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Water Purification by Removal of Pathogens Using Electrospun Polymer Nanofiber Membranes: A Review

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Abstract: Pure drinking water management is a biggest challenge due to contamination with pathogens and toxic chemicals from industries and manmade pollution caused by growth of population and industrialization. The presence of pathogens in the form ofminute biological organisms such as bacteria, virus, protozoa and algae in water causes epidemics. Although, various techniques have been used for purification of water, the membrane based filtration technology considered as most suitable for removal of pathogens. Electrospun nanofiber membranes (ENM) are highly suitable for water filtration due to presence of large number of nano sized poreswith interconnected pore structure that helps permeability for water filtration and rejection of nano sized particles and pathogens. Electrospinning is a versatile technique used for preparation of nanofibermembranes in the form of cloths / nets etc. with high level of porosity and surface area. In this article, attempts have been made to review the removal of pathogens from water using electrospunnanofiber membranes with their surface and bulk modification. Effect of various pathogen killing elements such as silver, zinc oxide etc. mixed with electrospining polymer solution for preparation of functionalized nanofiber membranes was discussed. Also, preparation of PAN nanofiber membrane in CSIR-NALand its suitability for removal of e-coli bacteria from water was discussed.

Keywords: Water Filtration, Pathogens, Electrospunnanofiber Membrane (ENMs), Electrospinning

1. Introduction

Water purification from very fine particles (nano particles) and with microbial elements (pathogens) is a great challenge as contaminated water is a serious threat to the human life. Improved water supply and sanitation can drastically reduce water-borne diseases. The increase in water pollution is due to rise in population, climate change, agricultural and industrialization, urbanisation etc. A survey report by World Health Organizationin shows, although more than 2 billion people have gainedaccess to improved sources of drinkingwater and to improvedsanitation, still 700 millionpeople lack pure drinking water facility and nearly 2.5 billion people lack of adequate sanitation (Figure 1) [1].

The drinking water quality is determined by presence / absence of microbial elements, chemicals, radioactive materials etc. Pathogens are minute organisms which include virus, bacteria, algae, protozoa etc. All water borne infectious diseases are mostly caused by these pathogens. Major diseases caused by various bacteria are gastric disorder, pulmonary disease, typhoid, dysentery etc. A list of water born bacteria and the diseases caused by them is presented in Table 1. Viruses are infectious pathogens and spread diseases through contaminated water and air. As parasites, viruses are totally dependent on living cells for their survival. The size of viruses ranges from 15-200 nm. The most concern water borne virus is the hepatitis-A virus. A list of major diseases caused by viruses present in water is presented in Table 2.

