NAME OF THE APPLICANT: DAYANANDASAGAR COLLEGE OF ENGINEERING

ABSTRACT

The present invention relates generally to cross linked hydrogels, specifically to herbal based bio-polymeric cross linked hydrogel composition for bone dressing, sweat absorption and skin care applications. More specifically, the present invention relates to stable bio-polymeric films (BF) with sustained release of herbal extracts from biodegradable polymer Poly Vinyl Alcohol (PVA). The herbal based bio-polymeric cross linked hydrogel composition comprising of: aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 to 12 (g) weight %; and blending agents, said blending agents are herbal extracts that include Turmeric extract - 2 to 4 (g); and *Cissusquadrangularis* extract - 8 to 10 (ml). The blending agents enhance stability of the hydrogel and biocompatibility.

F O R M 2

THE PATENTS ACT, 1970
(39 of 1970)
And
THE PATENTS RULES 2003

COMPLETE SPECIFICATION
(See section 10; rule 13)

"HERBAL BASED BIO-POLYMERIC CROSS LINKED HYDROGEL COMPOSITION"

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THE FOLLOWING SPECIFICATION PARTICULARLY DESCRIBES THE INVENTION AND THE MANNER IN WHICH IT IS TO BE PERFORMED:

HERBAL BASED BIO-POLYMERIC CROSS LINKED HYDROGEL COMPOSITION

FIELD OF INVENTION

The present invention relates generally to cross linked hydrogels, specifically to herbal based bio-polymeric cross linked hydrogel composition for bone dressing, sweat absorption and skin care applications. More specifically, the present invention relates to stable bio-polymeric films (BF) with sustained release of herbal extracts from biodegradable polymer Poly Vinyl Alcohol (PVA). Further, the present invention relates to cross linked hydrogels with optimized concentrations of herbal extracts to enhance the mechanical strength and functional properties of PVA.

BACKGROUND OF INVENTION

Generally, hydrogels were obtained by a cross linking process of polymers, which might be done by a chemical reaction e.g. free-radical polymerization, chemical reaction of complementary groups, using high energy irradiation, or enzymatic reaction or by a physical reaction e.g. ionic interaction, crystallization of the polymeric chain, hydrogen bond between chains, protein interaction, or design of graft copolymers.

Poly vinyl alcohol (PVA) is a water-soluble synthetic polymer. It has the idealized formula $[CH_2CH(OH)]_n$. It is white (colorless) and odorless. The PVA is sometimes supplied as beads or as solutions in water. The PVA is used in papermaking, textiles, and a variety of coatings. The PVA is a unique material as even in its atactic form and lack of stereo regularity, it is semi crystalline in nature. In aqueous solutions, entangled aggregates of hydrogen bonded PVA molecules are formed. Upon freezing the solutions, ice forms in the amorphous region and polymer crystallites grow until they meet the facets of other crystallites which assist in the formation of a porous network upon thawing.

The PVA hydrogels prepared using freeze/ thaw cycles are selected, as they exhibit a high degree of toughness, a rubbery elastic nature, are non-toxic, and can be readily accepted in the body [Bolto *et al., 2009;* Riccardi *et al.,* 2005]. All this makes the PVA preferable over other synthetic polymers from a bio-mimetic perspective, to be applied as matrices for tissue engineering

applications and as a vehicle for the controlled release of drugs [Hongbin *et al.,* 2000; Masanori *et al.,* 2010; Nayak *et al.,* 2005; Cheng *et al.,* 2006].

The PVA is one of the most frequent and the oldest synthetic polymer hydrogels that due to its good biocompatibility has been applied in several advanced biomedical applications e.g. wound dressing [Kenawy *et al.,* 2013], wound management [Zhao *et al.,* 2003], drug delivery systems [Muggli *et al.,* 1998), artificial organs [Yang *et al.,* 2008], and contact lenses [Hyon *et al.,* 1994]. However, the PVA hydrogel possesses insufficient elastic, stiff membrane, and very limited hydrophilicity characteristics which restrict its use alone as a wound dressing polymeric material.

Some of the conventional methods for orthopaedic dressing are:

- Using plaster bandages: Using plaster bandages, which consist of a cotton bandage that has been combined with plaster of Paris, which hardens after it has been made wet. The plaster of Paris is calcinated gypsum (roasted gypsum), ground to a fine powder by milling.
- Using plastic orthopedic fiberglass and polyester casting tape for medical wound dressing bone fracture: Immersing the plastic orthopedic fiberglass and polyester casting tape roll in room temperature water (21-24°C) for 4-6 seconds and squeeze it 2-3 times firmly. If the water warmer than 21-24°C shortens the immersing time, while cooler than 21-24°C water lengthens the time. Enwind spirally; Overlapping the previous layer by one-half or two-thirds of the plastic orthopedic fiberglass and polyester casting tape roll width.
- Using medical orthopaedic fiberglass casting: Orthopaedic splint is composed by manifold layers of orthopaedic casting tapes and specially nonwoven fabric. It is characterized by better viscosity, fast drying time, high rigidness and light weight. Due to better biocompatibility, polyurethane is widely used in medical fields.

Some of the prior arts are:

CN203749451 discloses a medical examination sweat collection device, comprising a roller, to the roller is centered on the central axis, with a plurality of semi-circular cross-section at the outer side of the roller is curved protrusion strip sweat collecting device for medical technology. The protruding strips are embedded in the inner side surface of the annular PVA water-absorbing

sponge sleeve, a U-shaped support is installed on a rotation shaft of the roller, an arc-shaped gasket which is bent towards the inner side is embedded in the top end of the annular PVA water-absorbing sponge sleeve, a medical polyvinyl alcohol sponge is placed on the arc-shaped gasket and a plurality of hollowed-out holes are formed in the arc-shaped gasket.

US7556610 discloses use of a multi-layer wrap for providing more comfortable gel treatment to skin and pressure therapy for underlying skin, the wrap allowing for migration of moisture away from the skin while providing compressive musculo-skeletal support to the treatment site.

CA1056100 discloses a uniformly expandable hydrophilic sponge, adapted for medical usage, characterized by instantaneous wicking and a high liquid holding capacity comprising a reaction product of polyvinyl alcohol and formaldehyde. The wicking and liquid holding capacity is attained by controlling the time, temperature and processing conditions while forming and curing the reaction product in an aqueous medium.

US20090062423 discloses orthopaedic bone cement mixtures that include a primary material that has a flowable state and a solid mass state and a low weight percent amount of polyvinyl alcohol (PVA) material. The PVA material distributed in the primary material PVA solutions (percentage PVA) is formulated to substantially match or accommodate a patient's elastic modulus of the target bone structure. Alternatively, the kits can be formulated and packaged with different low weight percents of PVA solution so as to provide different elastic modulus ranges and labelled with the particular range of elastic modulus for matching to a particular site and/or patient.

