Experimental Procedure

3. EXPERIMENTAL PROCEDURE

The experimental procedure pertaining to the study consisted of the following five phases.

3.1	Phase – I	: Pilot Study – Selection of Suitable Medicinal Herbs, Solvent, Material, Extraction Procedure, Optimization of Finish Process Parameters, Mordant, Finishing Methods, Microorganisms, Antibiotics and Antimicrobial Tests
3.2	Phase – II	: Application of Herbal Extraction on Cotton Fabrics and Evaluate the Antimicrobial Activity against Bacteria and Fungi
3.3	Phase – III	: Evaluating the Antimicrobial Effectiveness, Physical and Mechanical Property, Durability and Functional Property of Herbal Extractions
3.4	Phase – IV	: Product Development and their Performance
3.5	Phase – V	: Statistical Analysis

3.1 PHASE – I : PILOT STUDY – SELECTION OF SUITABLE MEDICINAL HERBS, SOLVENT, MATERIAL, EXTRACTION PROCEDURE, OPTIMIZATION OF FINISH PROCESS PARAMETERS, MORDANT, FINISHING METHODS, MICROORGANISMS, ANTIBIOTICS AND ANTIMICROBIAL TESTS

Pilot study is a preliminary study conducted to gain some primary information, on the basis of which the main project would be planned and formulated, as stated by Saravanavel (1999). Hence, a pilot study was carried to select the medicinal plants, solvents, material, extraction, application techniques, optimization of process parameters, microorganisms, antibiotics, material liquor ratios and antimicrobial tests.

3.1.1 Selection of Medicinal Plants

Plants have been one of the important sources of medicines since the dawn of human civilization. They are the only economical source of a number of well established and important drugs, as described by Aggarwal et al. (2007). A number

of natural plants are used in the traditional medical systems in many countries. It contains a wide range of undiscovered valuable medicinally active phytoconstituents that can be used to treat chronic as well as infectious diseases and alternative medicine for treatment of various diseases is getting popular also in western countries, as highlighted by Diallo et al. (1999), Brantner and Grein (1994), Soudahmini et al. (2005) and Essawi and Srour (2000). The plant-based, traditional medicine system continues to play an essential role in health care, with about 80 per cent of the world's inhabitants relying mainly on traditional medicines for their primary health care, as expressed by Owolabi et al. (2007).

Medicinal plants are part and parcel of human society to combat diseases from the dawn of civilization, as described by Kausik et al. (2002). In the last few decades medicinal plants have been the subjects of very intense pharmacological studies, as stated by Packialakshmi (2010). Due to their inherent distinct chemical and biological properties medicinal plants claim a unique place in development of natural products in health care, as stated by Nair et al. (2005) and Khan and Khanum (2008). Some of the bioactive compounds and antioxidants of plants which act on the human body are alkaloids, tocopherols, ascorbic acid, carotenoids, flavonoids, tannins and phenolic compounds, as described by Edeoga et al. (2005) and John and Malathi (2010). A number of Indian herbs do contain antibacterial, antifungal and antiviral properties. Some specific species of herbs having antimicrobial activity are suitable for textile application, as reported by Sarkar et al. (2003). Even today native communities use folk medicine for the treatment of common infections, as denoted by Rojas et al. (2006) and Gupta et al. (2005). But their properties and actual action remain unrevealed calling for scientific investigation as quoted by Nascimento et al. (2000). Considering the above facts a variety of Indian herbal plants are selected for the extraction of antimicrobial compounds. An in depth review of literature and the antimicrobial activities made the investigator to select aloe vera leaves, neem leaves, marigold flowers, kuppaimeni leaves, yashtimadhu roots, tanner's cassia flowers, tridex daisy leaves, vettiveru roots and flax seeds for the pilot study.

Aloe vera (*Aloe barbadensis*) called the 'miracles plant' or the 'natural healer' belonging to family Liliaceae is known as 'Lily of the desert'. It is known for its medicinal and healing properties. Aloe vera has been used in traditional medicinal practices of many cultures for a host of curative purposes ranging from dermatitis to

cancer, healing of wounds and burns, various skin conditions namely skin disorders, softening the skin, removing dead skin, making skin look young, pimple free and helps in antiageing, speeding up the rate of healing, reducing the risk of infection and scale preventing properties and finds uses in medical field and as an excellent moisturizer that keeps the skin flexible by giving oxygen to the cells, which in turn increases the strength and synthesis of the skin tissue, the hydrating capacity of the skin. Cosmetic products from make up to anti-wrinkle creams to facial masks, to conditioners and lipsticks, reduces the intensity of pigmentation, as well as for general health, as highlighted by Rodriguez et al. (2005), Singh and Maurya (2005) and Kemper and Chiou (1999). Aloe vera also possesses antifungal, antiseptic, antibiotic, anti-inflammatory, wound healing, anti tumour, immune modulating and antibacterial properties which can be exploited for medical textile applications such as wound dressing, suture, bioactive textiles, as revealed by Harini et al. (2007) and Anonymous (2008). In modern times, scientific research has shown that the aloe leaf contains over 75 nutrients and 200 active compounds including 20 minerals, 18 amino acids and 12 vitamins, enzymes, sugars, lignin, anthraquinones, saponins, salicylic acid and amino acids which has been used in the USA since the 1970s and is found today in virtually all cosmetic products, as reviewed by Park and Jo (2006). Aloe vera gives aloe gel, which has antimicrobial activity against various microbes and inhibited the growth of *Mycobacterium, Trichophyton* and *Bacillus subtilis* (http://www.internethealthlibrary. com).

Marigold (*Calendula officinalis*) has been used traditionally for treatment of wounds, sores, skin ulcerations, eczema, fever, boils, abscesses, bruises, sprains, foot cracks, pulled muscles, boils and to prevent recurrent vomiting. The whole flower heads (Calendula flos cum calyce) and petals (Calendula flos sine calyce) are used for medicinal and culinary purposes. More recently, anti-tumour and anti-oxidant properties of *Calendula* species have been established and utilized in research treatments for various cancers, as reviewed by Hadfield et al. (2008). The medicinal rating of aloe vera and pot marigold was reported as 5 star, (www.pfaf.org.).

The plant, kuppaimeni (*Acalypha indica* Linn.) is commonly known as Indian acalypha and it belongs to the family Euphorbiaceae, found in all parts of the tropics. It is found mostly in the backyards of houses and waste places through the plains of India. The plant is used as an expectorant in asthma, pneumonia emetic, emenagogue, anthelmintic in rheumatism and several other ailments, as reveald by

Shivayogi et al. (1999) and Bourdy and Walter (1992). The dried leaves of *Acalypha indica* was made into a poultice to treat bedsores, wounds and the juice of *Acalypha indica* is used to treat a variety of skin disorders. *Acalypha indica* contains acalyphine which is used in the treatment of sore gums, as said by Bedon and Hatfield (1982). It also has anti venom, wound healing, antioxidant activities, anti-inflammatory effects, acaricidal effects, diuretic effects and antibacterial activities, as stated by Ruchi et al. (2007), Annie et al. (2004), Suresh Reddy et al. (2002), Ayyanar and Ignacimuthu (2009), Singh et al. (2004), Das et al. (2005) and Govindarajan et al. (2008) respectively.

Neem (*Azadirachta indica*) is an evergreen tree of India. It has been used as a traditional medicine against various human ailments from ancient times in India and about 700 herbal preparations based on neem are found in Ayurveda, Siddha, Unani, Amchi and other local health prescriptions, as described by Chatterjee and Pakrashi (1994). The active ingredients of neem are found in all parts of the tree but in general, seed, bark, leaves and roots which are used for herbal extractions. More than 300 different active compounds have been reported from different parts of neem tree, but the most important limonoids are azadirachtin, salannin and nimbin, as pointed out by Schmutterer (1995). The neem extracts have been widely recognized as one of the most promising sources of compounds with insect control, antimicrobial and medicinal properties, as suggested by Singh et al. (1996).

Yashtimadhu (*Glycyrrhiza glabra*), belongs to genus *Glycyrrhiza*, commonly called as licorice or yashtimadhu is available in India. It is used as a remedy for skin infections and painful conditions. This herb claimed the efficacy of *Glycyrrhiza* species for a variety of pathological conditions indicated in traditional medicine for coughs and colds, as reported by Chopra et al. (2002) and Meghashri and Gopal (2009). Tanner's cassia (*Cassia auriculata* L.) commonly known as tanner's cassia also known as avaram in Tamil language is a shrub belongs to the Caesalpiniaceae family. The shrub is specially famous for its attractive yellow flowers which are used in the treatment of skin disorders, body odour and rheumatism, as remarked by Maneemegalai and Naveen (2010), Gaikwad et al. (2010) and Murugan (2010).

Tridex daisy (*Tridax procumbens*) is a hispid soberest annual herb, the leaves are 2-5 cm long.The extract of *Tridax procumbens* has been reported to have various pharmacological effects including antimicrobial activity against positive and negative bacteria (Patel et al., 2004, http://www.siv.edu/cbe and Nazeruddin et al., 2011).

Tridax procumbens exhibits antiseptic, insecticidal, hair growth promotion property, antiviral and antibiotic efficacies.The leaf juice can be used to cure fresh wounds and stop bleeding from incised wound of different kinds and stages, as quoted by Kurian (1984), Rasik et al. (1999) and (http://www.greenpharmacy.info/article .asp). The analysis of phytochemical compounds of *Tridax procumbens* L. showed good results for all major phytochemicals except acidic compounds, alkaloids, flavonoids and terpenoids, as stated by Siddiqui et al. (2009). The methanol extract of *Tridax procumbens* L. showed antibacterial activity against test organisms with the zones of inhibition ranging from 8-17 mm revealing its great medicinal potential for the treatment of microbial induced ailments. *Vetiveria zizaniodes* (L.), popularly known as kush-grass has been known in India since ancient time. It is the major source of the well-known oil of vettiver, which is used in medicine and in perfumery, as stated by Rao and Suseela (2000). The root extract is used for toothache, the leaf paste is used for lumbago, sprain and rheumatism, boils, burns, epilepsy, snakebite and sores in the mouth as revealed by Jain (1991) and Luqman et al. (2005). Flax seed most frequently is used as a laxative. It is also used in curing arthritis, acne, eczema and psoriasis (http://www.bodybuildingforyou.com). The details of the selected herbs are presented in Table – 2 and Plate – 1.

TABLE – 2

LIST OF SELECTED MEDICINAL PLANTS

Common name	Botanical name	Parts used	Family
Aloe vera	*Aloe barbadensis*	Leaves	Liliaceae
Marigold	*Calendula officinalis*	Flower	Asteraceae
Kuppaimeni	*Acalypha indica*	Leaves	Anacardiaceae.
Neem	*Azdirachta indica*	Leaves	Meliaceae
Yashtimadhu	*Glycyrrhiza glabra*	Roots	Solanaceae
Tanner's Cassia	*Cassia auriculata*	Flower	Caesalpiniaceae
Tridex Daisy	*Tridax procumbens*	Leaves	Asteraceae
Vettiveru	*Vetiveria zizanioides*	Roots	Graminae
Flax seed	*Linum usitatissimum*	Seed	Linaceae

3.1.1.1 Collection of Plants for Extraction

The selected medicinal plants were collected from the Tamil Nadu Agricultural University and Herbal Ayurvedic Shops in Coimbatore. The leaves, flowers, roots and seeds were washed thoroughly with soft water to remove extraneous matter and dried in shade.

ALOE VERA

MARIGOLD

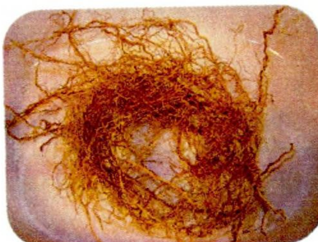
KUPPAIMENI

NEEM

YASHTIMADHU

TANNER'S CASSIA

TRIDEX DAISY



VETTIVERU

FLAX SEED

PLATE – 1

SELECTED MEDICINAL PLANTS

3.1.2 Selection of Solvent

The traditional techniques of solvent extraction for flavonoids from plant materials are mostly based on the correct choice of solvents and the use of heat agitation to increase the solubility of the desired compounds, as revealed by Zhang et al. (2009). A series of non polar to polar solvents like hexane, benzene, chloroform, diethyl ether, ethyl acetate, acetone, ethanol, methanol and water in 25, 50, 75 and 100 per cent give a first indication of the nature of the extracted substances, as viewed by Majorie and Kubo (1999), Bhattacharya and Patel (2003) and John and Malathi (2010). Herbs and spices can be extracted using distilled water and tested for its antimicrobial effect against *E. coli,* the most common intestinal non pathogenic organism, as stated by Amrita et al. (2009).

Normally herbal extraction is carried out by aqueous, alkaline, acidic or alcoholic methods. Water is universally known as a safe solvent. Initial screening of plants for possible antimicrobial activities typically begins by using crude aqueous or alcohol. The exceptional water-soluble compounds such as polysaccharides and polypeptides including fabatin (Zhang and Lewis, 1997) and various lectins are commonly more effective as inhibitors of pathogen adsorption and cannot be identified in the screening techniques. Occasionally tannins and terpenoids will be found in the aqueous phase, but they are more often obtained by treatment with less polar solvents (Appendix – I).

Organic solvent extraction methods are suitable to verify antibacterial activity and sensitivity of microorganisms against human pathogens including bacteria, fungi or virus, as described by Elastal et al. (2005). The row indicating the number of inhibitors extracted with each solvent points to two implications : first, that most active components are not water soluble and second, the most commonly used solvents (ethanol and methanol, both used as initial extractants in approximately 35 per cent of the studies appearing in the recent literature) may not demonstrate the greatest sensitivity in yielding antimicrobial chemicals on an initial screening. A suitable solvent for extracting the raw flavonoids is ethanol, it is also environmentally safe, as stated by Li et al. (2008), Zhang et al. (2003), He et al. (2005) and Sathishkumar et al. (2008). The ethanol and methanol extracts are also used for analysing the secondary metabolities such as carbohydrates, alkaloids, glycosides, flavonoids, tannins, steroids, saponins, triterpenoids, protein, resins, fixed oils and fats using standard procedures of analysis .

Hence, the solvents namely distilled water, ethanol and methanol were chosen on the basis of solvent polarity, solubility, volatility, economy, safety, nature and yield level of herbs.

3.1.3 Selection of Extraction Procedure

The efficient extraction of herbs while preserving their medicinal values is an art as well as science, as defined by Shrivastava (2006). Extraction refers to the separation of the desired material by physical or chemical means with acid as solvent. The choice of solvent, temperature and time determine the efficiency of extraction. The selected medicinal herbs were subjected to fresh and dry extraction method. In fresh extraction method, ten gms of fresh parts of selected medicinal plant namely leaves, flowers, roots and seeds except Aloe vera gel were ground with 100 ml of water, as remarked by Vankar et al. (2009). For aqueous extraction, plant parts of all the selected herbs were soaked in distilled water, blotted dry, made into slurry through blending. The solution was kept in a shaker for 24 hours and then filtered. This was then centrifuged for 30 min. The procedure was repeated twice. In the same method ethanol and methanol extracts were also prepared. These extracts were evaporated using flash evaporator and dried at 60°C. The dried residue was taken scraped from the plate and diluted with required percentage of water used for the study and screened against selected bacterial strains, as pointed out by Kivcak et al. (2002). For aloe vera, the aloe leaves were collected and washed with fresh water. Aloe vera gel was processed using the hand filleted processes. The inner gel was carefully removed avoiding the yellow sap (latex) found next to the rind. The latex contains aloin which was removed. Stabilizing the constituents within aloe vera gel was essential to develop effective products. Therefore the gel was cooled and processed within four hours of harvesting. This gel was smashed and converted into a solvent form.

The dry extraction method was used for all the parts of the plant namely leaves, flowers, roots and seeds of selected medicinal plants. Except aloe vera gel, the other plant parts of raw materials were cleaned with tap water and shade dried at room temperature for 48 hours till the moisture content was completely removed, as indicated by Chengaiah et al. (2009) and Sundararajan and John (2010). The root of the plants were air dried under shade for six consecutive weeks at room temperature. Great care was taken during drying to prevent contamination of the compounds. After drying, chopping and grinding was carried out to break down the parts of the plant

into very small units ranging from coarse fragments to fine powder, as expressed by Samanta et al. (2007) and Vankar et al. (2009). Aloe vera gel and selected parts of plants were dried and powdered.