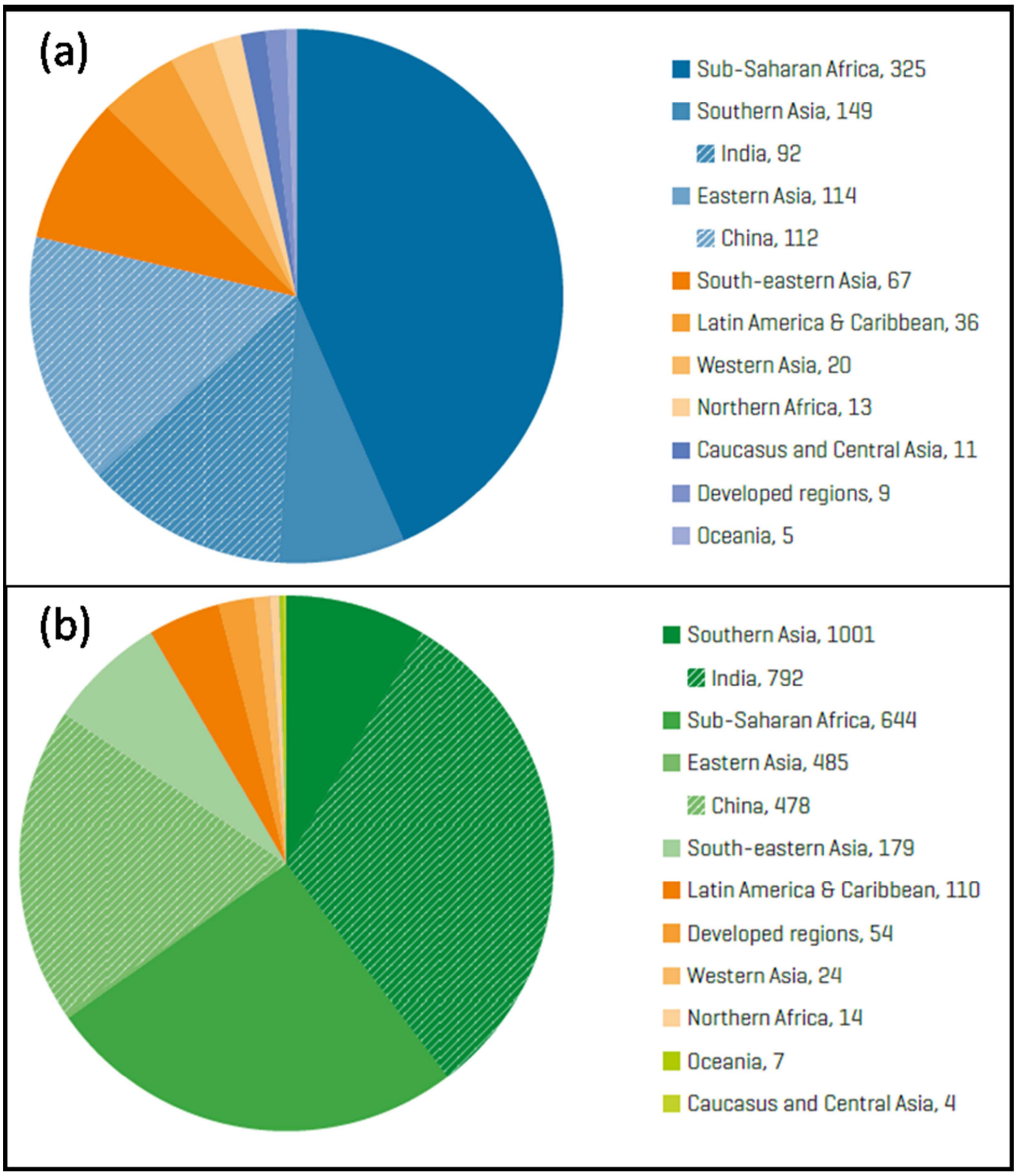

Figure 1. *Number of people (in millions) (a) without access to an improved drinking water source (b) without access to an improved sanitation facility (UNICEF Publication, 2014).*

Table 1. *Diseases caused by water born Bacteria.*

Bacteria	Major Diseases
Aeronomashydrophilia	Gastro enteritis
Bacillus	Gastro enteritis
Campylobacter jejuni	Gastro enteritis
Pathogenic Escherichia Coli (E-Coli)	Gastro enteritis
Helicobacter pylori	Peptic ulcers
Legionella pneumophilia	Legionellissis (a form of pneumonia)
Mycobacterium avium	Pulmonary disease
Pseudomonas aeroginosa	Secondary infections
Salmonella typhi	Typhoid fever
Salmonella paratyphi	Para typhoid fever
Salmonella (other species)	Gastro enteritis
Shigella	Bacillary dysentery
Yersinia enterocolitica	Gastro enteritis
Vibrio cholerae	Cholera

Table 2. *Major Diseases caused by viruses present in water.*

Virus	Diseases
Adeno viruses	Gastro enteritis
Astro viruses	Gastro enteritis mainly in children under 5 years of age.
Calici viruses	Gastro enteritis
Entero viruses	Enteric infections
Hepatitis A	Infectious hepatitis
Rota viruses	Gastro enteritis
Hepatitis E	Acute hepatitis E