US20090171264 discloses a medical device comprising poly (vinyl alcohol), said poly (vinyl alcohol) has a degree of hydrolysis of at least 90% and a weight average molecular weight of at least 50,000 Daltons, and a therapeutic composition. The medical device having 10-50 weight percent content of at least one of water and plasticizer. However, wear particles from such hydrophobic polymers often induce adverse immune responses such as osteolysis. Furthermore, these polymers, while being bio-inert, are not ideally suited for use as a cell scaffold or soft tissue replacement.

US8541484 discloses implants comprising poly vinyl alcohol (PVA), wherein said PVA has a degree of hydrolysis of at least 90% and a weight-average molecular weight of at least 50, 000. Some implants further comprise a therapeutic composition. The degree of hydrolysis is at least 95% or 98% in certain embodiments. Some preferred PVAs are cross-linked. Some embodiments concern orthopaedic implants. The orthopaedic implants of the invention include those having an articulating surface that comprises PVA. Some implants can contain additional materials such as water, a plasticizer such as glycerol, or therapeutic compositions.

EP2590689 discloses a composition comprising a macromer that can be in situ polymerized into a hydrogel wound dressing directly on a wound and one or more antimicrobial agents intended to achieve bacteriostasis and/or be bacteriocidal. The antimicrobial agent is released upon application and trapped in the hydrogel upon its formation and also released over a period of time into the wound.

CA2162372 discloses a water repellent dressing which is made from a soft, smooth, conformable, air-permeable, hydrophobic fabric. The fabric contains polymeric fibers of an extruded blend of a polymer and a fluorochemical oxazolidinone. These extruded fibers may be used to make nonwoven, knitted or woven fabrics.

CN103041601 discloses a papa ring capable of cooling and absorbing sweat. The papa ring is provided with an elastic curling strip and an outer layer wrapped outside the elastic curling strip. The outer layer is formed by a face layer and a back layer arranged on two sides of the elastic curling strip respectively. At least one of the back layer and the face layer is a polyvinyl acetate (PVA) chamois towel.

CA2672497 discloses an antibacterial sheet comprising an antibacterial composition and a sheet-like base material having the antibacterial composition attached thereto, wherein the antibacterial composition comprises a thermoplastic water-soluble polymer which takes a solid form at ambient temperature and can be dissolved in a body fluid and an antibacterial agent.

Limitations and drawbacks of prior art are as follows:

- The skin under the plaster becomes dry and scaly because the discarded outer skin cells are not washed or brushed off.
- The plaster of Paris casts can result in cutaneous complications including macerations, ulcerations, infections, rashes, itching, burns, and allergic contact dermatitis, which may also be due to the presence of formaldehyde within the plaster bandages. In hot weather, staphylococcal infection of the hair follicles and sweat glands can lead to severe and painful dermatitis.
- Unpleasant odour, burning sensation underneath of cast, improper circulation, cannot withstand re-wetting, fingers or toes may turn blue or white, may contract allergy, inadequate hydration.
- Does not absorb excess sweat to the surface of the dressing.
- Exposure to moisture and irritants, changes in skin pH and friction can cause infection and irritation. Extent of damage can range from erythema (redness) and rash to severe, partial thickness skin damage.
- Due to pressure and temperature damage, the peripheral circulation has been compromised, which leads to venous leg ulcers, pressure ulcers and diabetic foot ulcers. In some cases, especially in leg ulcer management, vulnerable peri wound skin can be the most challenging.
- Normal dry skin contains 10 to 1000 micro-organisms per gram of tissue while skin in exposed sweat deposition and increased temperature, bacterial growth may double and causes irritation.
- Adherent on the skin and cannot be easily removed without trauma, requires more material, labour, and repeated visit to hospital, which increases cost.

Accordingly, there exists a need for an herbal based bio-polymeric cross linked hydrogel composition for bone dressing, sweat absorption and skin care applications.

OBJECTS OF INVENTION

One or more of the problems of the conventional prior art may be overcome by various embodiments of the system and method of the present invention.

It is the primary object of the present invention to provide an herbal based bio-polymeric cross linked hydrogel composition for bone dressing, sweat absorption and skin care applications.

It is another object of the present invention to provide stable bio-polymeric films (BF) with sustained release of herbal extracts from biodegradable polymer Poly Vinyl Alcohol (PVA).

It is another object of the present invention, wherein the herbal extracts which acts as blending agent include Turmeric extract and *Cissusquadrangularis* extract.

SUMMARY OF INVENTION

Thus according to the basic aspect of the present invention there is provided an herbal based bio-polymeric cross linked hydrogel composition comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 to 12 (g) weight %; and

Blending agents,

wherein the blending agents are herbal extracts that include Turmeric extract - 2 to 4 (g); and *Cissusquadrangularis* extract - 8 to 10 (ml), and

wherein the blending agents enhance stability of the hydrogel and biocompatibility.

In another aspect of the present invention, the herbal based bio-polymeric cross linked hydrogel composition comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 (g) weight %;

Turmeric extract - 2 (g); and

Cissusquadrangularis extract - 8 (ml).

In yet another aspect of the present invention, the herbal based bio-polymeric cross linked hydrogel composition comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 4 (g) weight %;

Turmeric extract - 2.8 (g); and

Cissusquadrangularis extract - 8 (ml).

In yet another aspect of the present invention, the herbal based bio-polymeric cross linked hydrogel composition comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 8 (g) weight %;

Turmeric extract - 4 (g); and

Cissusquadrangularis extract - 10 (ml).

It is another aspect of the present invention, wherein the aqueous solution of the biodegradable polymer PVA includes glycerin, said aqueous solution of the biodegradable polymer PVA and glycerin are in the ratio of 4:1 respectively.

It is another aspect of the present invention, wherein the turmeric extract and aqueous solution of the biodegradable polymer PVA are in the ratio of 1:4.

It is another aspect of the present invention, wherein the turmeric extract and *Cissusquadrangularis* extract are in the ratio of 1:2.

It is another aspect of the present invention, wherein the turmeric extract is filtered turmeric extract.

BRIEF DESCRIPTION OF THE ACCOMPANYING DRAWINGS AND GRAPHS:

Figure 1: illustrates aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA), turmeric extract and *Cissusquadrangularis* extract mixture placed on the magnetic stirrer.

Figure 2: illustrates herbal based bio-polymeric cross linked hydrogel composition prepared by solution casting method.

Figure 3: illustrates measurement of thickness of the bio-polymeric films (BF) using screw gauge.

