

**Development of environmental friendly
approach towards the usage of sanitary napkin.**

Under the supervision of

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&

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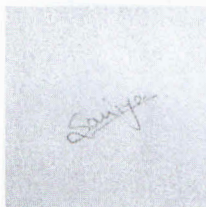
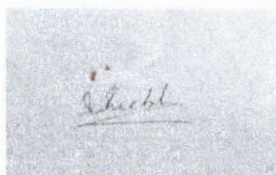
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CERTIFICATE

We, hereby declare that the work presented here in this report entitled “ **DEVELOPMENT OF ENVIRONMENTAL FRIENDLY APPROACH TOWARDS THE USAGE OF SANITARY NAPKIN**” in partial fulfilment of the requirements for the award of the degree of Bachelor of Technology in Biotechnology submitted in the department of Biotechnology and Bioinformatics , Jaypee University of Information and Technology Waknaghat is an authentic record of my own work carried out over a period from August 2019 to December 2019 under the supervision of Dr. Abhishek Chaudhary and Dr. Sudhir Kumar.

The matter embodied in the report has not been submitted for the award of any other degree or diploma.



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This is to certify that the above statement made by the candidate is true to the best of my knowledge.

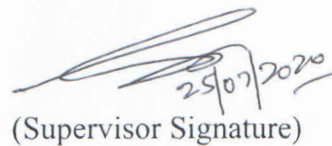
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1. Abstract

According to the estimated study there is insufficiency in proper menstrual hygiene facilities globally . There are specific preferences, sanitation needs and experiences for both woman and men. Poor menstrual hygiene can lead to many different issues including bacterial or fungal infections of the reproductive tract, can cause discomfort due to irritation of the skin and sometimes can also cause blisters. Taking into account the economic and social factors of the country it is essential to consider environmental and public health risk. Hence, our project is to customize an environmental friendly sanitary napkin with different beneficial factors like better absorbancy, leak protection, dryness, non- toxic, comfortable and easy availability. The Eucalyptus extract coated silver nanoparticles will be used to play an advantageous role in the production of sanitary napkin.

2. Introduction

Menstruation is considered to be an important part in the life of women. According to the recent studies India has approximately 355 million menstruating women, however the reproductive health is being neglected and poorly understood due to the different beliefs, myths and taboos associated with them, which can lead to different infections in the life of female as a whole. Hence understanding the menstrual health, it is important to devise mechanism to protect them against various infections through different awareness and social norms.

Approximately, 12% of women are observed to use hygiene products like sanitary pads, tampons etc. whereas 75% still use rags and cloths to absorb the menstrual flow resulting to 125 to 150 kg's of disposal products in her lifetime (1). It is important to make women aware regarding various menstrual hygiene product and making such products that are not just available to particular social class but available for lower and middle income classes as well.

Basic composition of sanitary napkin consists of polypropylene, non-woven fabric which is considered to a non-biodegradable material. Hence it became a major concern to make sanitary napkin which provides maximum absorbency along with minimum toxicity and maximum biodegradability.

Utilising nanotechnology and manipulating a matter to its optimum, both "Nano enhanced" and "Nano enabled" nanocellulose was produced from cellulose resulted in increased absorption property.

(1.) Nano cellulose possessing qualities like high tensile strength, great surface area and inert chemical nature

(2). It gave a major benefit for making of high absorption material for sanitary napkin. Further to avoid vaginal infection and urinary infections antiseptic and antibacterial qualities are required to be introduced on the surface of sanitary napkin.

Silver being used from the medieval period from 440 B.C, Hippocrates was the person to use it for healing wounds. Silver nanoparticles were synthesized using green synthesis. Green synthesis involves using of a plant extract, here in Eucalyptus plant extract was utilized, since it possessed Tannic acid which is responsible for reduction of the silver nitrate to silver nanoparticles. Introducing silver's antiseptic properties along with eucalyptus

antibacterial properties served as major contributor towards protection against various urinary and vaginal infections.

Nano cellulose being used in sanitary napkin gave a major benefit towards absorption during menstruation along with AGNPs giving antiseptic and antibacterial qualities enabled the designing of such sanitary napkin which will be commercially available to every social class .Educating the masses and literacy regarding hygiene and menstruation gives the women a better perspective towards development of better menstrual hygiene and using products that are beneficial to them along with the society as whole.