There is a great demand for removal of these pathogens to avoid various diseases. A number of techniques such as chemical disinfectants and membrane based filtration technology have been used for purification of water [2, 3]. Though chemical purification technology kills the pathogens, continuous use of these chemicals increase the resistant level of bacteria / virus and also produce some by-products which are again harmful for human life [4-6]. Therefore, membrane based filtration technology is currently considered as the most suitable technique for purification of water. Nanofiber based membranes prepared by electrospinning technique can improve the filtration efficiency to a great extent due to nano porous structure of the membranes with high surface area [7]. Electrospinning is a multipurpose technique used for preparation of very light weight nanofiber membranes in the form of clothes / nets from a biocompatible polymer solution. In this review article, purification of water by various techniques and removal of pathogens from water using electrospun nanofiber membranes with surface and bulk modification is reported. Also, preparation of PAN nanofiber membrane and its suitability for removal of e-coli bacteria from water is discussed.

2. Water Purification Methods

There are various methods used for water purifications. Traditionally, these include: (i) physical separation of particles through fine cloth, clay vessels etc, (ii) chemical treatment through chlorination, (iii) evaporation and condensation technique etc. Now a days, suitable membranes are also used to supplementthe above techniques for better purification of water [2, 3].

Membrane filtration technique was initially used for desalination of ground water, seawater, and brackish water. Membrane filtration processes are classifiedaccording to the membrane pore size, and size of the particles theyare able to retain. These are (i) microfiltration (MF), pore sizebetween 0.1 - 10μm (ii) ultra-filtration, (UF), pore sizebetween 30-100 nm, (iii) nano filtration (NF), pore sizebetween, 1-20 nm and (iv) reverse osmosis (RO), pore sizebetween, 0.1-1 nm. A comparison of various membrane filtration techniquesis presented in Table 3.

Table 3. *Comparison of various membrane filtration techniques.*

Filtration class	Particle capture size	Typical contaminants removed	Typical operating pressure ranges
Microfiltration (MF)	0.1-10 μm	suspended solids, bacteria, protozoa	0.1-2 bar
Ultrafiltration (UF)	0.003-0.1 μm	colloids, proteins, polysaccharides, most bacteria, viruses (partially)	1-5 bar (cross-flow) 0.2-0.3 bar (dead-end and submerged)
Nanofiltration (NF)	0.001 μm	viruses, natural organic matter, multivalent ions (including hardness in water)	5-20 bar
Reverse osmosis (RO)	0.0001 μm	almost all impurities, including monovalent ions	10-100 bar

Membranes are generally prepared from polymeric material for water purification application. Performance of the membrane mostly depends on the factors like Flux and Retention. Flux is the amount of water passing through the membrane of specific surface area per unit time and retention isthe ratio of concentration of a salt/ pathogens in the filtered water and the input feed water [8]. Membrane fouling is also a common phenomenon during filtration process. It mainly occurs due to deposition of various components (nano particles, colloidal particles, precipitated inorganic salts etc) in the pores and at the surface of the membrane. The membrane fouling can be reduced by pre-treatment of the feed solution and selection of suitable membrane material with appropriate porosity.

Among various filtration techniques, reverse osmosis is a common filtrationmethod which can remove dissolved soluteusing a semipermeable membrane. Except RO membrane all other filtration membranes are porous in nature with physical holes. The RO filtration membrane is generally hydrophilic in nature which allows easy diffusion of water molecules through the semipermeable membranes. But due to large trans membrane pressure and biological fouling it is difficult to remove the virus completelyby RO membranes [9]. Among various types of filtration membranes, nanofibrous membranes are found to be suitable for filtration application due to small interconnected pore structures, high permeability, low weight, high specific surface area [10]. The above properties of nanofiber membranes make them suitable for removal of viruses, bacteria with other inorganic and organiccontaminants and would help in production of cost-effective drinking water from various water sources [11].