Ten grams of the prepared dry powder of all the plants were taken in separate conical flask and mixed with the 100 ml of water, ethanol and methanol solvents in the ratio of 1 : 5. Cotton was plugged into the head of the conical flask and wrapped with the foil paper to prevent the evaporation of the solvent. These air tight flasks were kept on a rotary shaker at 190 – 220 rpm overnight. The temperature was maintained at 40°C to dissolve the active compounds. The supernatant was collected and filtered using filter paper to remove the extraction solution from the supernatant. The extracted solvents from the ethanol and methanol were evaporated under reduced pressure through flash evaporator to condense the compounds as suggested by Sithan and Kamaraj (2010). The solvents were allowed to evaporate for one hour and the extraction process was repeated twice. The dried extract was taken, centrifuged, filtered and the residue was scraped from the plate.

The water extract was taken from the shaker and boiled for two hours in mild heat and then cooled. The cooled extracts were filtered and evaporated for 24 hours. The dried powder was later stored at 4°C in air tight bottles for further studies as pointed out by Sofowora (1982). The extraction efficiency was quantified by determining the weight of each of the extracts (Gidado et al., 2005) (Plate – 2). Based on the extraction efficacy in different medium of solvents, nature of herbs and percentage yield of herbs, the fresh extraction method for aloe vera and dry extraction method for all the other herbs were selected for the final study.

3.1.3.1 Screening of Antimicrobial Property

The extracts of all the herbs were screened by Thin layer chromatography (TLC) and Agar well diffusion test methods for identifying the phytochemical compounds and antimicrobial effectiveness of the herbs against the gram positive and gram negative bacterias. The pure compounds were isolated from natural plant extracts using initial screening process.This exhibits high potent antibacterial properties, as stated by Kusumoto et al. (1995), Cowan (1999), Ramachandran et al. (2004), Kavitha et al. (2006), Thilagavathi et al. (2005), Ushamalini and Naik (2008). The maximum inhibitory effect was exhibited by the leaf extracts, as said by Barooah and Konwar (2010).The colour reflection of herbal extraction on TLC plates and the

ALOE VERA STEM

EXTRACTION OF GEL

MARIGOLD POWDER

MARIGOLD ETHANOL EXTRACTION

KUPPAIMENI POWDER

KUPPAIMENI ETHANOL EXTRACTION

NEEM POWDER

NEEM DISTILLED WATER EXTRACTION

YASHTIMADHU POWDER

YASHTIMADHU DISTILLED WATER EXTRACTION

TANNER'S CASSIA POWDER

TANNER'S CASSIA METHANOL EXTRACTION

TRIDEX DAISY POWDER

TRIDEX ETHANOL EXTRACTION

VETTIVERU POWDER

VETTIVERU ETHANOL EXTRACTION

FLAX SEED POWDER

FLAX SEED DISTILLED WATER EXTRACTION

PLATE – 2

EXTRACTION OF MEDICINAL PLANTS

zone of inhibition obtained from all the different medium of extracts of antimicrobial activity was measured in mm. The zone was compared with all the medium of solvent extracts against *Staphylococcus aureus* and *Escherichia coli* bacteria growth (mm). It has been suggested by Chung et al. (1995) and Vlietinck et al. (1995) that aqueous and ethanolic extracts from plants used in allopathic medicine are potential sources of antiviral, antitumoral and antimicrobial agents. The agar well diffusion test method showed the maximum zone of inhibition for all the selected herbs in particular solvents against both positive and negative bacteria. The maximum zone of inhibition was found in the solvents for the selected herbs namely distilled water for aloe vera (27 mm x 26 mm), ethanol for marigold (23 mm x 24 mm), ethanol for kuppaimeni (23 mm x 27 mm), distilled water for neem (27 mm x 24 mm), distilled water for yashtimadhu (27 mm x 25 mm), methanol for tanner's cassia (18 mm x 20 mm), ethanol for tridex daisy (19 mm x 18 mm), ethanol for vettiveru (18 mm x 18 mm) and distilled water for flax seed (17 mm x 16 mm). Based on the higher zone of inhibition, antimicrobial compound present in the herbs, herb yields in the solvents, cost effectiveness of solvents, the solvents and herbs such as distilled water for aloe vera, neem, yashtimadhu, ethanol for marigold and kuppaimeni were identified as the best solvents for the medicinal herbs. Hence, these best parameters were used for developing the research work.

3.1.4 Selection of Optimization of Finish Process Parameters for the Application of Herbal Extracts on Fabrics

Optimization is defined as the process of trying to find maxima and minima of function and also the methodologies used for improving the efficiency of the production process (www.wikipedia.com). The response surface method of Box-Benkhen design model is widely used to optimize the various process parameters used in textile industry, being reported by Iqbal and Pramanik (2011). The dried-ground leaves (2.5, 5, 7.5, 10 and 12.5 gm each) were soaked in sufficient water (app. 200-250 ml) at 70-75°C for 0.5, 1.0, 1.5 and 2 hours. Later the dye extract was filtered through ordinary filter paper and the filtrate was collected, conditioned for 30 mins, 60 mins and 90 mins respectively. After conditioning the absorbance was recorded for determination of concentration as discussed by Jayalakshmi and Amsamani (2008). The three temperatures namely 40°C, 50°C and 60°C was used for optimizing the plant extract finish parameters. The concentration of solute is usually expressed as the amount of a solute in a unit volume of a solution. The

amount of a solute can be in grams, kilograms and moles. The unit volume of a solution is always in litres, as revealed by Fan (2005).

The Box-Behnken design is an independent quadratic design, as reported by Thilagavathi and Kannaian (2008). This design is rotatable and requires three levels of each factor. Thus the optimization method was carried out using Box and Behnken response surface design with three independent variables at three levels of each variable. Since time, temperature and concentration play a major factor which influences the antimicrobial activity against bacteria and fungi organisms these variables were taken into consideration.

The three different concentrations namely 25 per cent, 50 per cent and 75 per cent of plant extracts at three different temperature (40°C, 50°C and 60°C) followed by 30 mins, 60 mins and 90 mins duration were taken for treatments. The independent variables were named as time of extract treatment (X_1), treatment temperature (X_2) and concentration of the herbal solution (X_3). The dependent variable (Y) was the microbial zone of inhibition by agar well diffusion test method (www.biomedcentral.com). The Box-Behnken three level and three variable experimental design for finishing process optimization is shown in Table – 3.

TABLE – 3

BOX AND BEHNKEN EXPERIMENTAL DESIGN VARIABLES AND CODED LEVELS

Variables	Coded levels		
	-1	0	+1
Time (in mins) (X_1)	30	60	90
Temperature (°C) (X_2)	40	50	60
Concentration (%) (X_3)	25	50	75

The various parameters of the extraction process were analyzed statistically. Effects of single factors such as temperature, time and concentration were initially investigated. On the basis of Box and Behnken three level and three variable experiments,15 trials were taken for different combinations and each combination showed the response in the form of microbial zone of inhibition in mm.

TABLE – 4

BOX AND BEHNKEN EXPERIMENTAL DESIGN FOR ALOE VERA

Test order No.	Time (mins) (X_1)		Temperature (°C) (X_2)		Concentration (%) (X_3)		Response Factor zone (Y) (mm)
	Coded	Actual	Coded	Actual	Coded	Actual	
1	0	60	-1	40	-1	25	15.1
2	-1	30	1	60	0	50	15.8
3	**0**	**60**	**0**	**50**	**0**	**50**	**22.4**
4	-1	30	0	50	1	75	17.3
5	1	90	-1	50	0	50	16.8
6	0	60	1	60	1	75	19.2
7	**0**	**60**	**0**	**50**	**0**	**50**	**22.4**
8	0	60	-1	40	1	75	20.7
9	-1	30	0	50	-1	25	13.2
10	**0**	**60**	**0**	**50**	**0**	**50**	**22.4**
11	0	60	1	40	-1	25	18.9
12	1	90	0	50	1	75	18.3
13	1	90	1	60	0	50	17.2
14	-1	30	-1	40	0	50	15.3
15	1	90	0	50	-1	25	14.8

$$Y = 22.40 + 0.63\,X_1 + 0.45\,X_2 + 1.68\,X_3 - 4.40\,X_1^2 - 1.82\,X_2^2 - 2.10\,X_3^2 + 0.07\,X_1X_2 - 0.15\,X_1X_3 - 1.32\,X_2X_3$$

(R = 99.61% ; R^2 = 98.91%)

TABLE – 5

BOX AND BEHNKEN EXPERIMENTAL DESIGN FOR MARIGOLD

Test Order No.	Time (mins) (X_1		Temperature (°C) (X_2)		Concentration (%) (X_3)		Response Factor Zone (Y) (mm)
	Coded	Actual	Coded	Actual	Coded	Actual	
1	0	60	0	50	0	50	20.9
2	0	60	-1	40	1	75	23.4
3	-1	30	0	50	-1	25	12.9
4	0	60	-1	40	-1	25	17.3
5	0	60	0	50	0	50	20.9
6	-1	30	-1	40	0	50	17.9
7	1	90	1	60	0	50	21.1
8	0	60	1	60	-1	25	17.2
9	0	60	0	50	0	50	20.9
10	-1	30	0	50	1	75	20.2
11	**0**	**60**	**1**	**60**	**1**	**75**	**24.9**
12	1	90	0	50	1	75	22.2
13	1	90	-1	40	0	50	18.7
14	-1	30	1	60	0	50	18.4
15	1	90	0	50	-1	25	16.9

$$Y = 20.90 + 1.21\,X_1 + 0.53\,X_2 + 3.32\,X_3 - 2.28\,X_1^2 + 0.41\,X_2^2 - 0.61\,X_3^2 + 0.47\,X_1X_2 - 0.55\,X_1X_3 + 0.40\,X_2X_3$$

(R = 98.97% ; R^2 = 97.12%)

TABLE – 6

BOX AND BEHNKEN EXPERIMENTAL DESIGN FOR KUPPAIMENI

Test Order No.	Time (mins) (X_1)		Temperature (°C) (X_2)		Concentration (%) (X_3)		Response Factor Zone (Y) (mm)
	Coded	Actual	Coded	Actual	Coded	Actual	
1	0	60	1	60	1	75	20.9
2	0	60	0	50	0	50	19.3
3	0	60	1	60	-1	25	15.8
4	-1	30	0	50	-1	25	15.3
5	1	90	0	50	-1	25	15.9
6	1	90	-1	40	0	50	18.4
7	0	60	0	50	0	50	19.3
8	0	60	-1	40	-1	25	15.6
9	-1	30	0	50	1	75	21.0
10	1	90	1	50	0	50	18.9
11	1	90	0	50	1	75	22.1
12	-1	30	-1	40	0	50	17.8
13	-1	30	1	60	0	50	17.8
14	0	60	0	50	0	50	18.3
15	0	60	-1	40	1	75	20.4

$$Y = 18.96 + 0.42 X_1 + 0.15 X_2 + 2.72 X_3 - 0.17 X_1^2 - 0.57 X_2^2 - 0.22 X_3^2 + 0.12 X_1X_2 + 0.12 X_1X_3 + 0.07 X_2X_3$$

(R = 98.16% ; R^2 = 94.85%)

TABLE – 7

BOX AND BEHNKEN EXPERIMENTAL DESIGN FOR NEEM

Test Order No.	Time (mins)(X_1)		Temperature (°C) (X_2)		Concentration (%) (X_3)		Response Factor Zone (Y) (mm)
	Coded	Actual	Coded	Actual	Coded	Actual	
1	0	60	1	60	1	75	18.6
2	-1	30	0	50	1	75	21.1
3	-1	30	1	40	0	50	21.0
4	0	60	0	50	0	50	17.2
5	1	90	1	60	0	50	19.9
6	0	60	-1	40	-1	25	15.4
7	1	90	0	50	-1	25	16.9
8	0	60	1	60	-1	25	16.7
9	-1	30	1	60	0	50	20.9
10	0	60	0	50	0	50	17.2
11	-1	30	0	50	-1	25	21.3
12	1	90	0	50	1	75	20.7
13	1	90	-1	40	0	50	18.4
14	0	60	0	50	0	50	17.2
15	0	60	-1	40	1	75	18.9

$$Y = 17.2 - 1.05 X_1 + 0.30 X_2 + 1.12 X_3 + 2.72 X_1^2 - 0.12 X_2^2 + 0.07 X_3^2 + 0.40 X_1X_2 + 1.00 X_1X_3 - 0.40 X_2X_3$$

(R = 98.86% ; R^2 = 96.81%)

TABLE – 8

BOX AND BEHNKEN EXPERIMENTAL DESIGN FOR YASTHIMADHU

Test Order No.	Time (mins) (X_1)		Temperature (°C) (X_2)		Concentration (%) (X_3)		Response Factor Zone (Y) (mm)
	Coded	Actual	Coded	Actual	Coded	Actual	
1	1	90	1	60	0	50	18.4
2	**0**	**60**	**0**	**50**	**0**	**50**	**20.9**
3	-1	30	-1	40	0	50	15.9
4	1	90	0	50	-1	25	16.7
5	-1	30	1	60	0	50	16.3
6	0	60	1	60	-1	25	17.8
7	**0**	**60**	**0**	**50**	**0**	**50**	**20.9**
8	0	60	1	60	1	75	20.2
9	**0**	**60**	**0**	**50**	**0**	**50**	**20.9**
10	-1	30	0	50	1	75	18.4
11	1	90	0	50	1	75	19.6
12	0	60	-1	40	-1	25	16.9
13	1	90	-1	40	0	50	17.4
14	-1	30	0	50	-1	25	16.5
15	0	60	-1	40	1	75	19.5

$$Y = 20.90 + 0.62\,X_1 + 0.37\,X_2 + 1.22\,X_3 - 2.35\,X_1^2 - 1.55\,X_2^2 - 0.75\,X_3^2 + 0.15\,X_1X_2 + 0.25\,X_1X_3 - 0.05\,X_2X_3$$

(R = 98.65% ; R^2 = 96.21%)

Here

Y = Antimicrobial efficacy of herbal finished samples (zone in mm)

X_1 = Treatment time

X_2 = Treatment temperature

X_3 = Concentration of herbal solution

The maximum zone of inhibition obtained from each trial at particular test order was selected as the best parameters for herbal finishing. The herbal extract of all the plants were applied on cotton fabric by pad dry and micro encapsulation techniques using the optimized process parameters by the same process sequence are furnished below in Table – 9.

TABLE – 9

OPTIMIZED FINISH PROCESS PARAMETERS FOR SELECTED HERBS USING BOX-BEHNKEN EXPERIMENTAL DESIGN

Medicinal plants	Optimized finish process parameters		
	Time (mins) (X_1)	Temperature (°C) (X_2)	Concentration (%) (X_3)
Aloe vera	60	50	50
Marigold	60	60	75
Kuppaimeni	90	50	75
Neem	30	50	25
Yasthimadhu	60	50	50

3.1.5 Optimisation of pH

pH is a scale between 0 and 14 used to express the concentration of hydronium (H_{30} + or H^+ ions in a solution. It is defined by equation ph = log (H^+). Bacteria does not grow at pH below 4.6. However, pH of human body range between 6.8 and 7.4, which is slightly basic, as reported by Orhan et al. (2007). Hence 7, neutral pH was selected for the study.

3.1.6 Selection of Material

Cotton is a cool, soft, comfortable, principle clothing fibre of the world. Its production is one of the major factors in the world prosperity and economic stability (http://www.fabrics/net/cotton.asp).The highlights of cotton are its properties like comfort, absorbency, ability to arrest microorganism's entry and its easy care, as remarked by Gupta and Laha (2007). The two main drawbacks of cotton are susceptibility to creasing and bacterial degradation. Human sweat provides a suitable shelter for bacterial growth, containing 1.4 million bacteria per gram which increases to 9000 million at 50 per cent moisture level, as remarked by Choudhary et al. (2009).

Cotton textiles in contact with human body offer an ideal environment for microbial growth providing oxygen, water and warmth as well as nutrients from spillage and body exudates. Cotton being a cellulosic material contains a large number of hydroxyl (OH) groups and few carboxyl (OH-C = O) groups which are polar in nature. Hence, 100 per cent plain weave cotton made up of 60^S count yarn with 82 ends / cm and 64 picks / inch and 63 GSM of fabric was selected for the study. The fabric details are shown in Appendix – II.

3.1.7 Selection of Mordant and Material Liquor Ratios

Mordants are the substances which create a link between the dye stuff and the fibre. It remains in the fibre permanently holding the dye, improving the quality of the fabric with better colour (http://oecotextiles.wordprocess.com). Mordants should be colourless, odour less, evenly thick, smooth with good adhesion property. Natural extracts require mordants in the form of metallic salt to produce an affinity between the fibre and dyes. The use of mordant increases the fastness property by forming an insoluble compound of the dye with material, as revealed by Aung and Win (2008) and Shivaprakash (2010). Mordants are eco-friendly in nature and does not affect the fabric or the finish applied. The most commonly used safe mordant is alum. It is usually used with as an additive, as remarked by Kavitha et al. (2006). Hence alum was selected as a fixing agent for all the herbs. Studies carried out by Singh and Gehlot (2009) proved that 5, 10 and 15 per cent of alum as best suited for herbal application. Considering the above facts mordanting was done using 10 per cent alum at 60°C.