3. Review of literature

In middle and low income countries there are many prohibitions regarding the monthly menstrual cycles. This waste disposal concerns combined with the urban waste collection systems are creating problems in many low- and middle-income countries (LMICs) which includes various risks and environmental pollution in many populated areas. The examination of peer reviewed and grey literature regarding the menstrual waste disposal with the objective of informing the water, sanitation and hygiene (WASH) to plan on more advanced facility user designs and waste management practices to support the menstrual hygiene needs of women and girls (2) . The primary focus was to understand the disposal and management of menstrual waste. Various factors included types of absorbents used and disposal method in urban and semi-urban areas of middle or low income countries; socio-economic factors that rule discarding practices; involvement of empowerment and dignity along with sanitation and various environmental and human health risks from menstrual waste and their improper disposal and from incineration; social and cultural factors related to use and acceptance of incineration & disposal practices in public and institutional settings in the developing world and policies or guidelines for menstrual waste disposal. The use of menstrual hygiene products, which is a basic component intimately linked with disposal practices. These materials are used for the better absorption of menstrual blood rather more safely. Our literature review disclosed no acceptable segregation and distinction of menstrual hygiene products; categorizations of these products were further done according to the type & quality characteristics of the products used in the sanitary napkin. Thus, for the making of this review, we have collectively segmented different menstrual hygiene products into sanitary napkins of commercial use such as disposable sanitary pads or handmade or formally made reusable napkins; the different absorbents utilized since medieval times were cloth, cotton, wool, toilet paper and commercially available products such as tampons and menstrual cups. The major role of these menstrual products concerning hygiene is regulated by various factors like leak protection, absorbency quality, dryness, regarding leak protection and comfort. It also includes size, thinness, toxicity and biodegradability. Various number of factors are taken into account while choosing the type of incinerator to be used depending on the types of absorbents used, also installation of the incinerator (e.g., houses, public toilet, schools or university settings), the amount of waste utilized for incineration, temperature utilized for treatment, retention capacity, budget amount available and the functioning and

maintenance also emission. Various examples of these technologies utilized were seen to be less complex than the incinerator technologies which included clay pots and low cost locally made incinerators. (2)

Usage of sanitary napkin in different low or middle income countries was studied and found to be minimum instead usage of cotton and wood pulp was primarily used apart from these banana fibres and bandages were used instead of cotton cloths. While the Indian government under their Solid Waste Rules 2016 classifies that “ sanitary pads solid waste, policy guidance on sanitary pad waste collection, handling, storage, transportation, treatment through incineration or any other methods need detailing”. Environmental factors and rules need to addressed with the fact that these available tampons and sanitary napkins contain poly ethane and chlorine that may produce dioxins and other potentially dangerous chemicals that can contribute to the health concerns which is caused due to the air emissions from combustion processes, or groundwater contamination as a result of leaching from unlined landfills and sites where solid waste is accumulated. (2)



Fig. 1 Various Waste disposal pathways and potential environmental and health hazards. (2)

The menstrual waste can be classified into 2 categories: disposal sanitary napkin & cloth napkin

- Solid waste system: incinerator, burning, waste bin, burying and discarding.
- Sanitation system: latrine, septic.

Where the waste accumulates in river, oceans or landfills, burning produces toxic fumes and clogs sewer and septic tanks. Further, inhaled by people also can be consumed by marine life leading to land and water pollution.

Incineration is considered to be an important factor for managing disposal of menstrual waste particularly in works places and educational settings maybe called as a method for achieving pathogen treatment, waste reduction and waste management on sight. (2)

Menstruation takes place between the ages of 11-48 yrs. on an average and disposal of the menstrual waste products are considered to be taboo in society for the difficulty in its disposal affecting the dignity of the person. Social and cultural thought process referring the disposal of menstrual blood and menstrual waste products maybe the reason behind some girls not preferring to discard the used pads in the school incinerator. According to a study shown girls extracted different benefits of utilizing an incinerator in schools and institutions which also gave an ease for schools going girls (34%) and beneficial and better school services for disposal and safety of sanitary napkin (42%), hence giving an indication for usage of incinerators in schools (3). Malawi school girls noted that need for sanitary napkin (26%) and incinerators (11.4%) were regarded as important requirement for menses maintenance in a better way. The majority of population felt comfortable disposal using communal incinerators, but a study carried out in state of Tamil Nadu concluded that incinerators which were installed in a communal toilet were not in use due to the lack of signage (4). In some communities, the burning of menstrual blood is considered to be a taboo, drawing on the belief that such actions will compromise a woman's reproductive capacity (5).

