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EFFICACY OF DUSHIVISHARIAGAD ON IMIQUIMOD-INDUCED PSORIASIS-LIKE DERMATITIS IN BALB/C MICE WITH REFERENCE TO KITIBHA KUSTHA: A STUDY PROTOCOL

Ayurveda

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ABSTRACT

Psoriasis is a long-lasting autoimmune disease characterized by raised areas of abnormal skin. These areas are typically red or purple on some people with darker skin dry, itchy, and scaly. . Prevalence of psoriasis in India is 0.44 to 2.8%. Causative factors of psoriasis are genetic, life style, HIV, microbes, medication. Life style Conditions reported as worsening the disease include chronic infections, stress, and changes in season and climate. Aacharyacharak2 described different dermatological manifestations of dushivisha out of which kitibhakushtha one of them it is one of the kshudrakushtha. Presentation of kitibhakushtha resembles like psoriasis.Produces lesions with symptoms like blakish or brown discolouration,dry,rough,hard in nature with itching.In Ayurveda different kinds of herbal products available for treatment of psoriasis effectively.Dushivishariagad is describe as a effective remedy for treatment of dushivisha by different Aacharya in samhita. All the dravyas(herbal drugs) in DushivishariAgad are pittakaphaghna, Vishaghna(neutralize toxicity), Raktaprasadak(blood purifier).some are kushthghan and kandughan. It mostly acts on skin disorders due to its properties and also other systemic disorders. So the study was taken to prove effective roll of dushivishariagad on psoriasis like dermititis in mice.**Aim-** Study the efficacy of DushivishariAgad on Imiquimod- Induced psoriasis like dermatitis in Balb/c Mice with reference to kitibhakushtha. **Material and Method:** 30 Male Balb a/c mice will be selected and divided in 5 groups of 6 mice in each group. In Group A (Normal Control group) Will be treated by the Application of Vaseline on shaved dorsal skin surface, Group B (positive control Group) will be treated by the application of 5% IMQ cream on shaved back, Group C (Standard control group) will be treated by the Application of IMQ cream on shaved back and oral administration of acitretin, Group D (Test group 1) will be treated by the application IMQ cream on shaved back and oral administration of DushivishariAgad with plain water and Group E (Test group 2) will be treated by the application of IMQ cream on shaved back oral administration of DushivishariAgad with Honey. **Analysis plan (Statistical test):**Statistical analysis will be done by applying suitable tests. (ttest,One way ANOVA test) **Results:** Results will be drawn from the observations of statistical test, biochemical parameters and histopathology reports. **Conclusion:** Conclusion will be drawn on the basis of results of Statistical Analysis and histopathology reports.

KEYWORDS

DushivishariAgad, Imiquimod-Induced Psoriasis-Like Dermatitis, kitibhakushtha

1] INTRODUCTION:

Psoriasis is a long-lasting autoimmune disease characterized by raised areas of abnormal skin. These areas are typically red or purple on some people with darker skin dry, itchy, and scaly. Psoriasis varies in severity from small, localized patches to complete body coverage. Injury to the skin can trigger psoriatic skin changes at that spot. Areas of the body most commonly affected are the back of the forearms, shins, navel area, and scalp. Prevalence of psoriasis in India is 0.44 to 2.8%.

Causative factors of psoriasis are genetic, life style, HIV, microbes, medication. Life style Conditions reported as worsening the disease include chronic infections, stress, and changes in season and climate. Others factors that might worsen the condition include hot water, scratching psoriasis skin lesions skin dryness, excessive alcohol consumption, cigarette smoking, and obesity. Drug-induced psoriasis may occur with beta blockers, lithium, antimalarial medications, non-steroidal anti-inflammatory drugs, calcium channel blockers, captopril, granulocyte colony-stimulating factor, interleukins, interferons, lipid-lowering medications[3].