3. Nanofiber Membranes by Electrospinning

Nanofiber membranes are prepared by various processing techniques such as drawing, template synthesis, self-assembly, electro-spinning technique etc. Among these, electro-spinning is the simplest method to prepare highly porous nanofiber membranes with low weight, fine interconnected open pore structure and high specific surface area [12-14]. The technique is used for fabrication of sub-micron to nano meter range fibers [15, 16]. In this process a strong electric field is applied to a polymer solution taken in a syringe. The metallic needle of the syringe is connected to the positive terminal and a plate covered with aluminium sheet (collector) placed at few cm apart is connected to the negative terminal of the high voltage source. On application of high dc voltage a stream of fluid comes out automatically through the needle, and deposited in the form of nanofibers on the collector plate. A thin sheet of nanofiber mat can be prepared by continuing the process for larger duration. A schematic drawing of the electrospinning process set upis presented in Figure 2. The technique has been used for the preparation of various polymer and metal-oxide/ ceramic nanofibers such as alumina, silica, zirconia, titania, nickel oxide, lead zirconatetitanate and other oxide materials [17-25]. Recently, few reports / research publications available on electrospunnanofibrous membranes (ENMs) for water treatmentapplications. Balamuruganet al. reportedpreparation of nanofiber membranes and their possible applications in watertreatment [26]. Fenget al. summarized few reports which emphasize the importance of ENMs in water filtration technology [27]. A number of polymershave been tried to remove the pathogens. The commonly used polymers are chitin/chitosan derivative [28], poly vinyl alcohol [29], chitosan/PVA [30], Poly acrylonitrile (PAN) [31-33], poly vinlypyrrolidone (PVP)[34], Polyethlyene terephthalate [35], polymethyl methacrylate (PMMA) [36] etc.

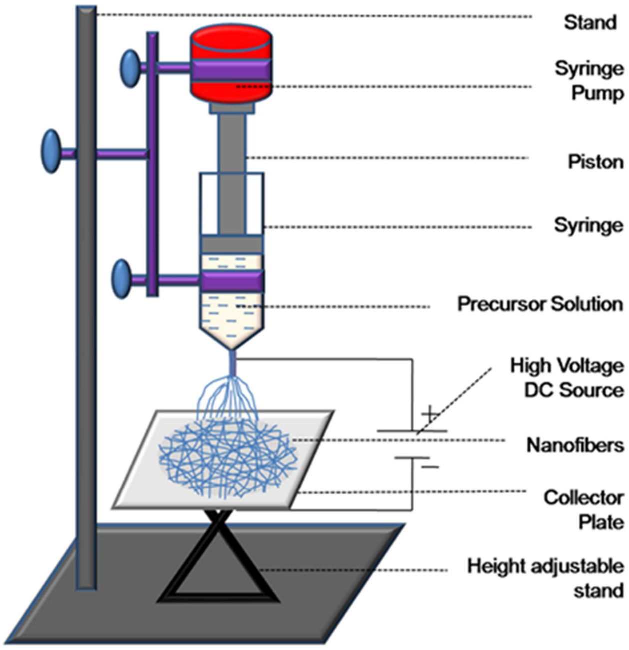

Figure 2. *Schematic drawing of the electrospinning process set up.*

4. Characterization of Nanofiber Membranes

A variety of techniques are used for characterization of physical and chemical properties of the membrane surfaces. The important parameters are contact angle measurement, zeta potential and surface morphology. Contact angle measurement provides the information on wetting properties of the membrane surface. Zeta potential measures the surface charge on membrane surface. Scanning electron microscopy / Atomic force microscopy is used to both quantify and visualize the morphology or roughness of membrane surfaces [37].

4.1. Contact-Angle

Contact angle (θ) measurement is used to provide information regarding the wetting properties of the membrane and generally measured by two techniques such as sessile dropand captive bubble method. A number of factors such as surface heterogeneity, surface roughness, volume of dropped liquid, measurement temperature and time, surface contamination etc can influence the contact angle measurement on a membrane surface since membraneshave porous and rough surfaces. The information on surface energy properties can also obtained from contact angle analysis.