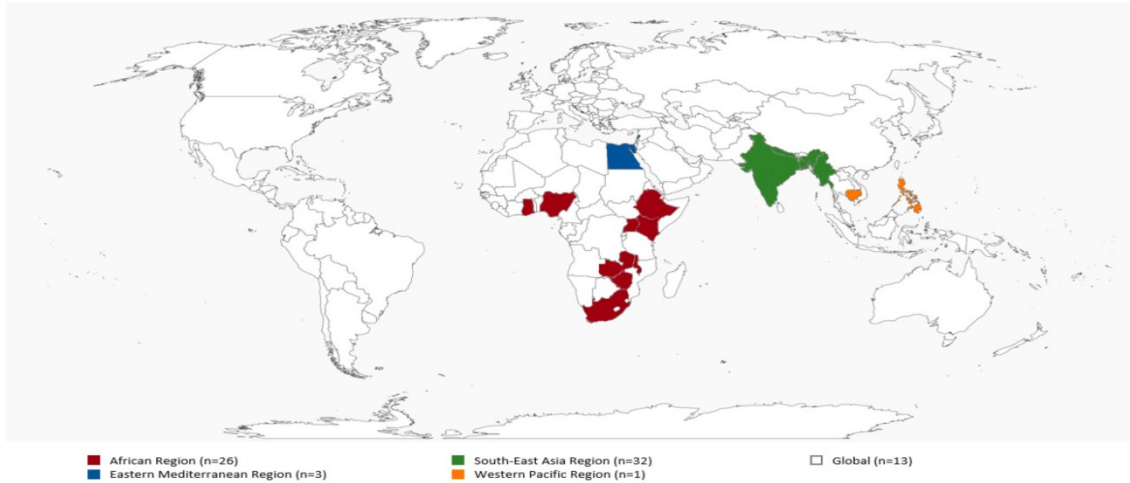


Fig. 2 LMIC's country & regions for articles reviewed on menstrual absorbents. (2)

Recent studies in low and middle income countries referring to the usage of sanitary pads, the most commonly used materials were cloth, tissue paper (6-10). Apart from these handmade pads, clothes, sponges and cotton, wool was also cited to be a absorbent (6-10). Egypt, Nigeria and many states commonly utilized sanitary napkin. Many Asian countries used cloth instead of sanitary napkin. Also usage of cloth was seen as major menstrual hygiene product among the ages of female 15-49 yrs. conferred from census data of 2007-2009. Urban areas in India showed usage of sanitary napkin after ages of 20 Various studies conducted in Nigeria, Uganda and India showed few Tampon users (11). Studies of menstrual cups are under trails; also the use of such products that are insert enabled menstrual hygiene product are conflicting with the religious beliefs and cultures connected to virginity (11-14). Usage of sanitary napkin are more preferable as it provides protection against odour, leak protection and gives comfort while usage of cloth cannot fully ensure these qualities , affordability is seen as a major drawback (10,13,15,16) cellulose and various super absorbent was seen as major contributor towards absorption.

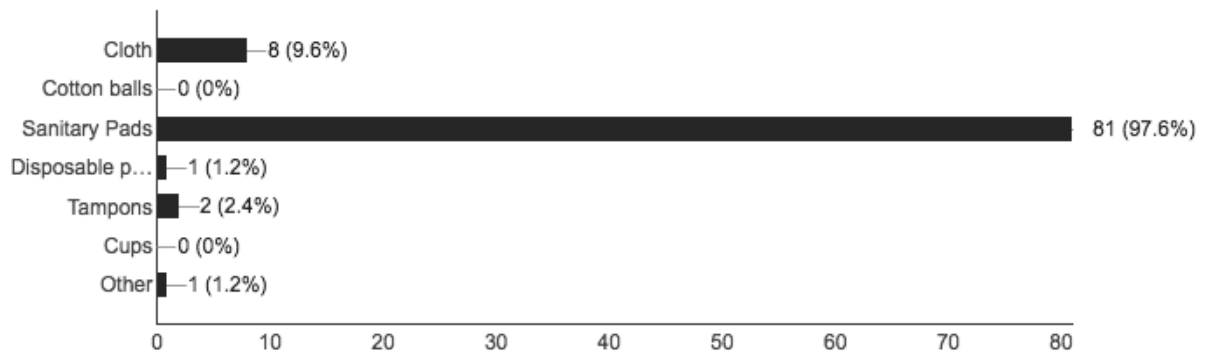


Fig. 3 Different types of menstrual absorbents used by the respondents. (17)

Urban areas of New Delhi referred a study that sanitary napkin usage was more common among young women aged 20 to 29yrs in comparison to women of 30yrs and above also the use was in direct correlation between educated mothers or women and usage of sanitary napkin as they are more aware (18). Also in secondary schools of Egypt usage of sanitary napkin was associated with the social class of family and its settlement in urban areas as they are more exposed towards the awareness of usage of sanitary napkin (19). Various studies brought the fact that usage of sanitary napkin was more common among the private institution in comparison to schools having government aegis (20,21) , referring to the reason that the family of users maybe be wealthy (10) . This review claims that the social classes and education play a major role towards the hygiene management of menstruation and its disposal of the waste; policies are being devised binding to the conclusion. Our review majorly focused on Sub Saharan and South Asian countries. Hence focusing of construction of efforts built globally on menstrual hygiene management, usage of sanitary napkin their material and absorbancy and age group underlying with the usage apart from this the socio-culture effects, location. It is important to understand the use of sanitary napkin among various demographic female populations (2).