Figure 4: illustrates disintegration of the herbal based bio-polymeric cross linked hydrogel composition during swelling analysis in distilled water and sweat composition.

Figure 5: illustrates standard solutions of turmeric extract for spectrophotometric analysis.

Figures 6a and 6b: illustrate surface morphology analysis of BF by scanning electron microscope.

Figure 7: illustrates estimation of protein in turmeric extract.

Figure 8: illustrates estimation of the amount of carbohydrates present in turmeric extract.

Figures 9a to 9f: illustrate skin sensitivity test performed on Wistar albino rats.

Figure 10: illustrates sequential optimization of the herbal based bio-polymeric cross linked hydrogel composition.

Figure 11: illustrates scanning electron microscope results for BF with filtered extract of turmeric.

Figure 12: illustrates scanning electron microscope results for BF with crude extract of turmeric.

Graph 1: Comparison of disintegration rate between membranes prepared by solution casting method and freeze thaw method using different concentrations of aqueous solution of biodegradable polymer PVA.

Graph 2: Estimation of carbohydrates in 5g of turmeric extract.

Graph 3: Estimation of cellulose in 1 g turmeric extract.

Graph 4: Fourier Transform Infrared Spectroscopy (FTIR) analysis.

Graph 5a: Swelling analysis membrane with filtered turmeric extract in distilled water.

Graph 5b: Water uptake capacity of membrane with filtered turmeric extract in distilled water.

Graph 5c: Swelling analysis membrane with filtered turmeric extract in sweat composition.

Graph 5d: Water uptake capacity of membrane with filtered turmeric extract in sweat composition.

Graph 5e: Swelling analysis membrane with crude turmeric extract in distilled water.

Graph 5f: Water uptake capacity of membrane with filtered turmeric extract in distilled water.

Graph 5g: Swelling analysis membrane with crude turmeric extract in sweat composition.

Graph 5h: Water uptake capacity of membrane with crude turmeric extract in sweat composition.

Graph 5i: Comparison of swelling analysis of membranes with filtered and crude turmeric extract in distilled water and sweat composition.

Graph 5j: A plot of force versus displacement for BF with crude turmeric extract.

Graph 5k: A plot of force versus displacement for BF with filtered turmeric extract.

Graph 5l: A plot of force versus displacement for comparison between bio-polymeric films with crude turmeric extract and BF with filtered turmeric extract.

Graph 6: Stress v/s strain curve for BF with crude turmeric extract.

Graph 7: Stress v/s strain curve for BF with filtered turmeric extract.

Graph 8: Spectrophotometric analysis for determination of dilution rate of turmeric from the membrane during swelling studies.

DETAILED DESCRIPTION OF THE INVENTION WITH REFERENCE TO THE ACCOMPANYING FIGURES AND GRAPHS

The present invention is thus directed to an herbal based bio-polymeric cross linked hydrogel composition for bone dressing, sweat absorption and skin care applications. The composition comprises of aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) and one or more blending agents. The blending agents are herbal extracts that include turmeric extract and *Cissusquadrangularis* extract. Basically, PVA is relatively biocompatible, it can swell to take up large water content and can be moulded into desired shapes but it lacks sufficient mechanical stability. Adding chemical cross-linking agents can increase stability of hydrogel composition, but it may affect biocompatibility. The drawback is overcome by addition of blending agents to improve stability and biocompatibility of hydrogel composition. The composition with optimized concentrations of herbal extracts enhances the mechanical strength and functional properties of PVA.

The composition comprises of:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 to 12 (g) weight %;

Turmeric extract - 2 to 4 (g); and

Cissusquadrangularis extract - 8 to 10 (ml).

In one aspect, the composition comprises of:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 (g) weight %;

Turmeric extract - 2 (g); and

Cissusquadrangularis extract - 8 (ml).

In another aspect, the composition comprises of:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 4 (g) weight %;

Turmeric extract - 2.8 (g); and

Cissusquadrangularis extract - 8 (ml).

In yet another aspect, the composition comprises of:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 8 (g) weight %;

Turmeric extract - 4 (g); and

Cissusquadrangularis extract - 10 (ml).

Process for preparing the composition:

The herbal based bio-polymeric cross linked hydrogel composition is prepared using known Solution Casting (SC) method and Freeze Thaw (FT) method.

Process for preparing Turmeric extract:

Different concentrations of raw turmeric 2g (25%), 2.8g (35%) and 4g (50%) were weighed and washed completely using distilled water and crushed using mortar and pestle. The extract was filtered using muslin cloth of 250 microns and used for further processing.

Preparation of *Cissusquadrangularis* extract:

The fleshy stem portion (1.5 kg) of *Cissusquadrangularis* stem was collected, washed, cut into small pieces and air-dried. It was then crushed using a mortar and pestle added to boiling water. Once, the slurry has formed it was filtered using muslin cloth. An extract filtrate of 1 liter was obtained.

Cissusquadrangularis weight in kg	Solvent quantity (lt)	Wt taken in ml
1 kg	1	8 ml
1.5	1	8ml

1 kg *Cissusquadrangularis* in 1 liter distilled water.

Solution Casting (SC) method:

Referring to Figure 1, aqueous solutions of biodegradable polymer PVA 2 to 12 (g) weight percent were prepared. The aqueous solution of biodegradable polymer PVA includes glycerin,

said aqueous solution of biodegradable polymer PVA and glycerin are in the ratio of 4:1 respectively. For complete dissolution, the solution was stirred using a magnetic stirrer. Turmeric extracts of 25% (2 g), 35% (2.8 g), and 50% (4 g) were added to the stirring gel. *Cissusquadrangularis* extract (approximately twice the amount of turmeric extract) of 8 (ml), 8 (ml), and 10 (ml) were then added to the strring gel. The magnetic stirring was done at 60^{o}C for approximately 2 hours. Solution was poured onto Petri plates and allowed to set for approximately 10 minutes and was incubated for approximately 48 hours at 37^{o}C as shown in Figure 2. The hydrogel were removed using forceps and its characterization was done.

Freeze Thaw (FT) method:

Aqueous solutions of biodegradable polymer PVA 2 to 12 (g) weight percent were prepared. The aqueous solution of biodegradable polymer PVA includes glycerin, said aqueous solution of biodegradable polymer PVA and glycerin are in the ratio of 4:1 respectively. For complete dissolution, the solution was stirred using a magnetic stirrer. Turmeric extracts of 25% (2 g), 35% (2.8 g), and 50% (4 g) were added to the stirring gel. *Cissusquadrangularis* extract (approximately twice the amount of turmeric extract) of 8 (ml), 8 (ml), and 10 (ml) were then added to the strring gel. The magnetic stirring was done at room temperature for approximately 2 hours. Solution was poured onto Petri plates allowed to set for approximately 10 minutes. The hydrogel was frozen at -20 ^{0} C for 18 hours and thawed at room temperature for 6 hours for maximum 5 cycles.