Material liquor ratio may be defined as volume of liquor to be taken in dye bath in proportion to weight of textile. The higher the affinity or strike rate of dye for fibre a higher liquor ratio would ensure formation of leveled shades. Dyes with higher affinity if applied forms a lower liquor ratio bath are bound to form unleveled shades, as remarked by Chakraborty (2010).The extracts were applied on to the cotton fabric using the optimized finish process conditions and along with the material liquor ratio of 1:10. According to Prince et al. (2010) the 1:10 material liquor ratio is followed for the application of finishes to the pure cotton woven fabric. Therefore the same material liquor ratio of 1:10 was selected for the study.

3.1.8 Selection of Functional Finishing Method

The application of herbal antimicrobial finishes on textiles has prevailed in the past when Egyptian mummies were wrapped with herbs and spices in order to preserve them. Clinical microbiologists have two reasons to be interested in the topic of antimicrobial plant extracts. First, it is very likely that these phytochemicals will find their way into the arsenal of antimicrobial drugs prescribed by physicians around the world. Second, the public is becoming increasingly aware of problems with the over prescription and misuse of traditional antibiotics.

Effective control of microorganisms particularly bacteria and fungi can be achieved by use of antimicrobial agents, as viewed by Wurtz and Pirmasens (2004). The use of antimicrobial finishing within textiles can help to avoid or control cross infection and extend life time of the product by stopping microbial growth,as highlighted by Bunce (2004). In addition many people are interested in having more autonomy over their medical care. Antimicrobial textiles based on eco-friendly agents which not only help to reduce effectively the ill effects associated due to microbial growth on textile material but also comply with the statutory requirements imposed by regulating agencies. Hence the herbal antimicrobial finish was selected for the study.

3.1.9 Selection of Microorganisms

Microbes are ubiquitous, part of our every day lives in nature and can cause innumerable trouble to both human beings and also to textile materials, as quoted by Srivastava (2010). Even the "clean" skin has a microbe's population ranging between 100-1000 / cm^2. The microbes can multiply at a faster rate and produces odour, discoloration of textiles and sufficient infection of skin, (http://www.articleonramp.com).

The most trouble causing organisms are fungi and bacteria. The growth of microorganisms on textiles can lead to functional, hygienic and aesthetic difficulties, as described by Harini et al. (2007). *Staphylococcus aureus* and *Aspergillus niger* are cloth damaging organisms,as described by Ramachandran et al. (2004). *Staphylococcus aureus* causes wound infection, pyogenic infection, pneumonia and toxic shock. It contaminates under garments and can cause infections like furunels and boils diseases of skin, as suggested by Teli and Prabhu (2006), Aggarwal et al. (2007) and Kan and Yuen (2006).

E. coli are non-pathogenic in normal conditions but if present in excess, will become causative agent of various diseases like urinary tract infection, diarrhea and vomiting, as expressed by Amrita et al. (2009). *Aspergillus niger* is a filamentous ascomycete fungus that is ubiquitous in the environment and has been implicated in opportunistic infections of human, as remarked by Perfect et al. (2001) and Baker (2006). *A. niger* is most widely known for its role as a citric acid producer, as revealed by Magnuson and Lasure (2004). In view of Gupta et al. (2005) the microbes which come in contact with the general household and hospital environment should be selected to conform the efficacy of any developed antimicrobial product. The standard stains of *Staphylococcus aureus* (ATCC 22925)

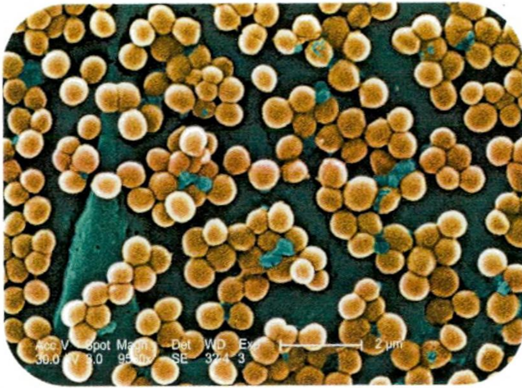
and *Escherichia coli* (ATCC 25922) were used as representative gram positive and gram negative. Similarly *Aspergillus niger* (ATCC 6275) and *Candida albicans* (ATCC 10239) of yeast cultures respectively fungi for testing antimicrobial activity can be used (Kivcak et al., 2002 and Rajendran et al., 2004). According to Wasif and Rubal (2007) to impart antimicrobial finishing on cotton woven fabric using aloe vera extract of various concentrations test against a gram positive bacteria *Staphylococcus aureus* and gram negative namely *Pseudomonas, E. coli, Klebsiella* and a fungus called *Candida albicans* is a must. Hence, the bacteria and fungi of gram positive bacteria-*Staphylococcus aureus*, gram negative bacteria-*Escherichia coli*, *Candida albicans* and *Aspergillus niger* fungi of microorganisms were selected for the study (Plate – 3). The selected microorganisms cultures were procured at microbiology laboratory, PSG Institute of Medical Science and Research Hospitals, Coimbatore, India.

3.1.10 Selection of Antibiotics

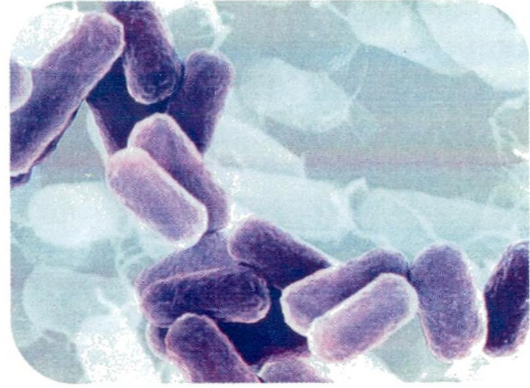
Antibiotics provide the main basis of the therapy for bacterial infections. However, genetic high variability of bacteria enables them to rapidly evade the action of antibiotics by developing antibiotic resistance. The discovery of penicillin in 1929 its use in chemotherapy in 1941 had a response to the great fatalities in the Second World War. So, penicillin and chloramphenicol were selected for the study. The antibiotic penicillin and chloramphenicol was obtained from Himedia Laboratory, Mumbai. The purity of the antibiotic is 99.8 per cent, as revealed by Venkatesan and Karrunakaran (2010). The penicillin img for positive bacteria, chloramphenicol img for negative bacteria were selected and used as standard antibiotics for comparison with extracts and fractions.

3.1.11 Selection of Herbal Finish Application Method

The method of preparation used by the practitioners can broadly be classified into four categories namely plant parts applied as a paste, juice extracted from the fresh plant materials, powder made from dried plant materials, decoction obtained from fresh plant materials, as expressed by Ramya et al. (2009). Antimicrobial textiles find a variety of applications in health and hygiene products, specially the garments worn close to the skin. The modern developments in science and technology has listed a different method of application for herbal extracts on textiles.

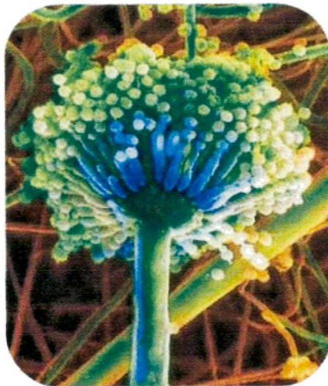


Staphylococcus aureus

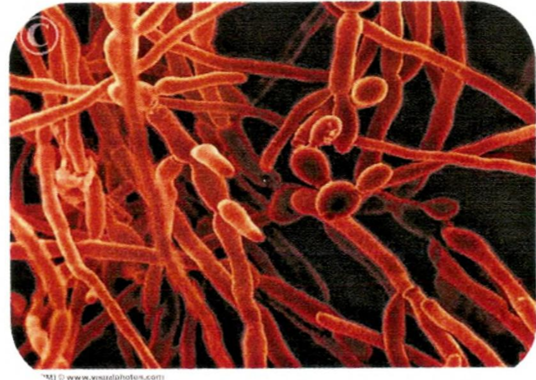


Escherichia coli

BACTERIA



Aspergillus niger



Candida albicans

FUNGI

PLATE – 3

SELECTED MICROORGANISMS

Gopalakrishnan (2007) reveals pad dry, micro encapsulation finish method as the recent trend in antimicrobial finishing. Ramachandran et al. (2004) added the pad dry cure, exhaust, spray coating and foaming techniques to this list. In padding, it is important to know the actual pick up of the fabric from the bath. To get the optimum pick up and penetration, the pad dry cure method is recommended for finishing the fabrics, as reported by Korgaonkar (2006). Microencapsulation is a micro packaging technique that has traditionally involved the deposition of thin polymeric coatings on small particles of solids and liquids. Micro encapsulation has more advantages to conventional process in terms of durability, economy, energy saving, eco-friendliness and controlled release of substances, as described by Cheng et al. (2008). Hence, the pad dry cure and microencapsulation finish application methods were selected for the research work.

3.1.12 Selection of Antimicrobial Standard Testing Method

The antimicrobial activity of the extracts as well as finished and unfinished samples were evaluated both qualitatively and quantitatively using the standard test method such as agar plate method (Rajendran et al., 2004). The different methods used for the testing of antibacterial activity namely assessment of antibacterial activity of textile (Qualitative) (AATCC Test Method 147-1998), quantitative (AATCC Test Method 100-1998) and assessment of antifungal activity of textile materials (AATCC Test Method 30-1998), as suggested by Goyal and Prabhu (2009) and Basu and Balasubramaniyan (2008).

The agar diffusion quantitative test is only used to detect the diffusive antimicrobial finish and it is not suitable for non-diffusive finishes. The agar diffusion testing is a method used for testing the antimicrobial effectiveness in the chemical agent by determining and measuring its zone of inhibition, as revealed by Sivaramakrishnan (2007). The zone of inhibition is the area around the treated substrate in which the antimicrobial chemistry leaches or kills or inhibits the growth of microorganism. This killing (or) inhibiting action of a leaching antimicrobial is witnessed when AATCC 147 test is run, as remarked by Gopalakrishnan (2006).

Quantitative analysis of antibacterial property of finished and washed fabrics were estimated as per AATCC-100 Broth dilution test method (Sathianarayanan et al., 2010). AATCC method 100 is adequately sensitive but is

cumbersome and time consuming (AATCC Technical Manual, 2006. Hence the qualitative (AATCC 147, 30) agar diffusion tests for bacteria and fungi, quantitative (AATCC 100) broth dilution test for bacteria were selected for the research work.

PHASE – II : APPLICATION OF HERBAL EXTRACTION ON COTTON FABRICS AND EVALUATE THE ANTIMICROBIAL ACTIVITY AGAINST BACTERIA AND FUNGI

3.2.1 Application of Finish on Fabric

3.2.1.1 Pretreatment

Pretreatments are used for enhancing the appearance, dye uptake and serviceability, as remarked by Gupta (1998). Pretreatments like desizing, scouring, bleaching is normally carried out for all market fabric. Desizing helps to remove the sizing material from the cellulosic materials, so that their absorbance can be improved. The selected material namely cotton was thoroughly desized before finishing. The 60^S count plain cotton fabric was desized with 0.5 per cent H_2SO_4 solution at 40°C for half an hour and then the fabric was rinsed in cold water and dried.

3.2.1.2 Finish Application Methods

The finish was applied onto desized cotton materials by using pad dry cure and micro encapsulation methods.

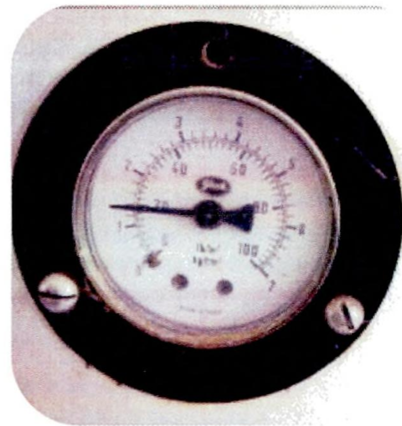
3.2.1.2.1 Pad Dry Cure Method

In the pad dry cure finishing method, the Box Behnken optimized extracted plant solutions of all the herbs were directly applied onto cotton fabric mixed with the mordant (alum) in the ratio of (9 : 1). Alum was added to a solution by simultaneous mordanting method. The liquor was finished onto the fabric at the optimized condition of 37°C for half an hour with a pH of 7 and m:L ratio of 1 : 10. The pressure was maintained at 3 psi. The liquor was stirred at regular intervals to ensure uniformity in finishing. Later the samples were passed through the rollers in a padding mangle (Plate – 4). After padding, the samples were taken and dried at 60°C for ten mins. Then cured at 150°C for 90 seconds for permanent fixation as described by Khanna (2005).

3.2.1.2.2 Micro Encapsulation Method

The Micro encapsulation-co-acervation spray drying method was procedured using all the selected herbal extracts as the core material and acacia gum as a wall

PADDING MANGLE



PRESSURE DIAL

PADDING OF FABRIC

PLATE – 4

PAD DRY CURE METHOD

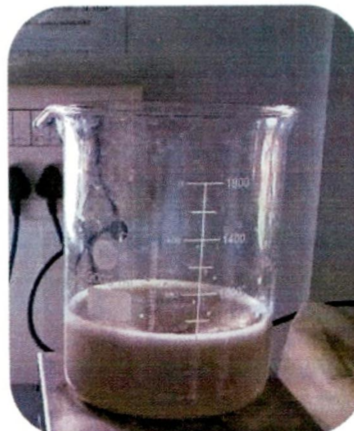
material. Ten gm of wall material was allowed to swell for half an hour in 100 ml of hot water. Later this was mixed with 50 ml of hot water, stirred for 15 min maintaining the temperature at 50°C. After the stipulated time, ten ml of core material was added and stirred at 500 rpm for further 15 min followed by drop wise addition of ten ml of sodium sulphate (20%) solution. This process was completed within ten mins. Later the speed was reduced to 100 rpm. Five ml of formaldehyde (17%) was added and stirred for another two mins. The stirrer was stopped and the mixture was freeze-dried. This was stored for later use.

The laboratory model lu222 spray dryer machine at advance M/s.Lab Ultima Enterprises, Bangalore, India was used to convert the freezed dried herbal plant extracts into powder. The liquid was fed through 0.77 mm diameter nozzle at the air pressure of 3 psi. The rate of liquid fed was maintained at two ml per minute.

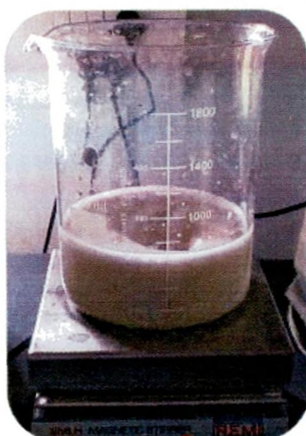
The automation was done in the aspirator with the vacuum of 35 mm water column. Spray drying was performed at an inlet air temperature (T_{inlet}) of 70°C corresponding to an outlet air temperature (T_{outlet}) of 40°C. The powder was collected from I and II cyclones of the spray dryer and stored in a air tight container for further applications (Plate – 5).

These parameters were followed as per the instruction given in the current mode of the machine. Each of the stored herbal extract powder was taken separately and mixed with alum in the material liquor ratio 1 : 10 and poured into the solution container of pneumatic padding mangle. The temperature was maintained at 50°C, the fabric to be finished was passed through the padding mangle to immerse into the solution. This process was repeated for five minutes. Later the fabric was squeezed dried at 70°C and cured for two minutes at 140°C as pointed out by Sathianarayanan et al. (2010) and Thilagavathi and Bala (2007). The core content present in the micro capsules are released only under specific conditions. In control release mechanism the micro capsules of herbal extracts was slowly released onto the fabric surface when any action took place. This leaching type of mechanism was very effective to control the growth of microorganisms on the fabric and to protect the textile materials and also the wearer from the extraneous elements in the environment, as stated by Kan and Yuen (2006). Therefore all the herbal powders were applied on to the fabric by following the above mentioned mechanism. The selected herbal finished samples are shown in Appendix – III.