In Bangladesh low income communities disposed the sanitary napkin in dumps and ditches also preferred throwing in toilets (22). In a study of communal ablution blocks (CABs) in Durban (South Africa) it was shown that none of the studied CABs had bins for disposal within the toilets (23). The public toilets were installed successfully with dustbins containing liners in capital of Bangladesh, Dhaka (22). Apart from this South African women had the fear of dogs as they may dig the used sanitary napkin when disposed in dustbins (24). In few areas buckets with covered lids were used in backyards to dispose the sanitary napkin

privately. Few women preferred keeping the used sanitary napkin beneath the bed until they found better way to dispose (24). Some references suggested that disposal of sanitary napkin was done using paper or plastic covering (24, 25, 26). A UNICEF studies showed that very few washrooms had dust bins and hence girls had to carry the used sanitary pads back homes (27). Disposal of sanitary napkin was referred as an awkward situation among Malawi females.

In a country, Nepal observed 28.2 % of school absentees due to lack of disposal system of used sanitary napkin (28). A percentage of 69.3 % of Ethiopian females faced discomfort due to lack of space available for changing the sanitary napkin or absorbent.

19.1% females couldn't access water for washing. And only 10% got disposal facility available (29). A study conducted in Ethiopia showed that only 8.5 % of females remained absent from school due to menstruation as there was absence of disposal facility (30).

In urban areas females are continuously among crowd and giving less privacy towards disposing the sanitary napkin (31).

Throughout the globe, females suffer major stress and pain in menstruation and while dealing with the menstrual wastes. The menses blood is considered to be impure and unhygienic (9). Various bondages comes along with menstruation are restrictions are imposed on females going through it. The sight of menstrual blood on different absorbents is believed to cause various harms to those who see it or come in contact with it, and some fear that the various myths and taboos are connected with the blood of menses such as the sight of it can cause harm (32). To make the stain of menses blood wash away females preferred washing it after use or covering using paper or polythenes. In order to remove the traces of blood from absorbents, (21, 32, 33) or also few preferred keeping them separately before throwing (24, 34, 35). Menstrual management in various camps was seen as a major challenge and to the fact when disposal system had been provided; burying the waste is also seen as an alternative to discarding (36). Requirement for segregation gives the reason why the napkins are required to be disposed in toilets (22, 29).

Wrapping the sanitary napkin with plastic gave more requirements and led the service provider synthesize napkin including plastic covers enhancing the mechanism of waste disposal , however this has resulted to increased environmental waste accumulation (2) Incinerators are being promoted by the Government of India as a mechanism of waste disposal resulting in reduction of environmental burden only once the design requirements are met .But due to the absence of proper emission regulation enforcements on incinerator

and hazards connected to the use of the incinerators and their emission is still a major problem (37,38).

3.1 Cellulose

Cellulose is estimated to be the most important part in the plant cell wall and it is from wood, grasses, seed fibres (39,40). Cellulose is widely available and the most important feature is that it is a renewable polymer which can be used for the manufacturing of food, paint as well as textiles etc. (41)

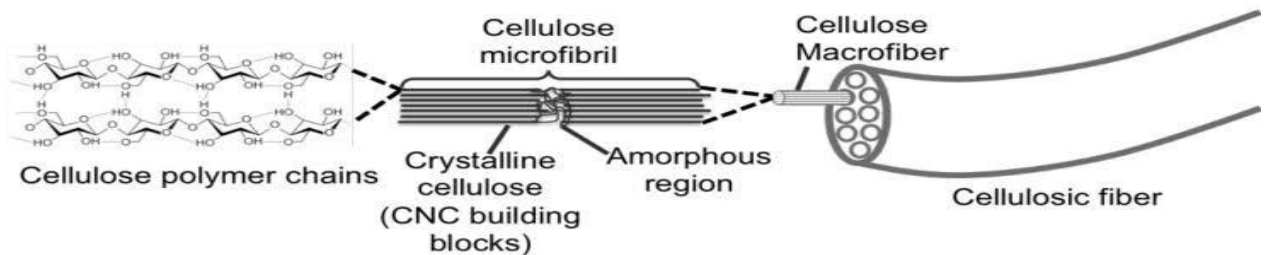


Fig. 4 cellulose macrostructure (92)

Payen was the first to discover about cellulose (42). After that there was an intensive study about the material properties of cellulose. Lignin and hemicellulose bind the microfibrils of cellulose polymer. Mostly its microfibrils tiny cells have a width of 10-50 micrometre. (43)

It is a steady polymer that is extracted from natural environment, its hydrogen bonding network pattern makes it insoluble in aqueous solvent and makes it difficult to show a melting point (44). These polymers are formed by D- glucopyranose molecules, and are linked together by β 1, 4 – glycoside bonds (45). These materials are formed by the degree of polymerization of cellulose molecules.