4.2. Zeta Potential

Potential difference between the bulk of solution and the sheer plane of the interfacial double layer created by the membrane and the liquid is termed as Zeta potential (ζ) [38]. It provides the information on charge developed on membrane surface and the membrane fouling potential. The properties like pH, ionic strength at the solid-liquid interface mostly affect the value of zeta potential. Zeta potential is related to streaming potential (potential generated when an electrolyte solution flows through a thin channel or porous media) by the Helmholtz-Smoluchowski equation:

$$(E/p)= [\varepsilon\varepsilon_0 / \zeta \lambda \eta] \quad (1)$$

Where, E: Streaming potential
p: Applied pressure for driving the flow,
ζ: Zeta potential,
λ: Conductivity of electrolyte,
η: Viscosity of the solution,
εpermittivity of the solution
ε_0: permittivity of vaccum.

A streaming potential analyzer is used to measure the values of E, p, and λ. while ε and η are calculated traditionally using viscometer and dielectric measurement equipment.

4.3. Porosity

Porosity is one of the important parameter in filterdesign and its performance. Proper amount of porosity in the nanofiber membrane helps to improve the performance of the filters. Conventionally, porosity is measure from the apparent density and bulk density of the material [39]. But, other methods such as image analysis [40] and mercury porosimeter [41] are quite frequently used for the measurement of porosity in nanofiber membrane. Recently, Sreedhar et al. demonstrated the porosity measurement by pycnometer (used for determination of volume of porous material) and measuring the thickness of membrane by FESEM image [42]. The method allows minimum damage to the membrane and calculates total porosity more effectively.

4.4. Clean Water Permeability (CWP) Test

Clean waterpermeability (CWP) test is used to measure the rate of waterpasses through the filtration membrane. To determine the CWP value experimentally, a known volume of water passed through the membrane of a fixed surface area under a particular pressure. The pressure was then varied to have different flow rates. Then the CWP value was calculated from slop of the graph (flow rate vs. change in pressure). This is an important characteristic of the membrane which decides the amount of water collected. At certain pressure, higher flow rate means the high CWP value and lower operation cost. As nanofiber membranes are highly porous with inter connected pore structure a high CWP value is observed when the load on the membrane is low. Therefore, nanofiber membranes are most suitable for water filtration application.

4.5. Log Reduction Value (LRV)

Log reduction value (LRV) is defined as the log_{10} (C_{BF}/C_{AF}), where $C_{BF}$is the initial concentration of pathogens in unfiltered water and $C_{AF}$is theconcentration of pathogensin filtered water. An LRV of 1 means 90% of the pathogens has been removed from the water and an LRV of 3 represents 99.9% removal of pathogens.

5. Functionalization of Nanofibrous Membrane for Removal of Pathogens

The pore size of the filter media determines the ability ofthe filter to trap particles of a specific sizerange. As the fiber size decreases, the filtermedia becomes a high retentive surfacefilter with limited loading capacity, reduced flow rate, and increased pressure. To overcome this bottleneck and toexpand the application of nanofibrousmedia in liquid separation, researchershave been functionalizing the surface ofnanofibrous mediaby adding functionalizingagents (such as inorganic particles or organic biocides) to themembrane which improved the pathogen removal efficiency and antifouling properties of thenanofiber membranes [43]. The functionalization of nanofiber membrane can be done intwo different ways i.e. (i) Surface modification and (ii) bulk modification. In the former case, the top layer ofthe membrane is modified with functional materials and in the latter case eachfiber in the membrane throughout ismodified with nano particles.

5.1. Surface Modification

The surface modifications of nanofiber membranes enhance the nanofibers matrix properties such as availability of functional groups on the surface of nanofibers. Various methods used for surface modification are (i) plasma induced surface graft copolymerization [44], (ii) chemical oxidation [45], (iii) organic chemical surface functionalization [46], (iv) radiation induced surface grafting method [47] etc. In plasma induced graft copolymerization, the surface of the membraneis exposed to plasma followed byoxygen and then polymerized. This technique has beenused to reduce the surface pores of nanofibrous filter media to below 1 micron, resembling an asymmetric structure. Most significantly, waterflux permeation studies revealed that thegrafted nanofibrous media had a betterflux throughput than a commerciallyviable phase-inverted membrane of the same pore-size.