Determination of water uptake by herbal based bio-polymeric cross linked hydrogel:

Water content in herbal based bio-polymeric cross linked hydrogel is not only to provide a local moist environment, but also to adjust the permeation of nutrients and gases into cells. Dried herbal based bio-polymeric cross linked hydrogel can swell in water or in a saline up to more than 1000 times their original weight. The amount of absorbed water is usually expressed as water uptake or swelling ratio (SW %) as shown in the following equation.

Water uptake or swelling ratio (SR) % = [{(Ws-We) /We}] *100

Where, *Ws* is the weight of swollen hydrogel at interval times,

We is the weight of dried hydrogel.

Procedure:

The herbal based bio-polymeric cross linked hydrogel were cut in cross section of 3*3 and the thickness of hydrogels was measured using manual micrometer at 3 different places as shown in Figure 3. Dry weights of herbal based bio-polymeric cross linked hydrogel and weight of watch glass were noted. The hydrogels were soaked in 25 ml of water/sweat composition. Weights of swollen gels were noted every 15 minutes, continued till hydrogels completely disintegrates. Swelling ratio was calculated for each herbal based bio-polymeric cross linked hydrogel in water as well as in sweat composition as shown in Figure 4.

Optimization of herbal based bio-polymeric cross linked hydrogels:

Herbal based bio-polymeric cross linked hydrogel hydrogels possess a high degree of swelling in water or biological fluids and an elastic or rubbery nature structure. After swelling studies and spectrometric analysis, compositions best suited for preparing optimized gels were analyzed.

Procedure:

Referring to Figure 5, spectrometry analysis showed that among 25% (2 g), 35% (2.8 g), and 50% (4 g) turmeric extracts, turmeric let out was found to be almost equal to the RDA value in 50% (4 g) extract. *Cissusquadrangularis* extract of 10 ml was added. Using solution casting method the swelling ratio was found to be better for 50% herbal based bio-polymeric cross linked hydrogels with 10 ml *Cissusquadrangularis* extract, which was comparatively stable than other hydrogels.

Estimation of Biomechanical properties:

Fourier Transform Infrared spectroscopy (FTIR) analysis or FTIR spectroscopy:

FTIR analysis is a technique which is used to obtain an infrared spectrum of absorption, emission, photoconductivity or Raman scattering of a solid, liquid or gas. The FTIR analysis or FTIR spectroscopy is an analytical technique used to identify organic, polymeric, and in some cases, inorganic materials. The FTIR analysis method uses infrared light to scan test samples and observe chemical properties. Both qualitative and quantitative information about the test sample can be provided. An FTIR spectrometer simultaneously collects high spectral resolution data over a wide spectral range. This confers a significant advantage over dispersive spectrometer, which measures intensity over a narrow range of wavelengths at a time.

The FTIR identifies chemical bonds in a molecule by producing an infrared absorption spectrum. The spectra produce a profile of the sample, a distinctive molecular fingerprint that can be used to screen and scan samples for many different components. The FTIR is an effective analytical instrument for detecting functional groups and characterizing covalent bonding information.

Procedure:

KBr Pellet procedure for solid samples:

About 1/8" of the solid sample was taken on a micro spatula and about 0.25-0.50 teaspoons of KBr was taken. It was thoroughly mixed in a mortar while grinding with the pestle. If the sample is in large crystals, the sample was grinded separately before adding KBr. Just enough samples were taken to cover the bottom in pellet die. It was placed in press and pressurized at 5000-10000 psi. It was carefully removed from the die and placed in the FTIR sample holder. The pressed disc should be nearly clear if properly made. Repeat the process (regrind and repress), if it is translucent.

FTIR sample analysis:

The first step was to collect a background spectrum to subtract from the test spectrum to ensure the actual sample was all that was analyzed. Next, the sample was analyzed by fully-computerized FTIR system which generates the absorbance spectra showing the unique chemical bonds and the molecular structure of the sample material. This profile was in the form of an absorption spectrum which shows peaks representing components in higher concentration. Absorbance peaks on the spectrum indicated functional groups (e.g. alkanes, ketones, acid). Different types of bonds, and thus different functional groups, absorb infrared radiation of different wavelengths. Although the analysis was performed in absorbance, it can be converted to transmittance, since they are simply the inversions of each other. The analytical spectrum was then compared with a reference library program with catalogued spectra to identify components or to find a "best match" for unknown material using the catalogued spectra of known materials.

Tensile strength analysis:

Tensile testing is a destructive test process that provides information about the tensile strength, yield strength and ductility of a material. The tensile test or tension test involves applying an

ever-increasing load to a test sample up to the point of failure. The process creates a stress/strain curve showing how the material reacts throughout the tensile test. The data generated during tensile testing is used to determine mechanical properties of materials and provides the following quantitative measurements:

Tensile strength, also known as Ultimate Tensile Strength (UTS), is the maximum tensile stress carried by the specimen, defined as the maximum load divided by the original cross-sectional area of the test sample.

Yield strength is the stress at which time permanent (plastic) deformation or yielding is observed to begin.

Ductility measurements are typically elongation, defined as the strain at, or after, the point of fracture, and reduction of area after the fracture of the test sample.

Procedure:

The test sample was securely held by top and bottom grips attached to the tensile or universal testing machine. During the tension test, the grips were moved apart at a constant rate to stretch the specimen. The force on the specimen and its displacement was continuously monitored and plotted on a stress-strain curve until failure. The measurements, tensile strength, yield strength and ductility were calculated after the test specimen has broken. The test sample was put back together to measure the final length, and then this measurement was compared to the pre-test or original length to obtain elongation. The original cross section measurement was also compared to the final cross section to obtain a reduction in area.

Scanning Electron Microscopy (SEM) analysis:

Scanning Electron Microscopy also known as SEM analysis or SEM microscopy is used very effectively in microanalysis and failure analysis of solid materials. Scanning electron microscopy is performed at high magnifications, generates high-resolution images and precisely measures very small features and objects.

Specifications:

SEM Analysis with EDS – qualitative and semi-quantitative results

Magnification – from 5x to 300,000x

Sample Size – up to 200 mm (7.87 in.) in diameter and 80 mm (3.14 in.) in height.