These factors undergo pathological changes in the body resulting in cumulative toxicity. This cumulative toxicity is similar to *dushivisha* concept describe by *aacharyasushrutaDushi visha*[1] is a part of animate, inanimate and artificial poison which cannot removed out of body completely and slowly accumulates in our body and produce toxic symptoms and different complication.*Aacharyacharak*[2] described different dermatological manifestations of *dushivisha* out of which *kitibhakushtha* one of them it is one of the *kshudrakushtha*. Presentation of *kitibhakushtha* resembles like psoriasis.Produces lesions with symptoms like blakish or brown discolouration, dry, rough, hard in nature with itching[5].

In *Ayurveda* different kinds of herbal products available for treatment of psoriasis effectively.*Dushivishariagad*is describe as a effective remedy for treatment of *dushivisha* by different *Aacharya* in *samhita*. As per *AacharyaVaghbhatDushivishariagad*contains *Pipalli (Piper longum), Dhyamak (cymbopogan martini), Jatamansi (Nardostachysjatamansi), Lodhra (symplocosracemosa), Ela (Elettariacardamomum), Suvarchika (Tribulusterrestris), Musta (Cyperusrotundus), Tagar (Valereniawellchi), Kushtha (Saussurealappa), Yashtimadhu (Glycyrrhizaglabra), Chandan (Santalum album), Gairik (Ferrous Oxide)*[4].

All the *dravyas* (herbal drugs) in *DushivishariAgad* are *pittakaphaghna*, *Vishaghna*(neutralize toxicity), *Raktaprasadak* (blood purifier). some are *kushthghan* and *kandughan*. It mostly acts on skin disorders due to its properties and also other systemic disorders.So the study was taken to prove effective roll of *dushivishariagad* on psoriasis like dermititis in mice.

2.] Rational of Study:

Dooshivisha is a form of toxin (animal origin, plant origin, artificial poison) that has not been completely eliminated or neutralized due to various reasons, remains in the body for some time and eventually gets manifested in the form of some disease.*Dooshivisha* vitiates the *rakta* and hence manifests symptoms like *aruh*, *kitibha* and *kotha*[6]. For contemporary relevance, several agents can be considered as *dooshivisha*. Such can be categorized into animate poison, inanimate poison and artificial poison. Inanimate poisons include cumulative toxicity of herbs (phytotoxins, toxa-albumins), chronic exposure to metal & mineral (dyes, paints etc.) Animate poison may be considered as rat bite, *keeta*, *lootavisha* etc. Artificial poison include agriculture pesticides and fertilizers, medicinal preparations, alcoholism, incompatible diet. Skin manifestations like *Vaivarnya* (vitiated body complexion), *Kustha* (skin diseases), *Mandal* (cicular skin lesions),

Kotha (skin rashes), *Aruh* (eruptions on skin) are observed on exposure to above said *dooshivisha*[7].

Psoriasis, is a chronic inflammatory skin disease affecting about 2–3% of the worldwide population, is characterized by hyper proliferation and abnormal differentiation of keratinocytes. At present, psoriasis is incurable, and its cause remains unelucidated.However, many studies have reported various factors contributing to the pathogenesis of psoriasis, including genetics, the immune system and environmental conditions, thus recognizing it as a multi factorial disease. In the present days, the use of chemical agents as medicine for treating different disorders and ailments had provoked other serious side effects for human beings& various dermatological manifestations. Hence it is need of hour to develop medicine with no side effects .In this regard the herbal medicine provides the best solution. Nature had provided the humankind with immense treasure since the dawn of ancient times. The herbs and medicinal plants provide an array of broad spectrum of activity. It was felt that the use of such medicinal plants would be beneficial for treating different skin ailments such as eczema, itching psoriasis.

So, the study was taken to evaluate *DushivishariAgad*effectiveness in psoriasis like dermatitis.

3.] Research Question:
Whether*DushivishariAgad* is effective inImiquimod- Induced psoriasis like dermatitis in Balb/c Mice?

4.] Hypothesis:
DushivishariAgad may be effective in Imiquimod- Induced psoriasis like dermatitis in Balb/c Mice.

Null Hypothesis:
*DushivishariAgad*may not be effective in Imiquimod- Induced psoriasis like dermatitis in Balb/c Mice.