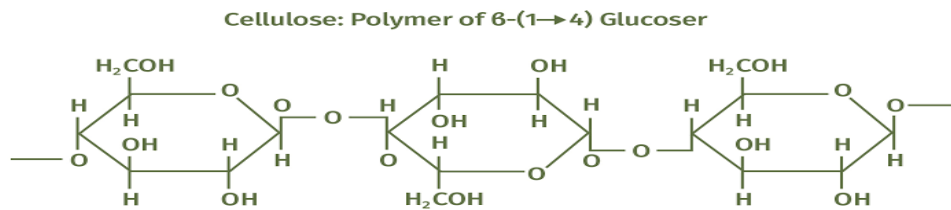


Fig. 5 structure of cellulose.

Cellulose polymer has four different classes i.e. cellulose i, ii, iii, iv.

- Cellulose i which is called the native cellulose is generally found in nature (46).
- the most steady form of cellulose is of class ii and it can be isolated from sodium hydroxide treatment of cellulose. The characteristic difference between the cellulose i and cellulose ii is the layout of their atoms, i.e cellulose i in parallel direction whereas the cellulose ii in the antiparallel direction (47-48)
- Cellulose iii is commonly extracted through the ammonia treatment of cellulose ii and cellulose i
- The modification of cellulose iii leads to cellulose iv(48).

3.2 Nano cellulose

The main reason for the increased use of nanoparticles nowadays is the fact that it is highly uniform in structure and contains biodegradable & mechanical properties. Nanocellulose is referred to as the cellulose molecules with 1-100nm in size (49).

3.2.1 Types of Nano cellulose:

- **Cellulose Nano crystals**

These types of nanocrystals are formed when cellulosic materials are dispersed in water using acid hydrolysis technique. During the acid hydrolysis treatment, the amorphous

regions of cellulose are dissolved whereas the remaining regions are crystalline , along with which a rod like rigid CNC is produced (50). The CNC's thus produced have a length of 200-500nm and diameter of 3-35nm.

- **Cellulose Nano fibrils**

These are the long entangled fibrils which are produced by the grinding of cellulosic pulp (51). Unlike CNC's, CNF's contains both the cellulose domains within the single fibres i.e amorphous as well as crystalline (52), having a diameter of 5-50nm and a length of few micrometres (53). CNF can be extracted from cellulose fibres by three processes i.e.

(1.) Mechanical treatment (e.g. grinding, milling, homogenization)

(2.) Chemical treatment (e.g. TEMPO oxidation)

(3.) A combination of chemical and mechanical treatment. (45)

- **Bacterial cellulose**

Also known as the “**microbial cellulose**” and is produced from *Acetobacterxylinum* bacteria (54). For the biosynthesis of these molecules bacterial body is continuously supplied with glucose chains and are expelled through the minor pores on the cell wall (53).

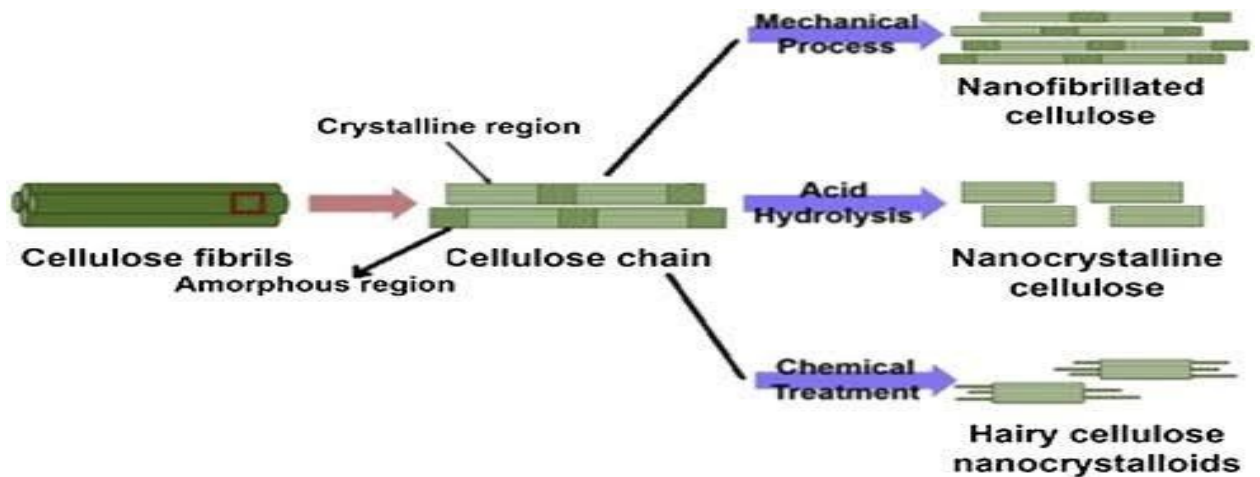


Fig.6 Formation of Nano cellulose from cellulose. (55)

3.2.2 Applications:

Nowadays, Nano cellulose composites are used for medical, automotive, electronic and in waste water treatment. It is believed that nanocellulose has replaced the synthetic materials and is more environment friendly in nature.