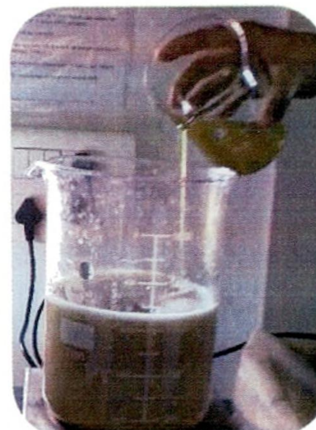
WALL MATERIAL WITH WATER



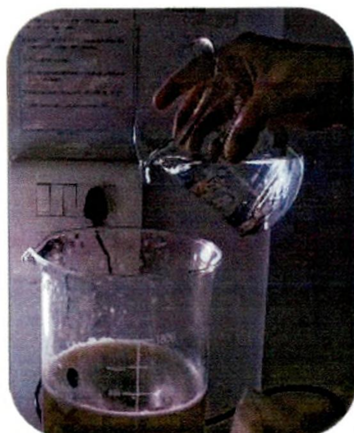
ALLOWED TO SWELL



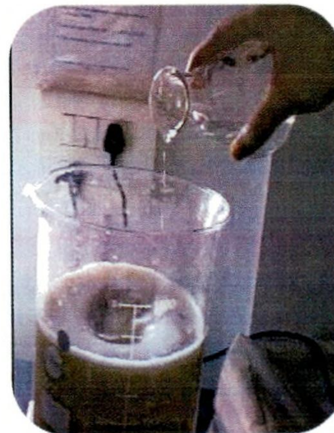
250 ML HOT WATER FOR 15 MINS.



500 ML OF CORE MATERIAL FOR 15 MINS.



20% OF SODIUM SULPHATE FOR 5–10 MINS.



125 ML OF 17% FORMALDEHYDE

PLATE – 5

PREPARATION OF MICRO CAPSULES

3.2.2 Preliminary Phytochemical, Solvent, Herbal Concentrations and Antimicrobial Activity against Antibiotic Screening Tests

Screening of medicinal plants for antimicrobial activities and phytochemicals is important for finding potentially new compounds for therapeutic use. Initial screening of potential antibacterial and antifungal compounds from plants may be performed with pure substances or crude extracts, as reported by Rojas et al. (2006), Freiburghaus et al. (1996) and Silva et al. (1996). The two most commonly used methods to determine antimicrobial susceptibility are the broth dilution test (Hess et al., 1995) and the disc or agar well diffusion test (Navarro et al., 1996).

3.2.2.1 Phytochemical Screening

The plant extracts were subjected to qualitative test for the identification of various biologically active compounds. The medicinal value of these plants lie in some active chemical substances called phytochemicals that produce a definite physiological action on the human body. The most important of these chemically active (Bioactive) constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds, saponins, pholabatannins, catachol, resins, lipids and fats, acidic compounds, terpenoids, reducing sugars, anthraquinone, carbohydrates, steroids and sterols tannin and glycosides found in crude extract and fractions. These phytochemical groups are known to possess antimicrobial compounds, as revealed by Scalbert (1991), Field and Lettinga (1992), Trease and Evans (1989), Ngbege et al. (2008), Meghashri and Gopal (2009), Edeoga et al. (2005) and Hussein et al. (1997).

The preliminary phytochemical screening tests were used to determine the active constituents present in the crude aqueous and ethanolic extracts and has been performed as suggested by Brindha et al. (1982) and Okerulu and Ani (2001). Phytochemical tests were carried out on the methanolic, ethanolic and aqueous extracts for the qualitative determination of phytochemical constituents as stated by Herborne (1973).

Based on the review of literature, two methods namely Thin Layer Chromatography (TLC) for identifying the phytochemical compounds through colour differentiation and agar well diffusion test for identifying the antimicrobial effectiveness in the form of zone of inhibition were selected for the research work.

3.2.2.1.1 Thin Layer Chromatography

The thin layer chromatography is used for assessing the antimicrobial phytochemical compounds present in the herbs. Chromatography is a broad range of physical methods meant to separate the compounds like phenol, tannin and flavonoids and identify the different colour compounds in natural dyes, as reported by Kharbade and Agarwal (1985), Nair (2004), Peng et al. (2006) and Reich and Schibli (2007). This test clearly represents the important quality aspects of fabric and the confidence level to the customer about the protection for which they have paid, as highlighted by White (2009).

In order to perform TLC analysis, 15 gms of silica gel was mixed with 30 ml of water and made into a paste (G_{60}). This paste was then applied onto TLC glass of dimension 20 x 20 cm. The prepared paste was diluted with distilled water at the ratio of 32 gms in 60 ml and applied onto the TLC 0.2 – 0.3 mm thickness plate (Medic-Saric et al., 2004). The plate was dried at 90°C for 30 minutes in a standard oven. A standard line was marked about 1.5 cm above the edge of the plate using standard markers (Rutin and quercetin). Twenty µl of each herbal extract was spotted separately on the marked line. The plates were placed in an air tight chromatography chamber containing ethyl acetate, ethanol, water in ratio of 5 : 1 : 5 known as mobile phase solvent mixture for 40 minutes. Then the solution along with the herbal extracts gradually rise up from the standard line and spread across the interpolate. The flavonoid and phenolic acid compounds appeared in different colour combinations. These plates were then dried. Later ammonia solution was sprayed on the plates which were transferred to UV chamber. The plates were subjected under typical intense fluorescence in UV light and normal light at 365 nm. On exposure, different ranges of colour was noted and analysed for the presence of antimicrobial compounds (Sathishkumar et al., 2008).

3.2.2.1.2 Agar Well Diffusion Test

Agar well diffusion test is a preliminary test to detect the diffusive antimicrobial finish. The important property of a useful antimicrobial is that its presence should be verifiable. Agar well diffusion method is also known as whole plate diffusion method (Branter and Gerin, 1999 and Chitravadivu et al., 2009). It put forths the inhibitory effect of plant extracts against the growth and multiplication of a particular bacterium. The agar well diffusion was done with the selected aqueous and alkaline solvents namely water, ethanol, methanol of different concentrations. The

nutrient agar was dispensed in pre-sterilized petri dishes (25 ml each). The plates were cooled and allowed to solidify. After solidification, wells of 6 mm diameter were punched into the agar with the help of flamed cork borer. Three wells were prepared for each plate. These agar plates were homogeneously inoculated with the *Staphylococcus aureus* test bacteria previously suspended in tryptose broth. The wells were filled with 50 μl and 100 μl of the selected herbal extracts and penicillin antibiotic respectively, as reported by Mishra et al. (2009). The petri dishes were incubated at 37°C for 24 hours. After the incubation period, the diameter of the inhibition zone formed around each well was measured and the values were recorded. The same procedure was focused for the *Escherichia coli* bacteria against with the chloramphenicol antibiotics. Two sets of controls were used, one control was the organism control and the other was standard antibiotic (Perez and Anesini, 1999). This was done to ensure the validity of the test. The same testing procedure was carried out for all the medium of solvents namely water, ethanol and methanol plant extracts. Three readings were recorded for all the herbal extracts. Then the selected herbal extracts microbial zone was compared with the selected antibiotics of penicillin and chloramphenicol against the *Staphyloccocus aureus* and *Escherichia coli* of positive and negative bacterias.

Based on the agar well diffusion method, the antimicrobial effectiveness of selected herbal extracts concentration of 100 μl against with *Staphylococcus aureus* and *E. coli* bacterias were identified and the best herbal concentration was selected for the final study. Similarly, based on the higher zone inhibition the best concentration, suitable solvent for each herb was identified and followed for the final study.

3.2.3 Fourier Transform – Infra Red Spectra (FT-IR)

The application of the infrared spectroscopic study is the diagnostic value in establishing the presence of certain organic constituents in plants, as opined by Gokulakumar and Narayanaswamy (2008) and Velmurugan (2006). More recently FT-IR has been introduced as a metabolic finger printing tool for the plant sciences, as stated by Parveen et al. (2007). FT-IR is the best choice to detect the surface composition of flavonoids structure and shows strong flavonoids functional group (C=O) at 1624.05 cm^{-1}. The compounds responsible for the disease are identified and quantified by the FT-IR analysis, as highlighted by Pellati et al. (2007).

Infra red spectra were recorded on a Nicole 5DX system FT-IR spectrophotometer. The unfinished, pad-dry and microencapsulation of all the selected herbal finished fabric samples were cut into very small pieces (2 mg), was mixed with the 198 mg of potassium bromide and pellets were prepared. Then these samples were transferred to the test tubes,which was inserted in the FT-IR spectrophotometer. The scanning was carried out from 3750 cm^{-1} to 650 cm^{-1} at slow speed, as reported by Aly et al. (2004). The absorption frequencies were indicated in the wave lengths (cm^{-1}).The same was repeated thrice.The FI-TR Graph representing the corresponding curves and wave lengths for different functional groups present were recorded. The same procedure was carried out for all the five herbal finished samples. These wave length frequencies were identified and compared with untreated samples. The presence of functional group was noted for all the samples and identified with the use of standard FT-IR Table (Appendix – IV). This FT-IR scanning was done at the Research Laboratory, PGS College of Arts and Science, Coimbatore.

3.2.4 SEM Study

The SEM study is mainly used to identify the finished molecules present in the fabric samples. The unfinished, pad dry and micro encapsulation finished samples of all the herbs were subjected under the study. The pad dry and micro encapsulation finished samples were analyzed using high resolution Scanning Electron Microscope JEOL M JSM-6360 with suitable accelerating voltage and magnification and used to conform the binding of microcapsules and alignment onto the fabric sample. The microscopic analysis was carried out with 3KV – 6 KV of voltage and below five vacuum levels as described by Wasif and Laga (2009). The pad dry and microcapsules were examined under the X450, X500, X1000, X3000, X7500, X10000 magnifications using light microscopy with image processing technique to analyze the morphology of capsules. The scanning was done for thirty minutes for each sample with four different magnifications depending on the thickness of the sample and images were pictured and presented in the Chapter – 4 Results and Discussion.

3.2.5 Sample Code of Samples

The research was carried out at two different stages namely preliminary assessment and standard test methods, based on which the sample code of samples were classified into two categories. The Table – 10 deals with the preliminary

assessment and Table – 11 deals with the herbal pad dry and micro encapsulation herbal finished samples.

TABLE – 10

SAMPLE CODE OF HERBAL PRE-TEST SAMPLES

S.No.	Medicinal herbs	Solvents			Microorganisms	
		Water	Methanol	Ethanol	*Staphylococcus aureus* positive bacteria	*Escherichia coli* negative bacteria
1.	*Aloe vera*	ALW	ALM	ALE	ALP	ALN
2.	*Marigold*	MAW	MAM	MAE	MAP	MAN
3.	*Kuppaimeni*	KUW	KUM	KUE	KUP	KUN
4.	*Neem*	NEW	NEM	NEE	NEP	NEN
5.	*Yashtimadhu*	YAW	YAM	YAE	YAP	YAN
6.	*Tanner's cassia*	TCW	TCM	TCE	TCP	TCN
7.	*Tridex daisy*	TDW	TDM	TDE	TDP	TDN
8.	*Vettiveru*	VVW	VVM	VVE	VVP	VVN
9.	*Flax seed*	FSW	FSM	FSE	FSP	FSN

TABLE – 11

SAMPLE CODE OF HERBAL FINISHED SAMPLES

S.No.	Sample name	Finish application method	Sample name
1.	*Original*	Unfinished	UF
2.	*Aloe vera*	Pad dry method	ALPF
3.	*Aloe vera*	Micro encapsulation method	ALMF
4.	*Marigold*	Pad dry method	MAPF
5.	*Marigold*	Micro encapsulation method	MAMF
6.	*Kuppaimeni*	Pad dry method	KUPF
7.	*Kuppaimeni*	Micro encapsulation method	KUMF
8.	*Neem*	Pad dry method	NEPF
9.	*Neem*	Micro encapsulation method	NEMF
10.	*Yashtimadhu*	Pad dry method	YAPF
11.	*Yashtimadhu*	Micro encapsulation method	YAMF

3.3 PHASE – III : EVALUATING THE ANTIMICROBIAL EFFECTIVENESS, PHYSICAL AND MECHANICAL PROPERTY, DURABILITY AND CURING PROPERTY OF HERBAL EXTRACTIONS

The pad dry and micro encapsulation finished fabric sample evaluation includes the antimicrobial effectiveness by qualitative and quantitative tests, visual inspection, physical, mechanical, comfort, absorbency, durability and functional properties.

3.3.1 Antimicrobial Assessment Tests

The antimicrobial activity of all the herbal pad dry and micro encapsulation finished samples were analysed by two different test methods namely antibacterial and antifungal assessments. For the assessments the inoculums were prepared with pure cultures of both bacterial and fungal types and the following microorganisms were used for the investigation of antimicrobial analysis namely *Staphylococcus aureus, Escherichia coli, Aspergillus niger* and *Candida albicans*.

The lyophilized pure bacterial cultures were thawed and inoculated onto nutrient broth and incubated overnight. Then, the cultures were swabbed onto sterile nutrient agar in petri plates. The growth and colony morphology of the cultures was carefully observed. Later, all the microorganisms were subcultured in conical flasks containing sterile nutrient broth. The cultures were incubated in bacteriological shaker-incubators. The nutrient broth cultures were allowed to grow for 24 hours. Then it was subcultured and preserved for further analysis. Similarly, potato dextrose agar was used for the analysis of antifungal activities. The fungal inoculum was prepared by the above mentioned protocol using potato dextrose for broth preparation as described by Mahesh and Satish (2008).

3.3.1.1 Antibacterial Activity

The antibacterial activity of herbal finished samples were analysed by qualitative and quantitative tests. The qualitative and quantitative tests includes agar diffusion test (AATCC 147-1998) method and broth dilution test (AATCC TM 100 – 1998).

3.3.1.1.1 Qualitative Antibacterial Activity Assessment by Agar Diffusion Method (AATCC 147 – 1998)

The textile fabrics treated with the diffusible agents are qualitatively tested using an agar diffusion test, as revealed by Bunce (2004). The agar diffusion test is particularly suitable for diffusive antimicrobial finishes and not for non diffusive finishes, as reported by Joshi and Joshi (2002). AATCC^{TM} 147 is a qualitative zone of inhibition test and is being used in the medical field for decades, as highlighted by Swoffold (2010). This is a quick, cheap, simple and well defined method. An unfinished control fabric was included in the test.

The clear zone of inhibition of bacteria was demonstrated around the herbal finished fabrics when a specimen of such fabric is placed in the surface of agar along

with the test organism and incubated at 37°C for 24 hours. The clear zone (mm) of inhibition around the finished sample was estimated taking the average of three separate measurements.

The antibacterial activity was evaluated by agar diffusion method. The zone of inhibition diameter shows the results of bacterial growth. For the bacterial evaluation, the assay was prepared by the standard principles. When a well is punched in the gel and filled with herbal extract on agar previously inoculated with the test bacterium, it picks up moisture and the antimicrobial compound diffuses radially outward through the agar, producing an antimicrobial compound concentration gradient. The antimicrobial compound present at high concentrations near the well affects even minimally susceptible microorganisms (resistant organisms will grow up in the well). As the distance from the well increases, the antimicrobial compound concentration drops and only more susceptible pathogens are harmed. A clear zone or ring is present around an antimicrobial compound present in the well after incubation if the agent inhibits bacterial growth. The wider the zone surrounding a well indicated the more susceptibility of the pathogen. Zone width also is a function of the antimicrobial compound's initial concentration, its solubility and its diffusion rate through agar. Thus zone width cannot be used to compare directly the effectiveness of two different antimicrobial compounds.

After the assay was prepared, the agar diffusion method was carried out. Three grams of nutrient agar was dissolved in 100 ml of distilled water and autoclaved for 15 minutes at 121°C. Later, 25 ml of sterile molten nutrient agar was poured into sterile petri plate and allowed to solidify. Then 0.1 ml of the bacterial cultures of *Staphylococcus aureus* and *Escherichia coli* (containing 1 x 10^6 CFU /ml) were inoculated on the agar plates and the culture was uniformly spread using a sterile glass rod (Spread plate technique). The wells were bored with eight mm borer in the agar. The selected five herbal extract pad dry and micro encapsulation finished fabric samples were placed on each well in the petri plates. The plates were incubated at 37°C for 24 hours. Then the zone of inhibition was measured and recorded for each sample. The inhibition diameters were measured in millimetres and averaged (Scholz et al., 2005). The minimum inhibitory concentration (MIC) is defined as the lowest concentration (µg) of the extract in agar plates showing no visible bacterial inhibition as reported by Ignacimuthu *et al.* (2006) and Nayak et al. (2008). The same procedure was followed for all the samples, three readings were

noted and the mean values were tabulated. Unfinished cotton fabric does not show any absorption peak in this area, as pointed out by Chen and Chang (2007).

3.3.1.1.2 Quantitative Antibacterial Activity Assessment by Broth Dilution Method (AATCC TM 100 – 1998)

The percentage of bacterial reduction was evaluated by broth dilution method in all the selected herbal finished fabrics. The bacterial reduction was compared with the original unfinished samples. The reduction values were tested against *Staphylococcus aureus* and *Escherichia coli* bacterias.