5.2. Bulk Modification

In bulk modification process each nanofiber of the membrane is coated with nano particles which helps the separation of particles / pathogens based on an affinitymechanism. Inorganic nanoparticles with known antibacterialproperties, such as silver, copper oxide, zinc oxide, and titanium dioxideis mostly used to enhance the filtration efficiency of nanofiber membrane [48-50]. It is observed that the pathogenremoval properties of polyamide nanofiber membranes can be significantlyincreased by adding nanosilver or organic biocides to the electrospinning solution [51]. Similarly, ArgonideCorporation has developed a technology that provides an effective incorporation of nano powders into a nanofiber membrane without affecting its surface properties. It claims nearly 99.999% of virus removal from water using a 1 mmthick membrane [52]. Bjorge et al. functionalized the nanofiber membrane with Ag nanoparticles which gave a log 4 removal of gramnegative Escherichia coli bacteria whereas the membrane is insensitive to gram positive bacteria [53]. Chen et al. prepared cellulose acetate fibers containingchlorhexidine as a bactericide which is used for removal of gram positive bacteria. Sundarrajan et al. studied preparation of polyethylene terephthalate (PET) blend with cellulose acetate and cellulosenanofibers by electrospinning technique [54]. Various metal oxide nano particles were deposited on nanofiber surface using liquid phase deposition (LPD) and electrospraying techniques. They observed high particle density on PET-Cellulose acetate and PET-cellulose blended surface compared to PET surface. Lala et al. prepared several silver impregnated polymeric nanofiber membranes and tested for its the bacteria removal efficiency against two gram negative bacteria such as E. coli and P. Aeruginosa [55]. The results were quite promising when the membrane were incubated with the above bacteria. Ignatova et al. reported the use of electrospun polyvinyl pyrrolidone-iodine complex (PVP-iodine) for antibacterial, antimycotic andantiviral applications by release of iodine during filtration process [32].

Poly-acrylonitrile (PAN) nanofiber has very good antibacterial activity against gram negative E. coli bacterial and is biocompatible [56]. Therefore, these nanofiber membranes are suitable for water filtration application for removal of pathogen. It has also ability to adsorb viruses when infused with cellulose. Infusion of cellulose reduces the pore size of PAN nanofiber and able to remove E. Coli bacteria up to 6.0 LRV, B. diminuta bacteria up to 4.0 LRV and MS2 bacteriophage virus up to 2 LRV [57]. Viruses are contains carboxyl and phosphategroups on their surface which are negatively charged; therefore, the positively charged nanofibers would able to adsorbvirus with higher LRV. Sato et al. modified electrospun PAN nanofiber with di-amine group which was successfully remove MS2 bacteriophage virus up to 4 LRV [58]. Li et al. reported similar result with positively charged alumina nanofiber filters which remove the virus about 2 LRV [59]. The virus removal efficiency of the filter increases with high surface area of the filter or long contact time. Bjorge et al. prepared polyamide nanofiber membrane and tested its water filtration efficiency after functionalizing with silver nano particles, biocide etc [60]. They observed that the efficiency of non-functionalized membrane was only 2 LRV. Its efficiency increased up to 4 LRV when functionalized with silver nanoparticles and ~6 LRV when it is functionalized with a biocide (WSCP). The lower removal of pathogens with silver nanoparticles compared to biocide could be explained by the fact that the silver nanoparticles only have a biocidal effect on gram-negative bacteria [61-64]. Various electrospunnanofiber membranes used for removal of pathogens are presented in Table 4.