- 1.) in paper industry.
- 2.) in composite industry
- 3.) in biomedical industry

3.2.3 Why Nano cellulose over cellulose?

Due to the its hygroscopic tendency and absence of melting point, there is a limitation in the cellulose applications and developments of various biocompatible products are therefore done at nanostructure level (56, 57). Although nanocellulose is originated from the natural cellulose and it consists of physical properties that are high in nature and also possesses exceptional chemical properties. In recent years, nanocellulose has accumulated high rate of interest in the material science and in fields of biomedical engineering due to its renewable nature, good biocompatibility, interesting optical properties and excellent

mechanical properties. The most beneficial property of nanocellulose is its green nature of particles, its physical and its chemical properties (58).

Table 1: distribution and description of Nano cellulose.

Nomenclature	Abbreviations	Size distribution(nm)	Method of preparation(sources)	Reference
Cellulose nanocrystals,nanocellulose, nanocrystallites of cellulose,crystalline cellulose, cellulose nanowhiskers.	CNC's ,NCC's ,CNXL's ,XNL's ,CNW's	Length : 50-500 Width : 3-15	Acid hydrolysis(wood,cotton,rice,ramie,sisal,tunicates)	59-68
Cellulose nanofibres,nanofibrillated cellulose,nanofibrous cellulose	CNF's , NFC's	Length:50-3000 Width :5-50	Mechanical treatment(wood, tunicates)	59-61,69-74
Bacteria cellulose, bacterial cellulose, bacterial Nano cellulose, bacterial Nano fibre.	Bacteria CN's , BNC's ,BNF's	Length :200-3000 Width : 10-75	Bacteria	59, 75-77

3.3 Green synthesis using silver nanoparticles

3.3.1 Nanotechnology

In the modern material sciences, nanotechnology is considered to be the most active areas of research. Green synthesis is making a great impact in all areas of human life. It consists of completely new & improved properties which are rising at an increased pace depending upon its size, distribution and morphology (78-80). In the areas of molecular detection, diagnostics and anti microbial and medicines usage of silver is being widely employed (81, 82). To synthesize silver nanoparticles economic, commercially viable as well as environmental friendly approach is still needed.

Nanoparticle synthesis have shown to be better biological method because of its slow kinetics and stability whereas chemical synthesis proves to be toxic when absorbed on the surface in the field of medical (83). Over chemical and physical methods green synthesis is more likely to be environmental friendly, cost effective, easy production for large scale synthesis without the requirement of high pressure, temperature, energy and toxic chemicals (84).

In the industrial and medical field, silver is proven to have inhibitory effects on microbes; it is used to prevent infection against open wounds and burns as typical ointments (85, 86). Due to the advantageous applications in food industries, agriculture, biomedical silver nanoparticles have attracted intensive research interest (87).

3.3.2 Green synthesis

For the development of nanotechnology, green processes consisting of non-toxic precursors and mild reaction conditions have been emphasized; it aims at implementing sustainable processes and minimizing generated waste. It is considered to be an advanced over other methods because of its relative reproducibility, cost effectiveness and in more stable results. Different approaches consisting of physicochemical properties of product, environmental applications and synthesis have been used (88).

production of nanoparticles through microorganisms are slow and comparatively ineffective than using plant based method. For the synthesis of green nanoparticles, high

temperature, energy and pressure and even toxic chemicals are not required, also the contamination rate is lower therefore many scientists nowadays are shifting themselves from artificial and synthetic methods to plant based production of nanoparticles(89).

Due to many advantages, green synthesis is accumulating popularity resulting in a greener environment. Plants parts are extensively used for synthesis of nanoparticles due to its large production.

The different methods and advantages used for the synthesis of nanoparticles are discussed briefly-

- **Microwave assisted technique**

Clean, simple, thermal gradient effect microwave assisted method for the production of oxides, hydroxides and sulphide nanoparticles. The most common advantage of microwave assisted technique over conventional biological technique is the improved rate of reaction due to the penetration and rapid heating involved, resulting in small distribution of particle size (89).

Other advantages include-

- Simple medium.
- Quick reaction.
- Short time for the temperature.
- Controlling the morphology of particles.

- **Mild reducing agent based technique**

For the synthesis of nanoparticles important reducing agents like glucose, starch, amine and a few amino acid like tyrosine. They can be utilised by simple chemical reaction with lower capital investment and less hazardous effluent (89).