The broth dilution method was carried out by following the principles. The antimicrobial activity of a substrate-bound antimicrobial agent is dependent upon direct contact of microbes with the active chemical agent. This test determines the antimicrobial activity of finished specimen by shaking samples of surface bound materials in a concentrated bacterial suspension for a one hour contact time or other contact times as specified by the investigator.The suspension was serially diluted both before and after contact and cultured. The number of viable organisms in the suspension was determined and the percent reduction was calculated based on initial counts or on retrievals from appropriate unfinished controls. This method was intended for those surfaces having a percent reduction activity of 50 per cent to 100 per cent for the specified contact time.

This test method is designed to evaluate the resistance of non-leaching antimicrobial finished specimens to the growth of microbes under dynamic contact conditions. This dynamic broth dilution test was developed for routine quality control and screening tests in order to overcome difficulties in using classical antimicrobial test methods to evaluate substrate bound antimicrobials.

The 2.5 x 2.5 mm of all the selected herbal pad dry and micro encapsulation finished sample were added into 50 ml nutrient broth inoculated in the sterilized flasks with the micro organisms of both positive and negative of 1.8×10^5 CFU/ml. Then the flasks were incubated under shaking for eight hours at 37°C. Immediately after the inoculation a sampling was performed from each flask to evaluate the microbial count at zero contact time (T0). Afterward the inoculated flasks were incubated for 24 h at 37°C under shaking, then microbial count was performed at 24 hours contact time (T24). The efficiency of the antimicrobial finished sample was determined by comparing the reduction in bacterial concentration of the finished

sample with that of the control samples expressed as a percentage reduction in standard time. The bacterial counts were reported as the number of bacteria per sample and not as the number of bacteria per ml of neutralising solution, '0' counts at 10 dilution was reported as less than 100. The percentage reduction of bacteria by the herbal finished samples was calculated against *Staphylococcus aureus* and *Escherichia coli* bacterias. The microbial inhibition was determined by the reduction in number of colony forming units with respect to unfinished control sample using the following formula.

$$\text{Reduction rate in the number of colonies (R\%)} = \frac{B-A}{B} \times 100$$

where

R = Percentage reduction

A = Number of colonies before shaking (CFU / ml at zero contact time of the reference sample)

B = Number of colonies after one hour shaking (CFU / ml of finished sample after specified contact time).

The same procedure was followed for all the samples. The percentage bacterial reduction values of all the samples were noted. Three reading were noted for each sample and the mean value was calculated as reported by Gupta and Laha (2007), Wallace (2001), Samanta et al. (2007) and Shanmugasundram (2008).

3.3.1.2 Anti Fungal Activity

The fungal activity was analysed by agar diffusion test method. The zone of inhibition was calculated against *Candida albicans* and *Aspergillus niger* fungi.

3.3.1.2.1 Qualitative Antifungal Activity Assessment by Agar Diffusion Method (AATCC 30 – 1998)

The AATCC30 method was used for assessing the antifungal activity. The zone of inhibition of microbial growth was measured in millimeter against *Candida albicans* and *Aspergillus niger* fungi. The same principle was followed as like AATCC 147 test method. The test method was carried out after the assay was prepared.

Potato dextrose agar (3.9 g) was dissolved in 100 ml of distilled water and mixed well. The mixture was boiled for five minutes with frequent agitation for proper dissolution. Later this solution was autoclaved for 15 minutes at 121°C. Then 0.1 ml of the fungal cultures (containing 1 x 10^8 CFU/ml) of *Candida albicans* and

Aspergillus niger were inoculated on the agar plates and the culture was uniformly spread using a sterile glass rod (Spread plate technique). The wells were bored with 8 mm borer in the agar. The selected five herbal finished fabrics were cut and placed in each well in the petri plates. About 200 μl of the herbal extract was added in each well. The plates were incubated at 37°C for 72 hours. After incubation the zone of inhibition was measured. Then the same procedure was followed for all the samples. Three readings were taken and the mean value was calculated.

3.3.2 Subjective Evaluation

Subjective evaluation of the finish includes sensory evaluation of the intensity of fragrance on the fabric. The pad dry and micro encapsulation herbal finished samples of selected herbs were subjected to the visual inspection to assess the general appearance, colour, texture, odour and luster.

3.3.2.1 Visual Inspection

A panel of 25 post graduate students specialized in the field of Textiles and Clothing and Apparel Technology were selected as judges for evaluating the fabric samples. The pad dry and micro encapsulation finished and unfinished samples were cut into 2 x 2 cms and pasted on a neat white chart and labeled. This was given to the selected judges along with the prepared rating scale (Appendix – V) for visual inspection.

3.3.3 Objective Evaluation

The objective of textile testing lies with the fact that the results of testing in research helps the scientists to decide which route to follow next. Besides, the findings on different properties of textiles, it gives the customer a better line to select what type of fabric is appropriate for their end use, because they are the consuming groups for whom the products are particularly intended,as indicated by Paul and Devi (1997).

Objective evaluation includes laboratory tests like physical, mechanical, comfort, absorbency and functional property tests. The objective evaluation tests were carried out to study the various properties of the fabric samples before and after pad dry and micro encapsulation herbal antimicrobial finishing treatments by laboratory examination using suitable instruments.

3.3.3.1 Tests to Evaluate Physical Properties

Fabric weight, fabric count and fabric thickness of the pad dry and micro encapsulated finished samples were tested to find out physical changes due to finish on the fabric.

3.3.3.1.a. Fabric Weight (ASTM D 3776)

The weight of the fabric can be expressed in two ways, either as "mass per unit area" (or) "weight per unit length", (Textile Committee (1993) and Grover and Hambay (1969)). The simplest unit is ounce per square yard. The weight of the fabric can be expressed as weight per unit area in terms of ounces per square yard or grams per square meter, weight per unit length in terms of ounces per yard or grams per meter, as remarked by Booth (1996). According to ASTM D 3776 standard procedure, the Quadrant Balance was used to determine the weight of the sample. It has quadrant copper scale graduated in ounce per square yard. Ten samples were cut at random from different places and each of the samples was suspended from the hook of balance and the reading was noted in grams per square meter (GSM) or ounces per square yard. The readings were noted for each sample to find out the fabric weight before and after finishing of herbal antimicrobial treatment on cotton fabric.

3.3.3.1.b. Fabric Count (ASTM D 3775)

According to Lyle (1977), fabric count is the number of ends (warp) or picks (weft) per inch in woven fabric. A fabric may therefore be described in terms of "ends and picks", as stated by Booth (1996). Pick glass was used to find out the number of warp and weft in the fabric samples. The glass was placed randomly at different places and ten such number of repeat was found by analyzing the weave. Ten readings were taken for warp and weft direction in different position and the counting number was recorded for all the samples to find out the shrinkage or elongation that had occurred during the antimicrobial pad dry and micro encapsulation finishing techniques.

3.3.3.1.c. Fabric Thickness (ASTM D 1777)

The ASTM D 1777 standard equipment used for the study was Sherley "Batty thickness tester", a hand operated instrument which has a dial that reads the thickness of the samples. Two clamps were attached to the dial. Each of the sample was placed in between the two clamps and pressed. Then the distance between the

plates is precisely measured. Ten readings were taken randomly and the mean was calculated to find out the fabric thickness before and after application of antimicrobial finish treatment on the fabric of selected herbal extracts. Care was taken to see that the places chosen were away from the selvedge and free from wrinkles.

3.3.3.2 Tests to Evaluate Mechanical Properties

The mechanical property tests include tensile strength, elongation and abrasion resistance. The pad dry and micro encapsulation herbal antimicrobial finished fabric samples were tested to find out the effectiveness of finish on the fabric.

3.3.3.2.a. Tensile Strength and Elongation (ASTM D 5034)

Strength is the ability of the fabric to withstand stress and strain applied at the time of processing of the fabric and during the usage of the fabric. Breaking strength is a measure of resistance of the fabric to a tensile load in either warp or weft direction, as stated by Angappan and Gopalakrishnan (1987). Breaking strength is the force required to break a fabric when it is under tension. Breaking elongation is increased in length that has occurred when the fabric breaks, as viewed by Pizzute (1985). The effective breaking strength and elongation of warp and weft of all the herbal finished samples were determined by the grab method using Instron universal tensile tester, as reported by Patel et al. (2004) and Aly et al. (2004).

According to ASTM D 5034 the instron universal tensile tester used the extension principle. A strip of 100 x 150 mm size of sample was cut from each of the herbal finished samples in both warp and weft direction. The grab test method was carried out for each sample. The gauge length was fixed as 75 mm. The options of all the parameters like type of fabric, machine speed, load force were fed onto the computer. Place the sample in the top and bottom jaws, ensure that the jaws are firm and tight after fixing the sample. The complete setting was finalized then the fabric sample was tested. To test the fabric specimen press the test key on control unit (or) green colour push button on the instrument. When the test was under process the display showed the signal of "in progress", after completion of test, the results of breaking strength and elongation were showed in the control unit automatically.The graphs were plotted immediately and it can be viewed. After the breaking force results are displayed automatically the bottom jaw returns back to its original position.

The same procedure was used for testing all the samples. Ten readings were taken for all the herbal pad dry and micro encapsulation finished samples.

3.3.3.2.b. Abrasion Resistance (ASTM D 4966)

Abrasion resistance is the ability of a material to resist the action of abrasive forces. Abrasion is one of the major criterias to assess the durability of the fabric, as stated by Basu (2001). Abrasion is just one aspect of wear and is the rub away of the component fibers and yarns of the fabric (Kotb and Megeid, 2011). Abrasion resistance is a test used to stimulate and measure the wear performance of textiles yarns, fabrics or floor covering (Textile Institute, 1991). According AATCC (2006), abrasion is the wearing a way of any part of the material by rubbing against any other surface.

The Eureka Martindale Abrasion Tester was used to determine the fabric abrasion resistance of the samples. The severity of abrasion varies with the nature of the abradant. Ten samples were cut at random from each of the herbal pad and micro encapsulation antimicrobial finished fabrics using a template dimension. The initial weight of the sample was taken accurately to the nearest of 0.001 gm using an electronic balance. Then the sample was mounted on the mushroom shaped sample holder. Sample holders with 200 grams weight was used for this purpose and silicon carborundum zero was used as abradant for cotton fabrics. The rubs were standardized to 50 revolutions. The samples were made to rub against the abrasive surface. After 50 revolutions the samples were removed and the final weight of each sample was found out, further weight loss due to abrasion. The appearance of a hole or rupture that occur on the sample determined the end point. The same procedure was repeated for all other samples and mean values were calculated. Each time a fresh abradant was used. Similarly the mean value of the ten readings for each of the samples was calculated. The loss in weight of each material was recorded separately. The formula given below was used.

$$\text{Weight loss} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100$$

$$\text{Abrasion resistance} = 100 - \text{Weight loss (\%)}$$

3.3.3.3 Tests to Evaluate Comfort Properties

The comfort property tests includes fabric stiffness, fabric drape and air

permeability. The selected pad dry and micro encapsulation herbal finished samples were taken for analyzing the comfort properties.

3.3.3.3. a. Fabric Stiffness (ASTM D 1388)

Stiffness is the resistance to bending (Textile Committee, 1993 and ASTM, 1999). Fabric stiffness indicates the resistance of the fabric to bending and it is a key factor to assist the handle and drape of any fabric, remark Angappan and Gopalakrishnan (1987). Fabric stiffness is a terminology that which expresses the character and quality of the fabric manifested by its performance in respect of fitting to human body, the feel of surface and comfort in wearing, as suggested by Harlapur et al. (2010). Fabric stiffness is explained as a combined effect of elastic property and mass per unit area (AATCC, 1995).

Eureka Shirley stiffness tester was used (cantilever principle) to determine the stiffness using in bending length using a scale of six inch (15 cm) length and one inch (2.5 cm) width. Ten samples were cut in the warp and weft directions. Each sample was mounted on the platform along with the scale which was moved slowly, until the fabric touched the edge of the platform and the tip of the fabric coincide with the index line which was viewed in the mirror. The bending length was read from the scale mark opposite to the zero line on the side of the platform. Ten readings were taken and the mean was calculated.

3.3.3.3.b. Fabric Drape (ASTM D 4850)

Drape is one of the subjective performance characteristic of a fabric that contributes to aesthetic appeal. Fabric drape is the extent to which a fabric will deform when it is allowed to hang under its own weight (ISI Hand Book, 1982). Booth (1996) describes drape as the ability of the fabric to assume graceful appearance in use. The characteristics of warp and weft yarns affect the graceful folding of cloth when it is draped other any circular support. Drapability is described as a phenomenon of fabric fold formation which arises when a fabric hangs down without the influences of external forces.

To determine drapability, Eureka Drape Meter was used. A circular fabric specimen was sandwiched between two horizontal discs of smaller diameter and the unsupported annular ring of fabric was allowed to hang down under the action of gravity. A planar projection of the controller of the drape specimen was recorded on a light sensitive paper. The drape pattern obtained was drawn along the outline and its

area is determined gravimetrically. The mass per unit area of the paper was determined by cutting a known area of the original paper and weighing the same. The mass per unit area of drape was determined by cutting the outline and weighing the same using an electronic balance. Drape coefficient was calculated for all the herbal finished sample, using the formula.

$$\text{Drape co-efficient} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100$$

3.3.3.3.c. Air Permeability (ASTM D 7377)

Air permeability of any fabric is important because ultimately it affects the comfort of the users. A comfortable garment textile should breathe and bench mark air permeability characteristics, as remarked by Schindler et al. (2001). Any fabric contains some porous portion which improves its breathable characteristics. So, the air porosity is an important functional property from the user's point of view. Air permeability increases for the fabrics made from finer yarns as expected, as described by Majumdar et al. (2010). Air permeability of a fabric is the volume of air measured in cubic centimeter passed per second through the square centimeter of the fabric. The digital FX 3300 air permeability tester was used to find out the air permeability of the unfinished and selected herbal antimicrobial finished samples. The sample was placed in between the sensing circular discs of 100 mm diameter without any wrinkles. After fixing the samples, the disc were fixed firmly to prevent release of air. Later the pressure was adjusted and set to zero. Then the machine was started and the values were displayed on the dial. The ten readings were noted. The same procedure was followed for all the herbal finished sample. The mean value was recorded.

3.3.3.4 Tests to Evaluate Absorbency Properties

Absorbency is one of the several factors that influence textile processing such as fabric preparation, dyeing and application of finishes. Often interchanged with the term wettability, the absorbency characteristics of a fabric can influence the uniformity and completeness of bleaching and dyeing. The absorbency of yarn or textile fabric can be determined by the test method (AATCC Technical Manual, 2008). Water absorbency mainly depends upon the porosity of the fabric, the type of fibre and yarn. The absorbency properties of pad and micro encapsulation herbal antimicrobial finished samples were determined by drop test and wickability test.

3.3.3.4.a. Drop Test (AATCC 79)

The ability of a fibre to take up moisture is termed as absorbency. The wettability is the time taken in seconds for a drop of water to sink into the fabric. If any fabric takes more than 200 seconds to absorb the water then it is considered as unwettable. In this test, the time taken for the absorption of water drop completely into the fabric was used to analyse the hydrophobic nature of fabric. As per the AATCC Test method 79 (2007), the conditioned fabric was tightly spread over an embroidery hoop circularly without wrinkles and kept 9.5 mm below a burette, which is positioned to allow 15 – 25 drops of water. A burette filled with distilled water was clamped on a stand. The distance between the sample and burette nozzle was kept constant. The nozzle of the burette was opened just to allow a drop of water to fall on the fabric sample. The stop watch was started simultaneously and it was stopped when the drop of water fully sank into the material. The time required for the drop of water to lose its specular reflectance and appear as a dull wet spot was noted. The fabric was illuminated at an angle of 45°C and then viewed from the opposite direction so that any water on the surface reflects the light to the viewer. The same procedure was repeated for ten times for the unfinished, pad dry and micro encapsulated herbal finished samples. The mean value was calculated and recorded.

3.3.3.4.b. Wickability Test (AATCC 79)

Wicking is the transportation of liquid water by capillary action. The ability of the fabric for wicking depends on the surface properties of the constituent fibres type, denier, yarn structure, fabric structure and their total surface area. Wicking behaviour of fabrics is extremely important from the point of view of comfort characteristics of fabrics, as remarked by Phukon and Phukon (1998). Transverse wicking is the transmission of water perpendicular to the fabric thickness. Transverse wicking is more important as the mechanism for removal of liquid, perspiration from the skin. Transverse wicking is measured by plate test method in which the rate of water uptake by the fabric is measured by timing the movement of the meniscus along the long horizontal capillary tube. In longitudinal wicking test, the edge of a strip of fabric is immersed in distilled water and the rate of rise of the leading edge of the water is measured, as described by Nayak et al. (2008). According to Behera et al. (2002) faster the rate of wicking, better will be the sweat transporting ability and the fabric will be more comfortable.