5.] Literary Review:
Critical review of *DushivishariAgad*and its ingredients will be done from various classical texts such as *Brihattrayi, Laghutrayi*andvarious *Nighantus*. Modern review and review from published research works related to this study topic will also be done and will be presented in systematic and scientific way.

5.1] Review *DushiVisha*:
*DushiVisha*which is week in nature and stay in the body for many years. Its action on the body become aggravated bya particular time (a bad whether, windy day and the rainy season), a particular place (damp country), by the particular diet (intake of incompatible food), and some other factors includes sleeping in daytime and indigestion[8]. *Acharya*Susrutahas described in detail regarding the same. Various *Acharyas*have classified *Visha*by different synonymous but all have the same opinion that there are mainly four types of *Visha*viz., *Sthavara*, *Jangama*, *Dushi*and*GaraVisha*.

Premonitory symptoms of *DushiVisha*: Drowsiness, heavy and painful limbs, droopiness in the joints,horripilation[9].

Symptoms of *DushiVisha*: After having toxic food, (Mandala) circular patches, *Kotha*(Urticaria), pallor and swelling of face, loss of the vital principles of the organism, swelling of extremity (Atrophy of the hands and legs), *Dusyodara* (Ascites), *Vamana* (Vomiting),*Atisara* (Diarrhea), epileptic fits, *Vishama-Jvara* (High-fever) and *Pipaasa* (Extreme thirst). Some of them are differentiated by *Anaha* (constipation), loss of hairs, involuntary discharge of semen, confused speech, *Kushtha* (leprosy), or some other similar disease[10].According to *AcharyaCharka* when *DushiVisha*attacks the *Rakta-Dhatu*, *Arunshika* (Dandruff), *Kitibha*(Psoriasis), *Kotha*(Urticaria)like disease are originated and then by arriving into all *Doshas*, It destroy the life of a person[11].

Concept of *DushiVisha*is very uniquely described by various *Acharyas*which is applicable to present condition of the universe. *DushiVisha*is no separate entity but the remnant part of *Sthavara*,*Jangama*and*KritrimaVisha*which enters the body and vitiates *Dhatu*when conditions are favorable[12].

5.2] Review *Dushivishari Agad*[13]-
Table- 1 Composition of *DushivishariAgad*

Dravy	Rasa	Vipaka	Virya	Guna	Karma
1.Pippali (Piper longum)	Katu	Madhur	Anushnashit	LaghuSnigdhaTikshna	KaphaVataghnaYogvahiDipan,Rasayan,Kushthaghn17
2.Jatamansi (Nordostachysjatamanis)	Tikta-Madhur Kshay	Katu	Sheet	LaghuSnigdha	KaphaghnaTridoshaghnaVishaghna, kushthaghna19
3.Lodhra (Symplocusracemosa)	Kshay	Katu	Sheet	LaghuRuksha	Kapha-Pittaghna,raktshodhak,kushtghna20
4. Ela (Elettariacardamomum)	Katu Madhur	Katu	Sheet	Laghu Ruksha	Tridoshaghna,kushtghna,kandughna21
5.Musta (Cyporusrotundus)	Tikta-Katu Kashay	Katu	Sheet	LaghuRuksha	Kapha-pittaghnaVishaghna,raktprasadak,twakdoshhar22
6. Suvarchika (Gynandropsispentaphylla)	Katu-Tikta	Katu	Ushna	LaghuRuksha	Kapha-Vataghna,vishghna22
7. Dhyamak (Cymbopogonmartini)	Tikta Katu	Katu	Ushna	Laghu Ruksha	Kapha-Pittaghna,raktajvikarhar,18
8.Tagar(Tabernaemontanadivaricata)	Tikta, Katu, Kashay	Katu	Ushna	Laghuruksha	Vishaghna, Kushthaghna23
9.Kushtha (Saussurealappa)	Tikta	Katu	Ushna	Laghu,ruksha	Vishghna,kandughna,twakroghar24
10.Yashtimadhu (Glycyrrhizaglabra)	Madhur	Madhur	Ushna	Guru, snigdha	Raktajvikar,kandughn, Tawakrog25
11.Chandan (Santalum album)	Tikta, Madhur	Katu	Shit	Laghu, ruksha	Kushthaghna, Raktashodhak26
12. Gairik Fe2o3 (Red oxide of iron)	Kashay Madhur	Katu	Sheet	Laghu Ruksha	Vishaghna27