- **Microorganism mediated technique**

For the synthesis of both extra and intracellular nanoparticles, a variety of organisms are used. The different biogeochemical cycles which are linked to different groups of bacteria by enzymatic actions reduces the metallic salts to its ionic forms (e.g. nitrate reductase present in nitrogen cycle linked bacteria). For the synthesis of extracellular nanoparticles, different enzymatic actions are taken into action, and for the synthesis of intracellular nanoparticles bacteria is employed by reducing it to a nano form and macrophaging which is afterwards excreted or stored in vacuoles for its disposal and further use. In the different nanoparticle synthesis fungal is considered to be the most advantageous because of its fastidious growth, mycelial meshwork, fabrication and easy handling (89)

- **Plant based technique**

Compared to the microbial mediated nanoparticle synthesis, the plant based technique is considered to be cost effective and requires an aqueous medium rather than solvent for plant extracts. Sometimes the percentage of metal is so high that it exceeds that of the surrounding ecosystem, which is therefore utilized through metallic salt reduction based synthesis (89).

- **Ultrasound assisted technique**

This technique has been extensively used for years for material synthesis and is considered to be the most powerful nanostructure technique. To most commonly employed techniques i.e, sonochemistry and ultrasonic spray pyrolysis are used for the nanoparticle synthesis.

3.3.3 Why plant based technique was chosen?

In ancient times, plant materials were enormously used because of its antimicrobial and antibacterial nature, therapeutic potential, for the treatment of infectious diseases and due

to its healing power. Growth of plant based bioactive agents have opened new avenues in this research field (90).

The plant based herbal products are utilized nowadays because of its:

- 1.) Bulk availability
- 2.) Ease of extractability
- 3.) Antimicrobial efficacy
- 4.) Durability
- 5.) Non- allergic reactions
- 6.) Shelf life
- 7.) Cost-effectiveness

Nowadays, different innovations are significantly required for improving the products. Therefore, the different health care textiles are demanding that the fluids should be retained, absorbed or distributed within the substrates such as nappies, sanitary pads, wound dressings and tampons. The products thus made are required to be non-toxic and non-allergic because they are in direct contact with the skin and antimicrobial treatments are generally given so that they could be protected from certain types of microbes which could certainly stop spread of different infections, can lessen inflammation, can fight vaginal infections, enhance comfort of the wearer, keep unpleasant odour away and maintain personal hygiene.

So, there was a great need for imparting herbal finishes in the sanitary napkins (89).

3.3.4 Taxonomy of different plants which were chosen for the application on fabrics used (90).

- ***Eucalyptus citriodora* Hook. (Safeda)**

Taxonomy

Kingdom: Plantae

Division : Angiosperms

Class :Eudicots

Order: Myrtales

Family: Myrtaceae

Genus :*Corymbia*

Species: *C. citriodora*

Bionomial name: *Eucalyptus citriodora* Hook.

Eucalyptus citriodora Hook., synonym *Corymbiacitriodora* Hook. of family Myrtaceae, is a native to Queensland, Australia which is a tall, graceful, evergreen tree spread in several regions of the world and throughout the tropics , which is commonly known as the “lemon scented gum” and used as a source of nectar in honey production , for the production of essential oil, timbers and fuel woods. The essential of these leaves are used overall as a powerful antiseptic, headache, pneumonia. It is usually applied on on skin infections and to cure external cuts.

- ***Pinuroxburghii*Sarg. (Chir)**

Taxonomy

Kingdom : Plantae

Division: Pinophyta

Class :Pinopsida

Order :Pinales

Family :Pinaceae

Genus :*Pinnus*

Species :*roxburghii*

Binomial name: *Pinusroxburghii*Sarg

Pinus roxburghii Sarg., synonym *Pinus longifolia* Roxb. of Pinaceae family, which is commonly known as chir or pine is usually found in the upper regions of Himalayas with a spreading crown found in the southern Indian hills, Afghanistan and from Kashmir to Bhutan. A clear, transparent oleo-resin is produced by the tapping of the stem which is usually used for the liver disorders, disinfectant and for the insecticides. These plants are reported to have antibacterial and antifungal properties, healing power and are shown to have spasmolytic actions. They also help in the treatment of cough and genito-urinary disorders.

- ***Woodfordia fruticosa* Kurz. (Dhawari)**

Taxonomy:

Kingdom : Plantae

Division: Magnoliophyta

Class: Magnoliopsida

Order: Myrtales

Family: Lythraceae

Genus: *Woodfordia*

Species: *W. fruticosa*

Binomial name: *Woodfordia fruticosa* Kurz.

Woodfordia fruticosa Kurz. belongs to the family of *Lythraceae*, which is commonly known as Dhataki, Dhawari, Dhawi and Dhai are present throughout India, up to an altitude of about 1500m. The leaves are found to be opposite or sub opposite in nature, whereas the barks are cinnamon brown in colour and are smooth. The flowers of these plants are found to be sedative, antibacterial and febrifuge in nature (90).

3.3.5 Extraction and applications techniques for plant extracts.

Several extraction methods are taken into consideration, whose sole purpose is to extract the therapeutically desired portion and to eliminate the inert.