Materials Analyzed – solid inorganic materials including metals and polymers

Procedure:

Referring to Figures 6a and 6b, is a scanning electron microscope illustrating how the desiccated samples were loaded, labeled and coated with a gold film. An electron beam was scanned across a sample's surface and the electrons struck and stimulated the sample. Almost instantaneously, as each element returns to its original energy state, it emitted X-rays of specific energies and at different wavelengths characteristic of the element. Energy dispersive X-ray spectroscopy plotted these results with X-ray wavelength on the X-axis and intensity on the Y-axis, and labeled each corresponding element. Identification of the elements was done by matching the peak values on the X-axis with known wavelengths for each element to reveal the sample's elemental composition.

Biochemical tests:

Carbohydrate estimation:

The need for an accurate, fast and reliable analysis of carbohydrate test is crucial for numerous biological processes. In that sense, Anthrone – sulfuric acid assay is one of the most efficient quantification techniques successfully applied to carbohydrate determination. Estimation of carbohydrate amount in aqueous solution of biodegradable polymer PVA is important to analyze because the stability of herbal based bio-polymeric cross linked hydrogel composition depends on carbohydrate content.

Procedure:

100 mg of the sample was weighed into a boiling tube. Hydrolysis was done by keeping it in a boiling water bath for 3 hours with 5 ml of 2.5N HCl and cooled to room temperature. It was neutralized with solid sodium carbonate until the effervescence ceases. The volume was made up to 100 ml and centrifuged. The supernatant was collected and 0.5 ml and 1 ml aliquots were taken for analysis. The standards were prepared by taking 0, 0.2, 0.4, 0.6, 0.8 and 1 ml of the working standard.'0' serves as blank. The volume was made up to 1 ml in all tubes including the sample tubes by adding distilled water. 4 ml of Anthrone reagent (100 mg was dissolved in 100

ml of ice-cold 95% H_2SO_4) was prepared fresh before use was then added. It was heated for 8 minutes in a boiling water bath. It was cooled rapidly and read the green to dark green colour at 630 nm as shown in Figure 7.

A standard graph was drawn by plotting concentration of the standard on the X-axis versus absorbance on the Y-axis. From the graph, the amount of carbohydrates present in the sample was calculated.

Cellulose estimation:

Principle:

Cellulose undergoes acetolysis with acetic/nitric reagent forming acetylated cellodextrins which gets dissolved and hydrolyzed to form glucose molecules on treatment with 67% H_2SO_4. This glucose molecule is dehydrated to form hydroxyl methyl furfural which forms green coloured product with anthrone and the colour intensity is measured at 630 nm.

Procedure:

3 ml acetic/nitric reagent (150 ml of 80% acetic acid and 15 ml of concentrated nitric acid was mixed) was added to a known amount (0.5g or 1g) of the sample in a tube and mixed in a vortex mixture. The tube was placed in a water bath at 100^{o}C for 30 minutes. It was cooled and then centrifuged for 15-20 minutes. The supernatant was discarded. The residue was washed with distilled water.10 ml of 67% sulfuric acid was added and it was allowed to stand for 1hour. 1 ml of the above solution was diluted to 100 ml. To 1ml of this diluted solution, 10 ml of anthrone reagent was added and mixed well. The tubes were heated in boiling water bath for 10 minutes. It was cooled and the colour was measured at 630 nm. Anthrone reagent (200 mg of anthrone was dissolved in 100 ml of ice-cold 95% sulphuric acid) was prepared fresh and chill for 2 hours before use and distilled water was taken.100 mg cellulose was taken in a test tube and preceded from step No. 6 for standard. Instead of just taking 1ml of the diluted solution a series of volumes (say 0.4 to 2 ml corresponding to 40-200 mg of cellulose) was taken and the colour was developed.

Protein estimation:

Referring to Figure 8, 0.2 ml of BSA working standard in 5 test tubes was taken and the volume was made up to 1ml using distilled water. The test tube with 1 ml distilled water serves as a blank. 4.5 ml of reagent I (48 ml of 2% Na_2CO_3 in 0.1 N NaOH, 1 ml of 1% Na K Tartarate in H_2O, 1 ml of 0.5% CuSO4.5 H_2O in H_2O) was taken and incubated for 10 minutes. After incubation, 0.5 ml of reagent II (1 part Folin-Phenol [2 N]: 1 part water) was added and incubated for 30 minutes. The absorbance at 660 nm was measured and the standard graph was plotted. The amount of protein present in the given sample was estimated from the standard graph.

Sensitivity test:

Skin prick test (SPT) is recommended as the primary method of diagnosis of immunoglobulin E (IgE) mediated allergies in most allergic diseases. It has relative sensitivity, specific rapid results, flexibility, low cost, low risk, good tolerability and clear demarcation to patients. Efficacy of skin prick test, among other tests was proven.

S.N	Sample(µg/ml)	No. of animals
1	Control	1
2	Histamine(500)	2
3	Test sample	
	1000	3
	1200	3
	1500	3
	1700	3
	Total No. of animals	15

The average weight of rats was taken as shown in Figure 9a and the surface of the rats was cleaned with 70% ethanol as shown in Figure 9b. The rats were grouped into positive, negative controls and 4 different concentrations of test liquid as shown in Figure 9c. Positions for skin prick were marked with numbers to identify allergens. A prick was made immediately adjacent to numbers to avoid confusion between allergens. Skin prick tests should be at least 2 cm apart to avoid overlapping reaction and fake reactions. A drop of allergen from a dropper bottle is added on to the skin prior to pricking as shown in Figures 9d and 9e. Time of initial and final pricking time was noted to estimate the time of reading results. Reaction to histamine started at 10 minutes, allergen reaction started after 15 minutes. Wheal and flare using glass sheet and ruler were

measured after 20 minutes as shown in Figures 9f. For further confirmation test organisms were under observation for 8 hours. Results were interpreted, if the wheal was circular diameter was measured, for ovoid shortest and longest perpendicular axis were added and divided by 2. A wheel diameter of 3 mm or greater indicates the presence of IgE to allergen tested.

Results:

Optimization and preparation of topical patches:

Optimization of Aqueous solution of biodegradable polymer PVA:

Concentration of aqueous solution of the biodegradable polymer PVA optimal to obtain stable polymeric blends with glycerin was determined by observing the differences in morphology of the bio-polymeric films (BF) having varying glycerin: PVA ratios as shown in Figure 10. The glycerin was added to increase plasticity and uniformity of the membranes.

It was observed that the 1:4 ratios between glycerin and aqueous solution of the biodegradable polymer PVA gave stable, flexible and smooth bio-polymeric films. Though maximum stability was observed at 12 (g) but the optimization was done at 8 (g) because any further increase in the concentration of PVA gave brittle films and a decrease in the concentration of PVA gave very soft and rapidly soluble bio-polymeric films [herbal based bio-polymeric cross linked hydrogel].

Optimization of procedure for the preparation of bio-polymeric films:

Data from swelling studies carried out on the bio-polymeric films was observed for its duration of stability and the results were tabulated in Table 1.

