't observed but they were present in 86% of samples which may cause severe illness in humans therefore drinking water's quality tests should have to perform even after treatment also post treatment such as boiling, chlorination should run to lessen the microbial count.

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