The different techniques taken into consideration for the preparation of plant extracts are as follows:

- (i) **Hot continuous extraction soxhletion:** In this technique, crude extracts are prepared using solvents. The material is placed in a container made of cellulose or cloth with a siphoning device in a central compartment and side arm which is connected to the lower compartment. The reflux condenser is attached above the central compartment and the solvent is placed in the lower compartment. The solvent is then heated till boiling and the vapours are then passed into the reflux condenser through the side arm. The heated solvent percolates through the powder and the extracts are gradually collected in the central compartment and repeated soxhletion is done till the powder decolourizes.
- (ii) **Maceration :** This is the most common method used for the plant extraction. In this technique, the pulverized material is shaken overnight with a solvent which is then followed by evaporation and filtration of the solvent. Repeated maceration is advisable than a single one as it is considered to be more efficient and the active constituents are more valuable.
- (iii) **Ultrasonication:** In this technique, the pulverized material is suspended in the solvent then under the influence of ultrasonic vibrations it is then subjected to cavitations. After that evaporation and filtration is followed, so that the dry extract can be obtained.
- (iv) **Microwave assisted technique:** In this technique, the heating occurs in a closed system. Microwaves have two oscillating perpendicular techniques which are electric and magnetic field. Microscopic traces of moisture in the plant cells serves as a target for microwave heating. After the moisture is heated up due to the effect of microwaves, it gets evaporated and generates a very high pressure on the cell wall which occurs due to the swelling of the plant cells that bursts and extractibles leach out in the solvent.

3.3.6 Application techniques for plant extracts:

Depending upon the active agent and fibre type there are many different approaches which are used for the antimicrobial functionalization. There are two different methods for antimicrobial finishing.

- (i) As an after treatment process or
- (ii) Prior to the extrusion, it can be incorporated into the polymer solution.

Before extrusion, if the different antimicrobial agents are incorporated into the polymer solution there can be a slow migration of the substances which are embedded into the fibre structure to make it active which are only suitable for synthetic fibres.

As an after- treatment process conventional exhaust and pad dry cure method is generally used for antimicrobial finishing. The different methods which are taken into consideration are nanoparticles, nano sized colloidal solutions and the cross linking of the active agent on to the fibre (90).

4. Materials and methods

4.1 Extraction of Nano cellulose from raw cotton

- The substrate used was Raw cotton (10g) from local market (Shoghi)
- Pectinase (1g) from *Aspergillusniger*
- Acetate buffer

The conventional bio scouring was carried out with 1g per litre pectinase in acetate buffer solution at pH 5 at 37°C for 15 hrs. with continuous shaking at 100/min. The scoured cotton was then visualised by measuring how long the cotton took to absorb water.

4.1.1 Preparation method for nanocellulose synthesis

- 12%(w/v) solution NaOH
- 1 M HCl
- 2% (w/v) solution NaOH

Alkaline acid pre-treatment was used for the solubilisation of lignin and hemicellulose. This included the following steps:

1. Cotton fibres were soaked in 12%(w/v) solution for 2 hrs which increased the surface area of the fibres that helped the hydrolysis.
2. Cotton fibres were then soaked in 1M solution at 70°C, which solubilized the hemicelluloses.
3. After that the fibres were treated with 2% (w/v) solution for 2 hrs.at 70°C , which disrupted the lignin structure , and the linkages between carbohydrates and lignin were then broken.

Alkaline pre-treatment is an effective method which can improve the yield and remove lignin and hemicellulose from the fibres.

4.1.2 Mechanical method

Cryo crushing

This method was used to break the cellulose wall in the nanocellulose fibres in which water soaked cellulose fibres were immersed in liquid nitrogen, which solidified the water content and after that it was subsequently crushed by mortar and pestle (91). By applying high impact forces on cellulosic fibres rupturing occurred due to the applied pressure through the ice crystals which converted it into nano cellulose.

4.1.3 Chemical hydrolysis

The nanocellulose fibres which were obtained after the mechanical methods, consisted of both crystalline and amorphous regions and in order to dissolve amorphous region and permit the longitudinal cutting of the microfibrils, acid hydrolysis treatment was again performed (91).

4.2 Extraction of plant extract

- Eucalyptus leaves were taken from JUIT campus, Solan.

Maceration technique was used in which the leaves of eucalyptus were grinded and boiled for 2 hrs at 60°C in a water bath, and the pulverised material which was obtained was then shake overnight followed by filtration and evaporation (91).

Repeated maceration was done for efficient results.

4.3 Synthesis of nanoparticles

- 10 ml Silver Nitrate
- 10 ml eucalyptus extract

In a typical reaction, 1ml eucalyptus extract was added to 9 ml aqueous Silver nitrate (pH- 9) and the mixture was then kept in the autoclave at 15 psi pressure, 121°C for 5 minutes and 10 minutes. The change in the colour indicated the synthesis of nanoparticles.

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