Table 1: Data obtained from swelling studies carried out at different concentrations of PVA in membranes to detect the disintegration time.

Serial No.	Membranes by FT method		Membranes by SC method	
	Amount of PVA (g)	Disintegration time (mins)	Amount of PVA (g)	Disintegration time (mins)
1	2	15	2	9
2	4	36	4	30
3	8	135	8	90
4	12	560	12	420

It was observed that the bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] prepared by FT method had higher stability and water uptake capacity compared to SC method. Graph 1 illustrates the comparison of disintegration rate between membranes prepared by FT and SC.

Thickness plays a very important role in determining the stability of membranes. Table 2 shows the data obtained from swelling studies carried out for membranes with different thickness to detect the disintegration time. The maximum disintegration was observed for herbal based bio-polymeric cross linked hydrogels of 3 mm thickness.

Table 2: Data obtained from swelling studies carried out for membranes with different thickness to detect the disintegration time.

Serial no.	Thickness of the membrane (mm)	Disintegration time (mins)
1	0.5	12
2	1	30
3	2	110
4	3	325

Estimation of components in turmeric extract:

Estimation of protein:

The quantitative analysis of protein in the turmeric extract was performed and the results were tabulated in Table 3.

Table 3: Data for estimation of proteins in the Turmeric extract.

Amount of protein (mg)	OD at 660 nm
0	0
20	0.26
40	0.52
60	0.72
80	0.96
100	1.04
Sample	0.84

Estimation of carbohydrates:

The quantitative analysis of carbohydrates in the turmeric extract was performed and the results were tabulated in Table 4. Graph 2 illustrates amount of carbohydrate v/s OD at 660 nm using different stock solutions of glucose by anthrone method. The OD value of 5 g of the turmeric extract was extrapolated against standard and found to be 0.48. The amount of carbohydrate present in the turmeric extract sample was found to be 0.64g/1g of turmeric**.**

Table 4: Data for estimation of Carbohydrates in the Turmeric extract.

Amount of carbohydrates (g)	OD at 650 nm
0	0
1	0.12
2	0.3
3	0.49
4	0.62
5	0.83
5g Sample	0.48

Estimation of cellulose:

The quantitative analysis of cellulose in the turmeric extract was performed and the results were tabulated in Table 5. Graph 3 illustrates the amount of cellulose v/s OD at 630 nm using different concentrations of cellulose by Anthrone method. The OD value of 1 g of the turmeric extract was extrapolated against standard and found to be 0.62. The amount of cellulose present in 1 g of the turmeric extract sample was found to be 470 micrograms**.**

Table 5: Data for estimation of Cellulose in the Turmeric extract.

Amount of cellulose (µg)	OD at 630 nm
0	0
100	0.13
200	0.26
300	0.38

400	0.51
500	0.65
600	0.81
1g sample	0.62

Optimization of turmeric:

Concentrations of turmeric optimal to obtain stable bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] was determined by testing the structural stability of bio-polymeric films having varying concentrations of the turmeric extracts under three different concentrations. Data from the stability studies are depicted in Table 6.

Table 6: Data obtained from swelling studies carried out for membranes with different ratios of aqueous solution of biodegradable polymer PVA to Turmeric extract to compare the disintegration time and % swelling.

Serial no.	PVA: Turmeric ratio	Weight of PVA (g)	Weight of turmeric (g)	Disintegration time (mins)	% Swelling
1	4:1	8	2	360	439
2	2:1	8	4	1175	1085
3	4:3	8	6	3000	1324
4	1:1	8	8	5600	1539

It was observed the herbal based bio-polymeric cross linked hydrogels with 50% (4 g) of the turmeric extract were stable. Bio-polymeric films with low concentrations of the turmeric extract were found to be unstable and tend to lose their stability at a faster rate and higher concentrations of turmeric in the gels are very stable, but tend to make the skin dry and cause irritation. Hence, 50% (4 g) of the turmeric extract was concluded to be the optimal cross linker concentration to develop bio-polymeric films.

Referring to Table 6, swelling studies were carried out for membranes with different ratios of aqueous solution of biodegradable polymer PVA to turmeric extract to compare the

disintegration time and % swelling. With increase in the turmeric extract concentration stability of membranes was increasing. Maximum swelling rate found for the aqueous solution of biodegradable polymer PVA sample (1:1) was found to be 1539%. Optimal PVA: turmeric ratio was found to be 2:1 with a disintegration time of 1175 minutes and swelling % of 1085.

The stability was compared in crude and filtered extracts of turmeric bio-polymeric films. The swelling studies and tensile strength studies were conducted, and it was found that the filtered extract bio-polymeric films were more stable, had higher tensile strength, Young's modulus and higher dilution rate when compared to crude turmeric bio-polymeric films. Hence, bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] of filtered turmeric extract were considered desirable.

Optimization of *Cissusquadrangularis* extract*:*

Cissusquadrangularis has been used as a medicinal plant since antiquity. *Cissusquadrangularis* has been used in various ayurvedic classical medicines to heal bones and injured, ligaments and tendons. It is considered as a tonic and analgesic and also provides moisture to the skin. Hence 1:2 ratios of turmeric and *Cissusquadrangularis* extract considered optimal for the skin application.

Physiochemical characterization:

FTIR Spectroscopy:

The spectral data obtained from the comparative FTIR analysis is depicted in Graph 4. The absorption bands observed at 3419cm-1, 2929cm-1, 1620cm-1, 1155 cm-1 and 1026 cm-1 and are due to the stretching vibrations of O-H, C-H, C=O, N-H, AND C-O bonds. The new band appears at 1753cm-1 in the bio polymer film.

Field Emission Scanning Electron Microscopy (FE-SEM):

The morphology of the bio-polymeric film [herbal based bio-polymeric cross linked hydrogel] with filtered turmeric extract depicted micro crystalline structures, the pores present and the cross linking between cellulose and aqueous solution of the biodegradable polymer PVA. When compared to the bio-polymeric films with crude turmeric extract, it was observed that, in the the

bio-polymeric films with crude turmeric extract, there was no cross linking, also clumps of the turmeric particles were observed. Hence the bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] with filtered turmeric extracts were considered to have desired morphological properties.

Swelling studies

Data from swelling studies carried out on bio-polymeric films was obtained as shown in Tables 7, 8, 9, 10, and 11 followed by graphical representation in Graphs 5a to 5l respectively. It was observed that with an increase in turmeric content there was an increase in swelling ability of the biomaterial in the first few hours owing the high absorptive nature of these fibers. However, it was found that with the increasing concentrations of turmeric, the water uptake capacity reduces but, the stability is high. Hence ratio of 1:2 of turmeric extract and aqueous solution of the biodegradable polymer PVA was considered optimal.