Table 4. *Various electrospunnanofiber membranes for removal of pathogens.*

Electrospunnanofiber membranes	Anti-pathogenic activity	Reference
Poly acrylonitrile (PAN)	E. Coli bacteria	[60]
Poly acrylonitrile (PAN)	S. aureus, E. Coli bacteria	[50]
Nylon-6 functionalized with Ag	S. aureus, E. Coli bacteria	[61]
Poly acrylonitrile (PAN) functionalized with Ag	B. subitilies, S. aureus, E. Coli bacteria	[62]
Chitosan in acetic acid	Feline calicivirus F9 (FCV-F9) Murine norovirus (MNV-1).	[63]
PAN infused with cellulose	MS2 bacteriophage virus (2 LRV) B. diminuta bacteria (4 LRV) E. Coli (6 LRV)	[50]
PVP)-iodine complex	Anti-bacterial and anti-viral	[32]
Alumina nanofiber	MS2 bacteriophage virus	[58]

6. Preparation of PAN Nanofiber Membrane and Its Characterization

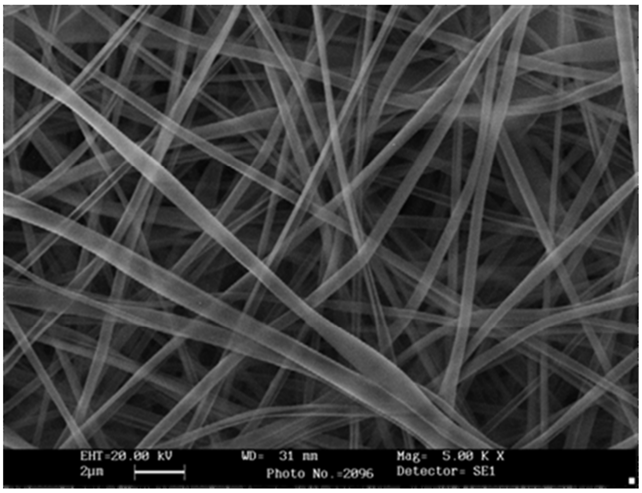

Figure 3. *SEM image of typical PAN nanofiber membrane.*

PAN nanofibers were prepared successfully by electrospinning technique using PAN of different concentrations (9-16 wt%). Solution of PAN prepared in dimethyl formamide was subjected to electrospinning by taking 2 ml of the solution in a syringe and connecting it to the positive terminal of the high voltage source. A rotating drum covered with aluminum foil served as counter electrode. The experiments were carried out by maintaining a distance of 15 cm between the tip of the syringe to the drum collector. The solution flow rate was maintained 1.0 ml/hr and humidity of the chamber was maintained in the range of 50–60%. The voltage was gradually increased till the liquid came out through the needle and split into web of fibers collected on the aluminum foil. It was observed that PAN at 10wt% concentration is efficient for fiber formation. A sandwich structure of PAN membrane was prepared by keeping it in between two polypropylene sheets to protect the PAN nanofiber membrane and give good strength and support to the membrane. A single layer of prepared PAN sandwich mat (thickness 200 µm) was tested for the microbiological reduction test. A known concentration of ($9x10^4$cfu/ml) E. coli bacteria was passed through the membrane. After filtration, the concentration of the E-coli in the filtrate was found to be 113cfu/ml, corresponds to 99.87% reduction. SEM micrograph of typical PAN nanofiber membrane is presented in Figure 3.

7. Conclusions and Future Work

Pure drinking water is necessary for maintaining a healthy life. Nanofiber based membranes found to be most suitable for purification of water. These nanofiber membranes are easily prepared by electrospinning technique with tailored porosity helpful for removal of pathogens as well as fine particulates. Various pathogen killing elements such as silver, zinc oxide etc can be mixed with electrospining polymer solution for preparation of functionalized nanofiber membranes which can effectively remove / kill the harmful pathogens from water. Similarly, nanofiber membranes with graded porosity can be tried for higher pathogen removal efficiency. Suitable packaging / casing of nanofiber membranes is most important so that the broken nanofibers should not contaminate the water.

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