The ability of the fabric to absorb water especially by a wicking or capillary action may be observed by timing the rate at which water climbs up a narrow strip of fabric suspended vertically with its lower end dipping into the water. Wicking test was conducted as per the procedure given by Saville (1999). Ten samples were cut into size of 30 cm length and 2.5 cm width from all the herbal antimicrobial finished samples. One end of the sample strip was pasted to a glass rod which was placed on heavy wooden blocks and at the other end two grams weight was added to keep the sample straight. Two cm of the sample was allowed to immerse in a tray of distilled water. The rise of the water level in the strip was measured on the scale after 30 seconds. Keeping the length of the fabric (cm) as constant, the same procedure was repeated for all the samples and the mean value was calculated. The capillary rise of each material was recorded carefully to find the absorbency of the fabric before and after finishing. High wicking value shows greater water transport. The resultant capillary force drives the liquid into the capillary spaces, as reviewed by Yuvarani et al. (2010) and Manohari et al. (2011). Wickability of the fabrics was also measured from the wicking height, by using the formula given below.

Wickability = Average height in rise x mass percentage.

3.3.3.5 Tests to Evaluate Fabric Durability Properties

A key issue to be considered in any finishing treatment is the durability to washing. Temporary finishes are good only for disposable articles (Gupta and Laha, 2007). The antibacterial activity of the finished samples were evaluated after (5, 10 and 15) wash cycles as per AATCC 124-1992 method.

3.3.3.5.a. Finish Wash Durability Test (AATCC 124)

Wash durability of pad dry and micro encapsulation herbal finished fabrics were evaluated for analyzing the antibacterial property after being subjected to a couple of repeated wash cycles as reported by Sathianarayanan and Bhat (2010).

All the selected herbal extract finished samples were laundered according to AATCC test method 124 – 1992 for antimicrobial testing. The durability of the antimicrobial finish was evaluated by repeated home laundering method. The standard detergent (5% owf) was used for each wash cycle and the samples were immersed in the solution for about 20 minutes as stated by Orhan et al. (2007) and (40°C) as suggested by Sathianarayanan et al. (2010). After 20 minutes, the fabric were rinsed with hot water (40°C). Later, the sample was squeezed and dried in

shade. After each wash 2.5 x 2.5 cm sample was cut and subjected to antibacterial test against both the *Staphylococcus aureus* and *Escherichia coli* bacterias. This process was repeated upto 30 washes.

The same procedure was followed for all the herbal finished samples. The values of zone of inhibition (mm) was noted for each and every laundered fabric. Then the bacterial reduction percentage was calculated against both the positive and negative bacteria.

3.4 PHASE – IV : PRODUCT DEVELOPMENT AND THEIR PERFORMANCE

The success of any research depends upon its application in day to day life. Considering the functional properties of each of the herbal solutions the investigator planned various end products.

3.4.1 Selection of Products

The textile products used for medical applications are called as medicated health care products. They are used for first aid, clinical or hygienic purposes and rehabilitation. The protective health care and hygiene textile products include both disposable and non-disposable products mainly used in hospitals such as surgical gowns, surgical masks, gloves, surgical drapes, surgical foot wear and head wear, staff apparel, towels, bedding, diapers, sanitary napkins, tampons, bed sheets, eye pads, pillows, panty shields, wipes and incontinence products. The use of such products helps to reduce the opportunity for contamination by biological toxins and infectious pathogens to other patients, as reviewed by Massey and Geol (2008) and Gulrajani (2008). Depending upon the functional properties of the selected five herbal solutions, the investigator designed five medicated herbal textile products. The products are hand gloves, head and face mask, fabric sandles, eye pillow and bed spread for aloe vera, neem, marigold, yashtimadhu and kuppaimeni herbal finished fabrics respectively.

3.4.2 Construction of Selected Health Care Products

3.4.2.1 Gloves

A glove is a type of garment which covers the hand. Gloves can serve to protect and comfort the hands of the wearer against cold or heat, physical damage by friction, abrasion or chemicals (http://medical-dictionary.thefreedictionary.com). They vary in size and length. In the modern days elbow length gloves are used by most of the ladies on two wheelers to protect them from UV rays.

The trunk is the rectangular silhouette that constitutes the front and back portions of the glove. The glove involves four components for construction. The narrow oblong piece is to provide room for fingers. The small triangular sections of quirks are inserted for extra freedom movement for the fingers. The thumb is bulky and provides freedom of movement for the wearer.

Measurements Required

Full length (160 cm), wrist circumference (25 cm), elbow circumference (30 cm), palm width (12.5 cm), bicep circumference (32.5 cm).

Drafting Procedure

Draw a rectangle ABCD,AB=bicep circumference + 5 cm

AC = full length + 7.5 cm, AB=CD, AC=B D

AE = BF = 20 cm, EG = FH = 7.5 cm, CI = DJ = 5 cm. Draw line EF, GH, IJ.

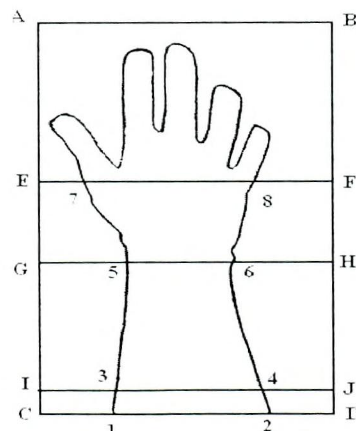
Mark 1 and 2 – 2.5 cm inwards on line CD.

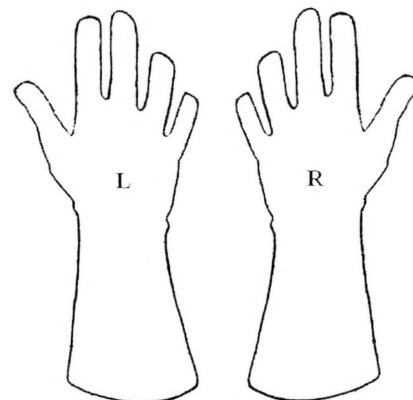
Mark 3 and 4 – 3.5 cm inwards on line IJ.

Mark 5 and 6 – 7.5 cm inwards on line GH.

Mark 7 and 8 – 5 cm inwards on line EF.

Trace out the outline of the palm and hand as shown in figure.





Construction Procedure

The left and right drafted patterns were placed on the aloe vera herbal finished fabric and two pieces for right and left arms were traced. One centimeter allowance was given for all the sides except the hemline. The cut fabrics were placed right side facing each other and stitched from one edge of the hem line through the trunk across each finger till the other side of the hem line. The hem line was folded

and machined placing an elastic at the edge. The gloves are placed on forms and pressed, as reported by Diamond and Diamond (2007) (Plate – 6a).

3.4.2.2 Fabric Sandles

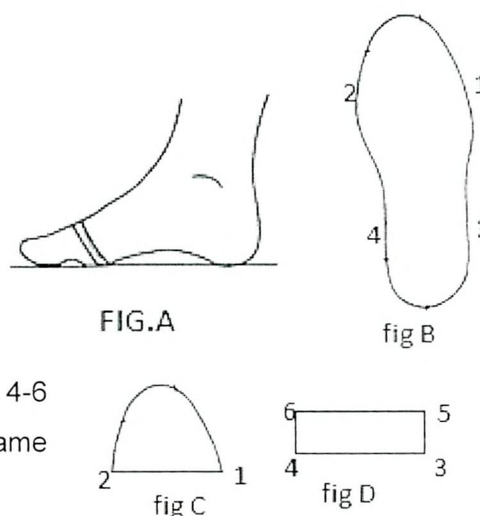
Sandles are intended to protect and give comfort to the human foot while doing various activities. Fabric sandles are worn during winter. The main components of a sandle are bottom piece, cup piece and strap piece.

Measurements Required

Foot length measurement (22.5 cm), foot width measurement (25 cm) and toe measurements (12.5 cm).

Drafting Procedure

To trace the base of the footwear, the posture in the Figure A can be used. Trace the base as in Figure B. Mark 1 and 2 from the base for the front piece as shown in the Figure C and mark 3 and 4 from the base for the back piece as shown in the Figure D. Mark 4-6 as 5 cms (height of the back piece). Mark 5-3 same as 4-6 and 5-6 same as 3-4.



Construction Procedure

Place the sandle patterns on the herbal finished fabric and cut three pieces of the bottom, cup and strap patterns. Place three layers of bottom patterns one on top of the other and sew all the sides. Stitches are made throughout the layer to give a quilting effect. The three layers of the cup and strap patterns were stitched together placing one on top of the other. Then the cup part was attached at the top portion in the finger area. Later the strap piece was attached at the bottom of the foot at back of the ankle (Plate – 6b).

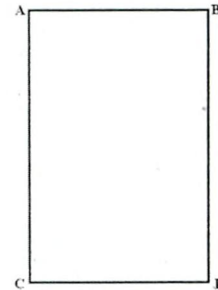
3.4.2.3 Bed Sheet

Bed sheets are fabrics over the bed enhancing its appearance. Bed sheets are available in different widths. They offer softness, tear resistance and comfort feeling. They are very close to traditional textile fabrics, as highlighted by Pal (2009).

Measurement Required

Full length of the bed (187.5 cm), width of the bed (87.5 cm).

Drafting Procedure



Draw a rectangle ABCD

AB = CD = width of the bed + 5 cm

AC = BD = length of the bed sheet required (187.5 cm)

Join 2 or 3 pieces and work a seam .

Construction Procedure

The kuppaimeni finished fabric was cut using the drafted pattern on straight grain. The designed bed sheet consisted of four layers. Hence first two layers were cut to the drafted bed spread size. The next two layers were cut with extra 10 cms.

The first two layers of fabric were placed on the table. The third layer of fabric was placed on top of the first layer fabric. The seam allowances were folded inside. Then fourth layer of fabric was attached at the bottom of these layers and the extra seam allowances were folded inside. Then the plain seam was sewed to all the four corners of fabric (Plate – 6c).

3.4.2.4 Head and Face Mask

The head and face mask is a piece of material around the head covering the ears over the face sides. This is used to protect the wearer from very cool climate conditions and dust.

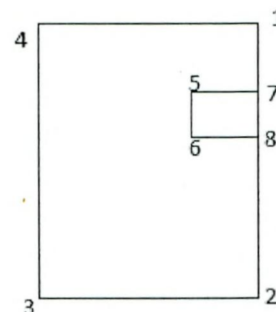
The mask is constructed using cool, comfortable, hypo allergic materials which are naturally fluid resistant to improve wearer safety. Surgical masks, used in medical sector have higher filter capacity and air permeability, as revealed by Pal (2009). These masks prevent the medical worker from infections. The components of head and face mask consists of top piece and side pieces.

Measurements Required

Length (25 cm), head circumference (57.5 cm), neck circumference (37.5 cm), face width allowance (27.5 cm) and eye opening length (17.5 cm).

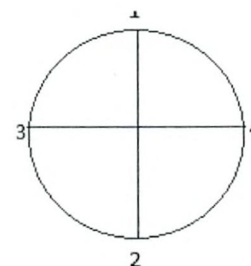
Drafting Procedure

Side Piece



Draw a rectangle 1234, where 1-2 is the height of the mask (top of the head to neck). Mark 3-4 same as 1-2 and mark 1-4 same as 2-3. Mark 1-7 = 7.5 cm, 7-8 = 10 cm, 6-8 = 5 cm. Mark 5-6 same as 7-8 and mark 5-7 same as 6-8. Join 5,6,7,8. 2-3 is ½ of the head circumference.

Top Piece



Mark 1-2 length of the top of the head, mark 3-4 as the width of top of the head. Join 1, 2, 3, 4 as a circle as shown in the figure.

Construction Procedure

The top piece and side pieces of fabric were cut from the neem herbal finish samples. The two side pieces were attached by plain seam. Then the eye opening length was adjusted and it was finished by shaped facing method. Then the top piece and side pieces were joined together. Then the bottom of the mask was finished with elastic attachment (Plate – 6d).

3.4.2.5 Eye Pillow

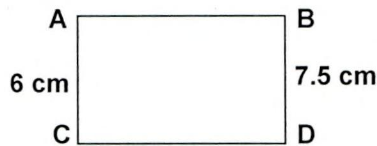
Eye pillow are breathable, have no toxic off gassing and are designed to facilitate optimal alignment breathing and true comfort. The eye pillows quieten the muscles around the eyes and this quietens the brain which induces relaxation. It also reduces eye strain, helps to relieve the symptoms of computer vision syndrome, eliminates headaches caused by stress, anxiety, tension and fatigue. The components of eye pillow is eye shaped square piece and panel piece.

Measurement Required

Eye length (7.5 cm), eye width (5 cm) and head circumference (5.5 cm).

Drafting Procedure

Draw a square ABCD. The fabric of size 6 x 7.5 cm along with seam allowance of 0.5 cm is taken.

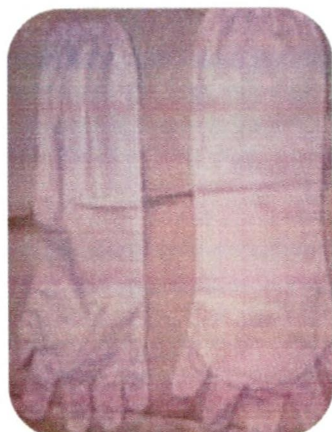


Construction procedure

The drafted patterns were placed on the fabric. Eight pieces were cut. Two layer of fabric pieces were taken and stitched. The pouch was attached along with elastic and centre strip to the outer layer of fabric. The same procedure done for other side of the eye and they joined together. Four layer of fabric pieces are taken and stitched along with elastic and centre strip and turned inside out. The same procedure is done for other side of the eye and they are joined together to form an eye pillow (Plate – 6e).

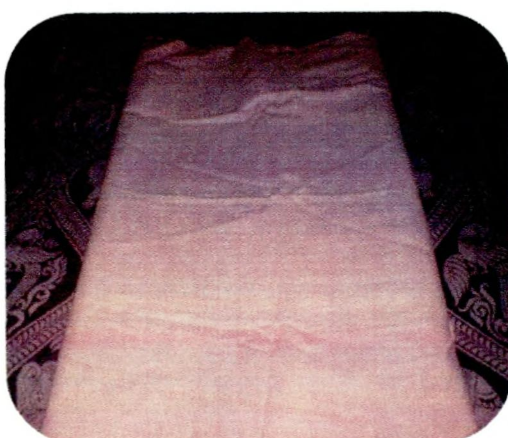
3.4.3 Selection of Samples and Age Groups

Sample for investigation method is one in which some members of population were selected and studied intensively, as explained by Beck (1993). Gupta (2005) defines sample as a part of the universe which is selected for the purpose of investigation.The success of the study depends on the careful selection of the sample. Selecting samples randomly from the population is called as random sample method. The investigator selected different samples for each product. The traffic police, sports person, working women in the age group of 20 – 50 years were selected as samples for aloe vera finished hand gloves to control UV black coating, UV / Sun rays tanning, college students in the age group of 15 – 25 years for neem finished head and face mask for controlling the dandruff hair fall and black pimple spots, homemakers in the age group of 30 – 45 years to control the foot cracks by marigold finished fabrics sandles, software / Information Technology professionals between the age group of 21 – 40 years to control eye irritation and red eye problems by yashtimadhu finished eye pillow and patients in the age group of 25 – 40 years for bed sheets to control bed sores and skin allergies in skin. For aloe vera – UV protection, the traffic police, working women and sports student's samples were selected from Kumaraguru College of Technology and PSG Medical Sciences and Hospital at Coimbatore and Erode town.



6a. ALOE VERA – HAND GLOVES

6b. MARIGOLD – FABRIC SANDLES



6c. KUPPAIMENI – BED SHEET

6d. NEEM – HEAD AND FACE MASK

6c. YASHTIMADHU – EYE PILLOW

PLATE – 6

CONSTRUCTED HEALTH CARE HERBAL MEDICATED PRODUCTS

Neem-dandruff and hair fall, the adolescent and adult girls of Kumaraguru College of Technology, Marigold-foot cracks, homemakers in Coimbatore, yashtimadhu-eye irritation, the software and Information Technology professionals from Vanenburg-KCT Tech Park Company, Sri Gayathri Nature Cure Hospital and Indian Ayurvedic Hospital and Research Limited, Coimbatore, computer science students from Kumaraguru College of Technology, Coimbatore. Kuppaimeni-bed sores, the patients from PSG Institute of Medical Sciences and Research Hospital, Coimbatore were selected for conducting the trial study with the constructed products.