Table 7: Data obtained from swelling studies performed on bio-polymeric films with filtered extract of turmeric using sweat composition.

Time (mins)	Volume of water uptake (ml)	Weight of membrane (g)
0	0	7.62
15	1.083	8.703
30	1.476	10.179
45	1.764	11.943
60	2.589	14.532
75	2.94	17.472
90	3.27	20.895
105	3.63	24.186
120	3.423	26.745
135	3.291	28.968
150	2.559	30.924
165	2.223	32.826
180	1.956	34.686
195	1.902	36.492
210	1.86	38.172
225	1.806	39.681
240	1.68	41.091

255	1.509	42.423
270	1.41	43.626
285	1.332	44.589
300	1.203	45.471
600	0	49.044
900	0	49.044
1200	0	49.044
1500	0	49.044
1680	0	49.044
1695	0	49.044
1710	0	49.044
1725	0	49.044
1740	0	49.044

Table 8: Data obtained from swelling studies performed on bio-polymeric films with filtered extract of turmeric using distilled water.

Time (mins)	Volume of water uptake (ml)	Weight of membrane (g)
0	0	7.62
15	2.304	9.324
30	4.545	13.869
45	3.969	17.838
60	3.24	21.078
75	2.61	23.688
90	1.65	25.338
105	1.185	26.523
120	1.158	27.681
135	1.134	28.815
150	1.062	29.877
165	1.011	30.888
180	0.99	31.878
195	0.837	32.715
210	0.822	33.537
225	0.813	34.35
240	0.81	35.16
255	0.807	35.967

270	0.798	36.765
285	0.795	37.56
300	0.792	38.352
600	0.645	52.806
900	0.477	63.942
1200	0.357	72.222
1500	0.237	78.102
1800	0.117	81.582
2100	0.045	83.07
2115	0.042	83.112
2130	0.039	83.151
2145	0.036	83.187
2160	0.024	83.211
2175	0.015	83.226

Table 9: Data obtained from swelling studies performed on bio-polymeric films with crude extract of turmeric using sweat composition.

Time (mins)	Volume of water uptake (ml)	Weight of membrane (g)
0	0	7.02
15	1.083	8.703
30	1.476	10.179
45	1.764	11.943
60	2.589	14.532
75	2.94	17.472
90	3.27	20.895
105	3.63	24.186
120	3.423	26.745
135	3.291	28.968
150	2.559	30.924
165	2.223	32.826
180	1.956	34.686
195	1.902	36.492
210	1.86	38.172
225	1.806	39.681
240	1.68	41.091

255	1.509	42.423
270	1.41	43.626
285	1.332	44.589
300	1.203	45.471
600	0	48.44
900	0	48.44

Table 10: Data obtained from swelling studies performed on bio-polymeric films with crude extract of turmeric using distilled water.

Time (mins)	Volume of water uptake (ml)	Weight of membrane (g)
0	0	7.02
15	1.305	8.925
30	1.767	10.692
45	2.082	12.774
60	2.88	15.654
75	3.39	19.044
90	3.84	22.224
105	3.57	24.831
120	3.18	27.039
135	2.607	29.211
150	2.208	31.305
165	2.172	33.273
180	2.094	35.184
195	1.968	36.99
210	1.911	38.652
225	1.806	40.161
240	1.662	41.628
255	1.509	42.999
270	1.467	44.262
285	1.371	45.312
300	1.263	46.26
600	0.15	54.696
615	0.114	54.738
630	0.078	54.738
645	0.042	54.738
660	0	54.738
900	0	54.738
1200	0	54.738
1500	0	54.738

1800	0	54.738
1815	0	54.738
1830	0	54.738
1845	0	54.738
1860	0	54.738
1875	0	54.738
1890	0	54.738
1905	0	54.738

Table 11: Data obtained for % swelling and time of disintegration.

Serial No.	Sample	Initial wt(g)	Final wt (g)	% Swelling	Disintegration time(mins)
1	BF with crude extract in sweat composition	7.02	48.44	618.35	1905
2	BF with crude extract in distilled water	7.02	54.738	344.31	900
3	BF with filtered extract in sweat composition	7.62	49.044	1085.56	1740
4	BF with filtered extract in distilled water	7.62	83.226	543.26	2175

It was observed that the bio-polymeric films (BF) with filtered turmeric extract had high stability and high water uptake capacity due to its cross linking and porous nature. It was observed that the water uptake capacity band stability of the bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] reduced in the sweat composition as compared to the studies carried out in distilled water.

Tensile strength:

Studies were conducted to evaluate the stress resistance capacity of bio-polymeric films. Data obtained from these studies are tabulated as Table 12, it was observed that the bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] with filtered turmeric extract had

higher values for tensile strength and hence, has higher elasticity when compared with crude turmeric extract films as illustrated in Graphs 5j to 5l.

Table 12: Data obtained from Tensile strength test of different bio-polymeric films.

Serial No.	Sample	Peak Load (N)	Displacement at peak load (mm)
1	BF by Freeze thaw method	31.74	83.69
2	BF by Freeze solution casting	19.93	34.38
3	BF with crude turmeric extract	13.83	26.69
4	BF with crude turmeric extract	54.72	159.05

From the Table 12, it is noted that the peak load for bio-polymeric films prepared by freeze-thaw method was higher than the ones prepared by solution casting method. The values were 31.74N and 19.93N respectively. The maximum peak load for bio-polymeric films with crude turmeric extract was 54.72N and displacement was 159.05mm. Tensile strength was found to increase 4 times for the sample with filtered extract over the sample with crude extract. Stress v/s strain was plotted. At the peak load, the Young's modulus was calculated and found to be 1.03MPa for BF with crude extract and 0.89MPa for BF with filtered extract. Hence, it is concluded that BF with crude extract is more stiff compared to BF with filtered extract.

Table 13: Data obtained from tensile strength test.

Serial No.	Sample	Maximum force, F (N)	Thickness, t (mm)	Width, w (mm)	Area, A A=t*w (mm^2)	Tensile strength, TS TS=F/A (N/mm^2)
1	BF with crude extract	13.38	1	2	2	6.915

2	BF with filtered extract	52.42	1	2	2	26.211

Skin sensitivity test:

In order to accurately determine the sensitivity of the bio-polymeric films it was crucial to observe the reactivity of the bio-polymeric films in animal models.

Wistar albino rats were injected with varying concentrations of the liquid gels; they showed no sensitivity when compared to the positive control with histamine which had wheal and flare on its skin. Hence the bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] were concluded to be biocompatible with the skin and cause no allergic reactions.

Table 14: Data obtained from skin sensitivity testing on animal models.

Serial No.	Group	No. of animals	Injected fluid	Concentration of fluids (µg/ml)	Volume of fluid (µl)	Observation	Inference
1	Negative Control	1	Distilled water	-	10	No change	No sensitivity
2	Positive control	2	Histamine	500	10	15 mm wide wheal with flare	sensitivity detected
3	Test Group 1	3	Test fluid	1000	10	No change	No sensitivity
4	Test Group 2	3	Test fluid	1200	10	No change	No sensitivity
5	Test Group 3	3	Test fluid	1500	10	No change	No sensitivity
6	Test Group 4	3	Test fluid	1700	10	No change	No sensitivity

Absorption Distribution Metabolism Excretion (ADMET) Studies:

Data rendered from ADMET studies showed that the skin permeability values were negative which are ideal.

Spectroscopic analysis:

The percentage release of turmeric from bio-polymeric films [herbal based bio-polymeric cross linked hydrogel] is directly proportional to the water uptake capacity. Higher the water uptake capacity, higher is the dilution rate. The spectrophotometric analysis was carried out at 445 nm, to determine the percentage release of turmeric. The values were noted and tabulated in Table 15.

Table 15: Data obtained by spectrophotometric analysis to obtain the dilution rate of turmeric from BF.

Sample no.	Weight of turmeric (g)	Spectraphotometric reading
1	0	0
2	0.5	0.58
3	1	1.16
4	1.5	1.8
5	2	2.3
6	2.5	2.89
BF with Crude extract	1.44	1.64
BF with filtered extract	2.05	2.37

The standard graph of weight of turmeric v/s spectrophotometric reading was plotted as shown in Graph 8. The samples of crude and filtered sample were extrapolated against standard and were found to have OD of 1.64 and 2.37 respectively**.**

Figure 11 illustrates Surface morphology (SEM) results for bio-polymeric films with filtered extract of turmeric. The bio-polymeric films prepared had clear formation of microcrystalline structures and pores in the blended gel. Figure 12 illustrates SEM results for bio-polymeric films with crude extract of turmeric. The images of the bio-polymeric films with crude extract showed abrupt surface with clumps and no uniformity of the structure.

Applications of the herbal based bio-polymeric cross linked hydrogel composition:

1. Orthopaedic dressing/orthopaedic cast/body cast/plaster cast/ surgical cast: Underneath of cast, layer of herbal based bio-polymeric cross linked hydrogel will enhance durability of cast by absorbing sweat and giving enhanced skin care, which will reduce casting cycle, casting price and time of repeated dressing.
2. Loincloth and skin care patch: Sweat absorbing foundation garments with different application.
3. Sweat controller on forehead: Food manufacturing and hotel management as protective layer for head (hair) and face (sweat).
4. Sunrays protection and absorber in helmet: As protective layer for skin in summer and UV protection. Sweat absorbing layer in helmets.

Advantages of herbal based bio-polymeric cross linked hydrogel composition:

- Herbal based bio-polymeric cross linked hydrogel composition for bone dressing, sweat absorption and skin care applications.
- Maintain a moist environment at the wound/dressing interface between cast.
- Absorb excess sweat to the surface of the dressing.
- *Cissusquadrangularis* has been used for its effects osteoporosis.
- Provide bacterial protection from turmeric.
- Allow gaseous and fluid exchange.
- Be non-adherent on the skin and easily removed without trauma.
- Provide some debridement action (remove dead tissue and/or foreign particles).
- Be non-toxic, non-allergenic and non-sensitizing sterile.
- Increase in bio-availability, osteoblastic activity, antioxidant, anti-inflammatory, anti-bacterial activities in hydro gel product.
- Cost effective.
- Enhanced biodegradability.

It is thus concluded that PVA hydrogels are hydrophilic polymers having a pendant hydroxyl group. As per many reports, PVA hydrogels have been proven to exhibit biocompatibility, mechanical properties, moldability and required porosity. The composition of the present

invention turmeric and *Cissusquadrangularis*, are reported to facilitate in bone fracture healing, antioxidant, antitumor, anti-inflammatory, antibacterial, antifungal, antihelmintic, antihemorrhoidal and analgesic activities. Based on the results of swelling and spectrophotometric analysis, the concentrations were optimized for the preparation of these films. Spectrophotometric analysis was done to test the controlled release of turmeric. FTIR analysis proved that there are no other new compounds formed as a result of solution casting at high temperature. Results showed that, they all have common functional groups and characteristic peaks that were almost overlapping. Mechanical properties of herbal based bio-polymeric cross linked hydrogels were defined by SEM analysis and tensile strength analysis. The composition of the present invention was tested for absorption of distilled water and sweat composition by swelling analysis. Then the sensitivity test (skin prick test) was performed on male Wistar albino rats. Positive control was taken as Histamine, negative control as distilled water and the hydrogels were injected in different concentrations to analyze the skin sensitivity reactions. No sensitivity was observed. So, the herbal based bio-polymeric cross linked hydrogel composition was proven to be safe and efficient.

NAME OF THE APPLICANT: DAYANANDASAGAR COLLEGE OF ENGINEERING

WE CLAIM:

1. An herbal based bio-polymeric cross linked hydrogel composition comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 to 12 (g) weight %; and

Blending agents,

wherein the blending agents are herbal extracts that include Turmeric extract - 2 to 4 (g); and *Cissusquadrangularis* extract - 8 to 10 (ml), and

wherein the blending agents enhance stability of the hydrogel and biocompatibility.

2. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 1 comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 2 (g) weight %;

Turmeric extract - 2 (g); and

Cissusquadrangularis extract - 8 (ml).

3. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 1 comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 4 (g) weight %;

Turmeric extract - 2.8 (g); and

Cissusquadrangularis extract - 8 (ml).

4. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 1 comprising:

Aqueous solution of biodegradable polymer Poly Vinyl Alcohol (PVA) - 8 (g) weight %;

Turmeric extract - 4 (g); and

Cissusquadrangularis extract - 10 (ml).

5. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 1, wherein the aqueous solution of the biodegradable polymer PVA includes glycerin, said aqueous solution of the biodegradable polymer PVA and glycerin are in the ratio of 4:1 respectively.

6. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 1, wherein the turmeric extract and aqueous solution of the biodegradable polymer PVA are in the ratio of 1:4.

7. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 1, wherein the turmeric extract and *Cissusquadrangularis* extract are in the ratio of 1:2.

8. The herbal based bio-polymeric cross linked hydrogel composition as claimed in claim 7, wherein the turmeric extract is filtered turmeric extract.

Dated this 5th day of October 2015

P. ILANANGAI [IN/PA-1365]
AGENT FOR APPLICANT