3.4.4 Human Ethical Committee

Considering the safe use of the designed products, they were presented for the analyses and approval to the Human Ethical Committee constituted at Avinashilingam Deemed University for Women, Coimbatore [Proposal Number HEC.2011.20 dated on 02-02-2011] and PSG Institute of Medical Sciences and Research Institutional Human Ethics Committee [Proposal Number 11/098 dated on 05-05-2011] as shown in Appendix – VI. The developed hand gloves were given to 30 women, 20 traffic police and 50 sports persons who spend more than 6 to 7 hours in sunlight. The eye pillows were given to 30 software professionals and 70 computer science students working for more than 5 to 8 hours on the computer. The police were requested to use the gloves for a minimum of four hours (25 days) and the software professionals were requested to wear the eye pillow for minimum of 30 minutes twice a day for 30 days. A questionnaire (Appendices – VII and VIII) was circulated for collecting their opinion about the performance of the product. Then the collected data was consolidated.

TABLE – 12

DETAILS OF WEAR STUDY

S.No.	Medicinal herb	Medicated Products	Diseases/skin problems	Subjects for wear study	Age group of the subjects	Time and Duration for wear
1.	Aloe vera	Hand Gloves	UV black coating, Tanning	Traffic police, sports persons, working women	20-50 years	6 hours / 30 days
2.	Marigold	Fabric Sandles	Foot cracks	Homemakers	30-45 years	6 hours / 30 days
3.	Kuppaimeni	Bed sheets	Bed sores, skin allergies	Patients	25-40 years	1 hour / 30 days
4.	Neem	Head and face mask	Hair fall and black pimple spots	College students	15-25 years	6 hours / 30 days
5.	Yashtimadhu	Eye Pillow	Eye irritation and red eyes.	Software/IT professionals, Computer science students,	21-40 years	1 ½ hours / 20 days

3.4.5 Recommended Ethical Committee Products for Wear Study

The committee carefully analysed the characteristics of the herbs, method of extraction and application of the herbal solutions. They reviewed the relation between the herbal properties, durability, efficacy of the products and disease. Considering these parameters the committee recommended only two products namely aloe vera hand gloves for UV protection and yashtimadhu eye pillow for eye irritation and redness for human wear study (Appendix – IX).

3.4.6 Wear Study (ASTM D 3181)

A test in which textiles are subjected to wear service conditions and evaluated for performance. Any individual uses a control textile during a wear test. The wear study was conducted using the standard ASTM method D 3181 – 95 (ASTM, 2002).

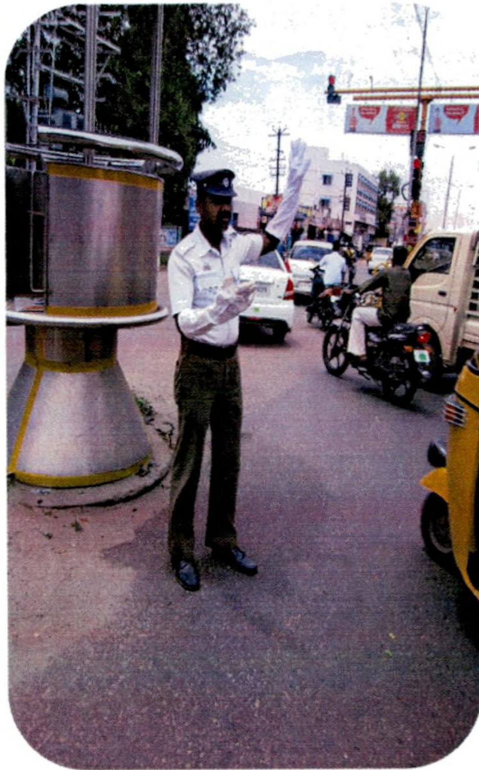
The clinical trial or field trial method is adopted to collect first hand information related to the study. This study is a kind of pilot study and was conducted in order to get an idea about the performance of herbal products, as remarked by Ramachandran et al. (2008). Hence the investigator planned a wear study to evaluate the efficiency of the designed products (Plates – 7, 8, 9) (Appendix – X).

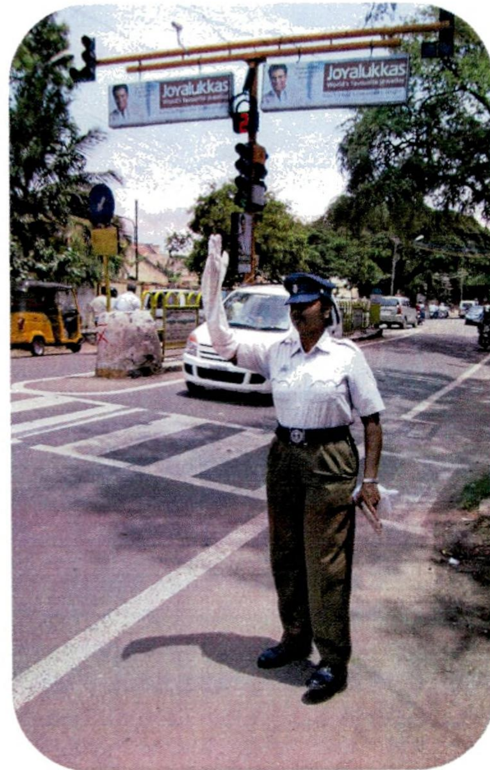
3.4.7 Tests for Functional Properties of Recommended Products

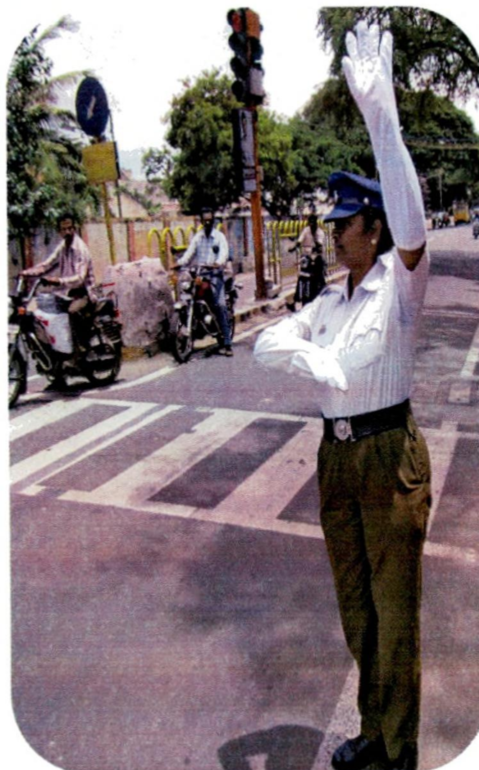
UV radiation of both UVA and UVB fall into the regions of 315 – 400 nm and 280 – 315 nm respectively of the solar spectrum. UV radiation can result in skin damage allergies and even skin cancer,as revealed by Hatch (2003). Cancer is a most harmful and multiple etiological disease. Roughly one person out of five dies of cancer, as narrated by Wasif and Deshpande (2011). Growing concern of health and hygiene has increased the demand for UV protecting textile, as described by Purwar and Joshi (2004). Considering the above facts the aloe vera hand gloves were tested for UV protection.

3.4.7.1 Ultra Violet Protection Test for Aloe Vera Herbal Finished Fabrics

The ultra violet protection factor was assessed in the aloe vera herbal finished fabrics by two different standard methods namely *in vitro* and *in vivo* study.







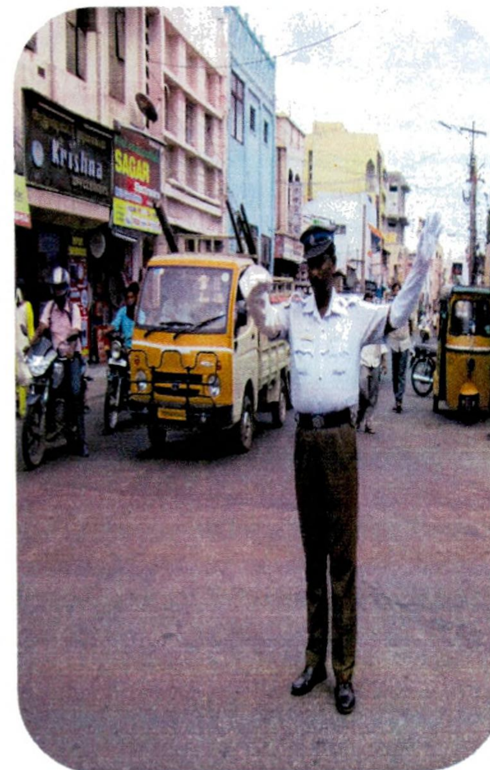
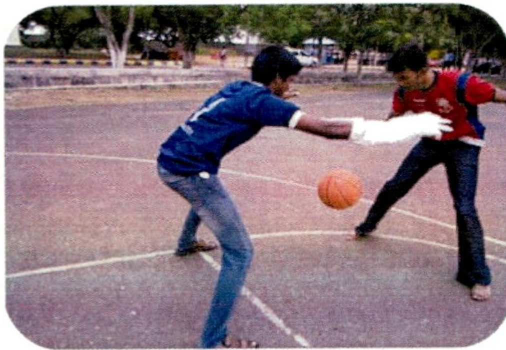
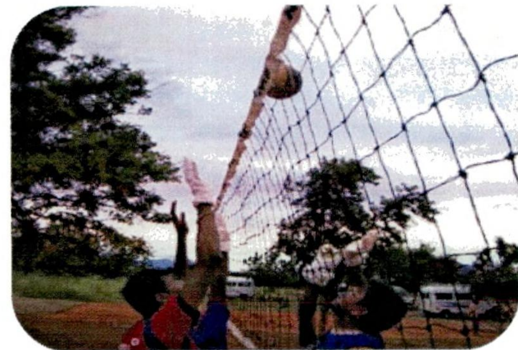
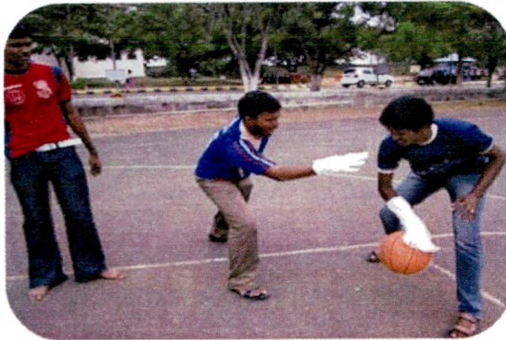


PLATE – 7

WEAR STUDY – TRAFFIC POLICE







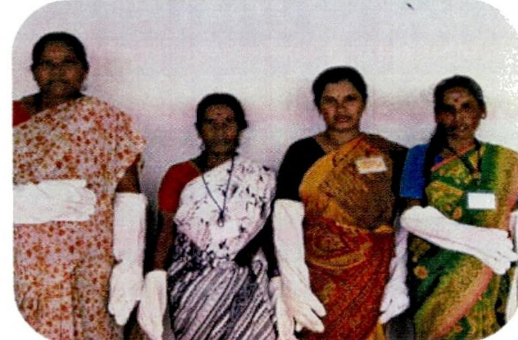
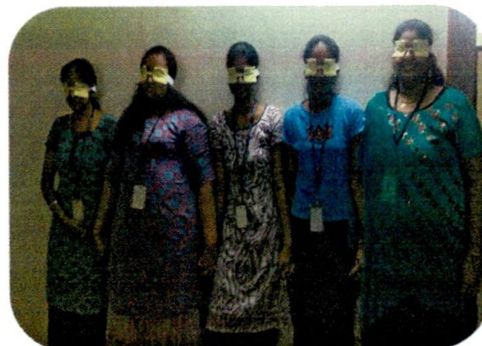


PLATE – 8

WEAR STUDY – SPORTS STUDENTS AND WORKING WOMEN



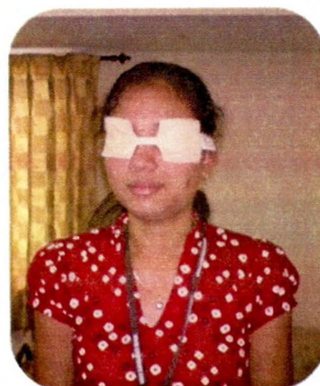


PLATE – 9

WEAR STUDY – SOFTWARE PROFESSIONALS AND COMPUTER SCIENCE STUDENTS

3.4.7.1.1 *In vitro* Study – Tests for Transmittance (or) Blocking of Erythemally Weighted Ultra Violet Radiation through Fabrics (AATCC 183)

The *in vitro* method is also called as instrumental method, because of the use of spectrophotometer. AATCC 183 method defines the UPF rating for a fabric as the ratio of UV measured without the protection of the fabric compared with the protection of the fabric (http://en.wikipedia.org).The transmittance and absorption values of unfinished and aloe vera herbal extract finished fabrics were measured using optical spectrophotometer with an integrating sphere according to AATCC test method 183 (2004). This test was carried out in SGS Laboratory, Pune, India. This standard method was used to determine the ultra violet radiation blocked or transmitted by textile fabrics intended to be used for UV protection (Diffey, 1998). The transmission of ultraviolet radiation (UV-R) was measured on a spectrophotometer having a spectral band pass of five nm over the spectral range of 280 nm to 400 nm as reported by Gorenesk and Sluga (2004) and AATCC (2006). The measured wave length interval over this spectral range should not be greater than five nm. The two specimens of 50 x 50 mm (2.0 x 2.0 in) in diameter were cut from the each sample for wet and dry testing. The specimen was fixed on the spectrophotometer equipped with an integrating sphere. The spectrophotometer was calibrated as per the recommended standards for validating the measurement of spectral transmittance.

After the calibration, the wet evaluation was carried out. First the test specimen was weighed and thoroughly wetted in distilled water by placing it flat at the bottom of a beaker, pouring distilled water into the beaker until the specimen was covered. Allow the specimen to remain submerged for 30 minutes. Stop and turn out the specimen from time to time to ensure good and uniform penetration. Then take out the fabric and check the wet pick up. Then the sample was conditioned to a wet pick up of 150 $\pm$ 5 per cent by squeezing the wet specimen between blotting paper through a hand wringer. Later the sample was placed on the spectrophotometer and the test was conducted. For dry evaluation, the specimen was placed against the sample transmission port opening in the sphere. One UV transmission measurement was taken with the specimen oriented in one direction, a second measurement at 0.79 rad (45°) to the first and a third at 0.79 rad (45°) to the second and the individual measurements were recorded. The same procedure was carried out for unfinished samples. Three transmitted measurements were taken for each specimen. The ultra

violet protection factor (UPF) of each specimen was calculated by using the equation (1)

$$UPF = \frac{\sum_{280\,nm}^{400\,nm} E_\lambda \times S_\lambda \times \Delta\lambda}{\sum_{280\,nm}^{400\,nm} E_\lambda \times S_\lambda \times T_\lambda \times \Delta\lambda} \quad (1)$$

Where,

E_λ = relative erythemal spectral effectiveness

S_λ = solar spectral irradiance

T_λ = average spectral transmittance of the specimen (measured)

$\Delta\lambda$ = measured wavelength interval (nm)

Calculate the average A-range ultraviolet (UV-A) transmittance using equation 2.

$$T(UV\text{-}A)_{AV} = \frac{\sum_{315\,nm}^{400\,nm} T_\lambda \times \Delta\lambda}{\sum \Delta\lambda} \quad (2)$$

Calculate the average B-range ultraviolet (UV-B) transmittance using equation 3.

$$T(UV\text{-}B)_{AV} = \frac{\sum_{280\,nm}^{315\,nm} T_\lambda \times \Delta\lambda}{\sum \Delta\lambda} \quad (3)$$

Calculate the per cent blocking for UV-A and for UV-B using equation ® and equation (5)

$$= 100\% - T(UV\text{-}A) \quad (4)$$

$$= 100\% - T(UV\text{-}B) \quad (5)$$

where,

T(UV-A) or T(UV-B) is expressed as a percentage.

The percentage of UV protection was compared with UPF ratings of ASTM (6544) standard norms. The rating was developed in 1998 by committee RA 106 for AATCC 183 Test methods (http://en.wikipedia.org/wiki/ sun-protective-clothing.html) shown in Appendix – XI.

3.4.7.1.2 *In vivo* Study - Phototherapy for Evaluating the Performance of the Product

The *in vivo* method of UV testing is a MED clinical study of performance on the human skin. The human subjects were selected for the study. With human volunteers, use of sun as the UV source is impracticable to test the UPF of fabrics. Generally xenon arc solar simulators are used with BB-UVB source. The phototherapy panel (V care system) with 8 TL 100 W (12 bulbs) were used to absorb wavelengths below 290 nm and to reduce visible and infra red radiation, as reported by Menzies et al. (1991), Lowe et al. (1995), Gambichler et al. (2000), Greenoak and Palithrope (1996) and Hoffmann et al. (2000). Based on skin phototype, the MED was determined by using incremental UV-B doses on the upper back portion of a subject and was read after 24 hours. Dermatologists classify skin into six types in which 5 and 6 are not affected and 3 and 4 are much affected by sun rays, as stated by Yadav (2011). Thirty five volunteers with Fitzpatric skin type 3 and 4 were selected and study period was from June 2010 to December 2010 at PSG Institute of Medical Sciences and Research Hospital, Coimbatore, Tamil Nadu, India (Appendix – XII). Minimal Erythema Dose (MED) to BB-UV-B was determined by standard method. To measure the MED on protected skin, an aloe vera finished fabric was placed over the skin on the other side of the back, because it is less likely to be influenced by environmental UV exposure. A template with 20 apertures of 1 ½ x 1 ½ cm^2 was made over the back of a cotton suit used by the operation theatre staff. Aperture was made in such a way that two rows of five apertures on each half of the back will closely approximate the skin of the back. Cotton flaps were made over the apertures enabling us to either shut or keep the apertures open by using velcro. The different types of skin with different age group of men and women subjects were randomly selected. The human subjects were seated on a stool at a distance of 20 cm from the source of irradiation for 1 minute. All the ten apertures on one half of the back was irradiated with increasing doses of BB-UVBC. The apertures were closed one after another after delivering one MJ more than the previous aperture. Each irradiated site was marked with genetian violet for identification purpose. Readings were taken after 24 hours of irradiation. The perçeptible erythema visible to the naked eye was taken as MED, as reported by Hoffmann et al. (2000). If MED was not determined at these doses, additional incremental exposures were administered (Plates – 10, 11, 12) and Appendix – XIII. The fabric sun protection factor was determined by using the following equation.

$$\text{SPF} = \frac{\text{Radiation dose to produce just pereptible erythema under fabric covered skin}}{\text{Radiation dose to produce just perceptible erythema of uncovered skin}}$$

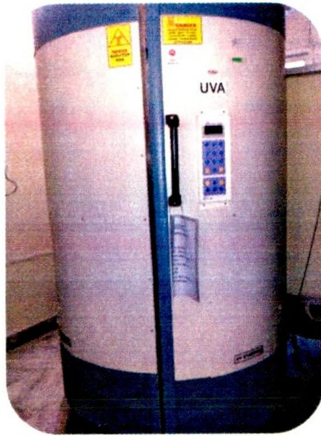
3.4.8 Thermal Comfort Test for Yashtimadhu Herbal Finished Fabrics

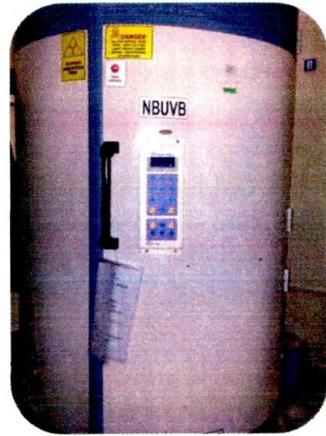
The acceptability of textile fabric largely demands on the comfort aspects which involve the thermal properties. The comfort properties determine the way in which the heat, air and water are transmitted across the fabric. During heavy activities, the body produces a lot of heat, energy and the body temperature rises. When the temperature is raised, discomfort increases in the whole body, especially eyes resulting in irritation and redness. So the garments should allow the perspiration to pass through to ensure comfort. Besides the warm-cool feeling on the first contact of the fabric with the human skin is also a very important parameter that influences the comfort properties of textile fabrics. The thermal properties were measured by two methods namely thermal conductivity and thermal resistance. Hysieope offers one number (index) to characterize the thermo-physiological comfort and another number to determine the sensorial comfort, as revealed by Hes (2008) and Mecheels (2007). The thermal resistance is a reciprocal of thermal transmittance (ASTM, 2007). In view of the above the coolant properties of yashtimadhu finished fabric was tested (Appendix – XIV).

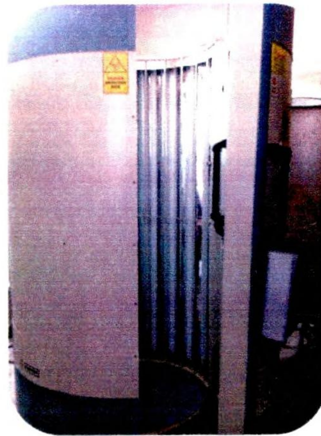
3.4.8.1 Thermal Conductivity Test (ASTM 1518)

Thermal conductivity is an intrinsic property of the material that indicates its ability to conduct heat. It is the flux of heat divided by the temperature gradient, as reported by Varkiyani et al. (2011).Time rate of undirectional heat transfer per unit area, in the steady state between parallel planes separated by unit distance, per unit difference of temperature of the planes (ASTM 1518-98).

The apparatus used was a modification of the standard Lee's disc method for the measurement of thermal conductivity by the absolute plane parallel plate technique. The fabric sample was kept between copper plates and the power to the heater was switched on. The Lee's apparatus consists of a heavy brass disc B suspended from a ring by three equal strings. Over the brass disc is kept a bad







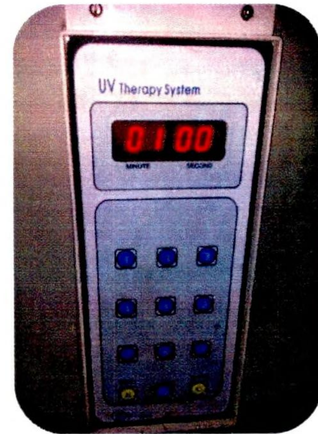
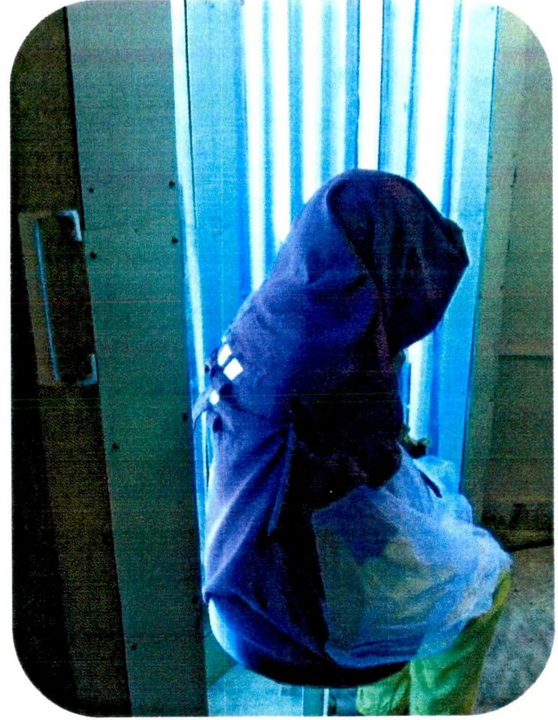


PLATE – 10

UV PROTECTION – IN VIVO STUDY



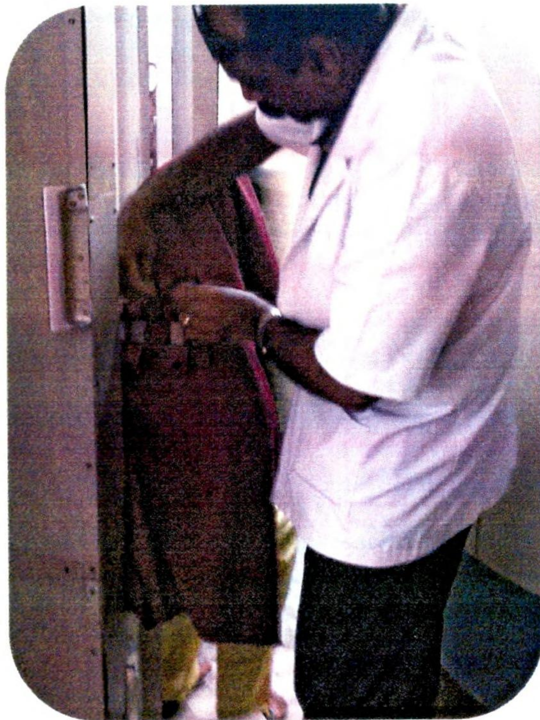


PLATE – 11

UV PROTECTION - IN VIVO HUMAN STUDY

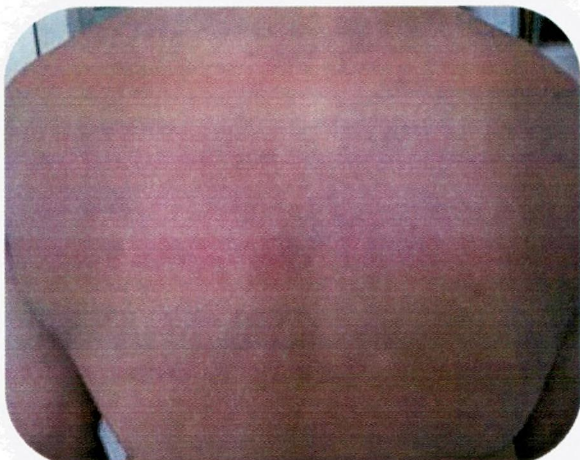
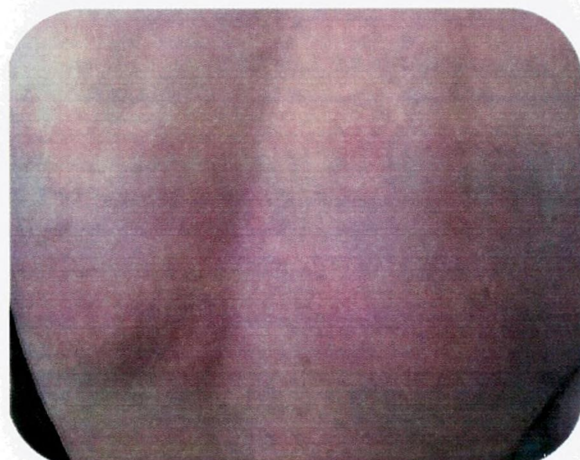
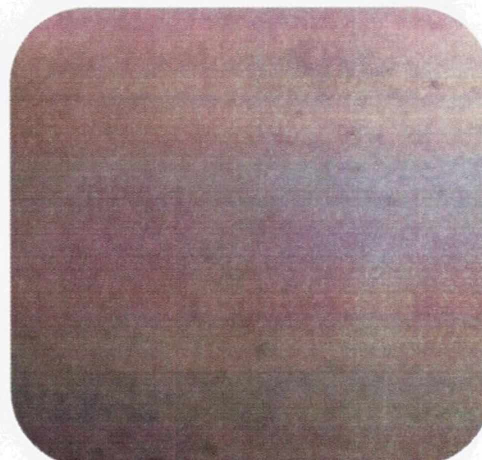
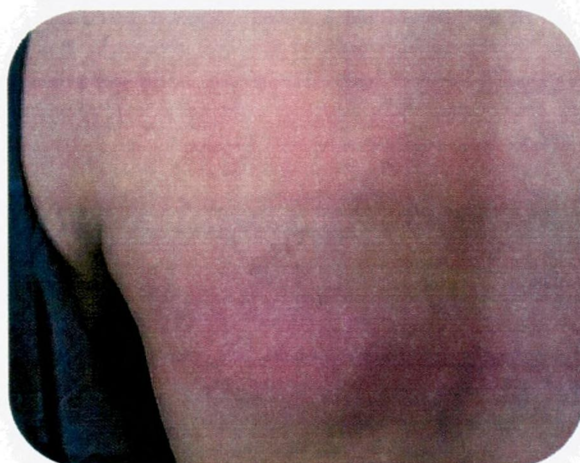
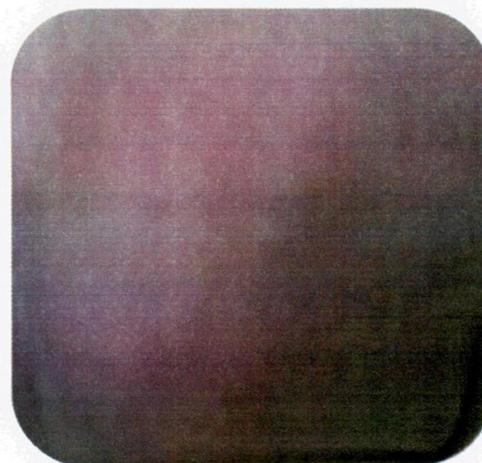
MEN	WOMEN
	
	
	

PLATE – 12

MED TESTING

conductor of the same diameter and over this is placed a cylindrical steam chamber(s) also of the same diameter as the brass disc. The steam chambers and the disc B are provided with two thermometers to note the temperature θ_1 and θ_2 of steam and slab B respectively. The specimen was placed over the plane parallel plate and the steam was passed through the chamber. The heat was applied on the brass disc through bad conductor, the plate gets heated up. The temperature was noted from time to time. Once the temperature is steady it is noted (θ_2°C).

The temperature of the steam was noted (θ_1°C). Later the cardboard was removed and the brass disc was heated with direct contact in the steam chamber until the temperature rises by 5°C above the steady temperature. The disc was separately suspended from the ring after removing from the steam chamber. Temperatures were noted for every 30 secs. The θ_2°C values were tabulated. Then the value of seconds were converted into minutes. A graph was drawn with the temperature on the Y axis and the time on X axis. A horizontal line was drawn corresponding to the steady temperature θ_2°C. It is found by taking two points 1° above 2° and the other degree.

The measurement of thermal conductivity was calculated by using Lee disc method. The coefficient of thermal conductivity of a bad conductor :

$$K = \frac{Msd(r+2l)}{\pi r^2 (2r+2l)(\theta_1 - \theta_2)} \left(\frac{d\theta}{dt}\right)_{\theta=\theta_2} \quad wm^{-1}k^{-1}$$

where,

K = Thermal conductivity of material

M = Mass of the disc placed over the experimental disc x 10^{-3} kg

s = Specific heat of the material of the disc J/Kg/K

d = Thickness of the bad conductor in metres

r = Radius of the brass disc in metres

l = Thickness of the brass disc in metres

θ_1 = Temperature of the steam in degrees

θ_2 = Steady temperature in degrees

$\frac{d\theta}{dt}$ = Rate of heat radiation of the brass disc at θ_2 k/sec.

The yashtimadhu herbal finished fabrics were tested by following the above methods to evaluate the conductivity of the heat.

3.4.8.2 Thermal Resistance Test (ISO 11092)

Thermal resistance (R_{ct}) is defined as the temperature difference between the two faces of material divided by the resultant heat flux per unit area in the direction of the gradient. Thermal resistance depends on the ratio of thickness and thermal conductivity of the fabric, as reported by Majumdar et al. (2010).

The thermal resistance was measured by sweating guarded hot plate test method (ISO 11092). The sweating guard hotplate simulates the heat and mass transfer processes which occur next to human skin. The 30 x 30 mm of specimen with the thickness of less than five mm was cut and used. The specimen to be tested was placed on an electronically heated three mm porous plate with air conditioned ducted to flow across and parallel to its upper surface which is maintained at a standard temperature of 35°C. The conditions of thermal guard temperature control – 35°C, air temperature – 18°C and relative humidity – 30-70 per cent was maintained. After the steady state of temperature condition the fabric is tested. The continued exposure of the fabric over the heated surface helps to absorb the heat. The thermal resistance R_{ct} test range was 0.002 mKW. The air flow speed was controlled and the experiment was repeated for every 15 min. Then mean value was calculated. If the heat loss of the measuring unit is more, it denotes the fabric has cooling effect and vice versa. The thermal resistance was calculated by using the following equation.

$$R_{ct} = (T_m - T_a) A / H - \Delta H_c - R_{ct0}$$

where,

T_m = Temperature of the measuring unit in degree Celsius.
T_a = Air temperature in the test closure in degree Celsius.
A = Area of measuring unit in square meter.
H = Heating power supplied to measuring unit in watts.
ΔH_c = Correction term for heating power for the measurement of R_{ct}.
R_{ct0} = apparatus constant in square meter Kelvin per watt for measurement of thermal resistance

The dry heat flux may consist of one or more conducive, corrective and radiant components. Thermal resistance was expressed in square meters kelvin/watt, is a quantity specific to textile material composite which determines the dry heat flux across a given area and responds to a steadily applied temperature gradient. This test was conducted for the prevention of irritation produced by heat for aloe vera finished fabric and yasthimadhu finished fabrics at SITRA Laboratory, Coimbatore, India.

3.5 PHASE – V : STATISTICAL ANALYSIS

Statistical analysis is done to confirm the validity of the data obtained. The changes in various parameters from base fabric and after finished fabrics can be effectively reviewed based upon statistical tests, as reported by Vyjayanthi et al. (2004). The level of significance difference was calculated and the values are discussed with the original, as remarked by Amrita et al. (2009). Hence the unfinished and herbal finished fabrics were subjected to various tests (ANOVA, paired and unpaired't' test) to evaluate their properties and later analysed using mean values. The significance of these results were put forward and discussed in the Chapter IV – Results and Discussion.