***5.3] Review of psoriasis*[14]-** Psoriasis is a common skin condition where the skin develops areas that become thick covered with silvery scales.Psoriasis is a disordered immune system.The T-cells, a type of white blood cell, become over-stimulated. These areas become the reddened, inflamed, patches with white scale on them. Psoriasis areas are worsened by scratching and minor skin injuries or irritations. Psoriasis may itch or burn. The skin may split or crack in areas that bend. Psoriasis is regarded as an autoimmune disease in which genetic and environmental factors have a significant role. Psoriasis is a non-contagious, dry, inflammatory and ugly skin disorder, which can involve entire system of person. The most commonly affected sites are the scalp, tips of fingers and toes, palms, soles, umbilicus, gluteus, under the breasts and genitals, elbows, knees, shin etc.

5.3.1] Types of psoriasis:
Plaque Psoriasis (Psoriasis Vulgaris)- It is the most common form of psoriasis. It affects 80 to 90% of people with psoriasis. Plaque psoriasis typically appears as raised areas of inflamed skin covered with silvery white scaly skin.These areas are called plaques.

Pustular Psoriasis-Appears as raised bumps that are filled with non-infectious pus(pustules). The skin under and surrounding pustules is red and tender. Pustular psoriasis can be localised, commonly to the hands and feet, or generalised with widespred patches occurring ranodomly on any part of the body.

NailPsoriasis- Produces a variety of change in the appearance of finger and toe nails. These changes include discolouring under the nail plate, pitting of the nails, thickening of the skin under the nail, and the loosening(onycholysis) and crumbling of the nail.

Guttate Psoriasis- It is characterized by numerous small oval (teardrop- shaped) spots. These numerous spots of psoriasis appear over large areas of the body, such as the trunk, limbs and scalp. Guttate psoriasis is associated with streptococcal throat infection.

Flexural Psoriasis (Inverse Psoriasis)-Appears as smooth inflamed patches of skin. It occurs in skin folds, particularly around the genitals

(between the thigh and groin), the armpits, under an overweight stomach (pannus), and under the breasts (inframammary fold). It is aggravated by frication and sweat, and is vulnerable to fungal infections.

Erythrodermic Psoriasis-Involves the widespread inflammation and exfoliation of the skin over most of the body surface. It may be accompanied by severe itching, swelling and pain.It is often the result of an exacerbation of unstable plaque psoriasis, particularly following the abrupt withdrawal of systemic treatment.

5.3.2] Pathophysiology-

Psoriasis is inflammatory immune mediator disease it is develops areas in various body's part. A host of abnormalities seen in psoriasis, like increased level in cyclic adenosine monophospate (cAMP), Epidermal growth factor receptor binding, protein kinase c and transforming growth factor (TGF collectively point to a disturbance in T cell fuction. Skin is a primary lymphoid organ with an effective immunological surveillance system equipped with antigen presenting cells, cytokine synthesizing keratinocytes, epidermotropicTcells dermal capillary endothelial cells, draining nodes, mast cells, tissue macrophages, granulocytes, fibroblasts, and non-Langerhans cells. Skin also has lymph nodes and circulating T lymphocytes. Together these cells communicate by means of cytokine secretion and respond accordingly via stimulation by bacteria, chemical, Ultraviolet (UV) light, and other irritating factors. The primary cytokine released in response antigen presentation is tumor necrosis factor-alpha (TNF). Generally, this is a controlled process unless the insult to the skin is prolonged, in which case imbalanced cytokine production leads to a pathological state such a psoriasis.The pathogenesis of psoriasis has been much studied and fairly well characterized; however, the precise inciting event in its initial development remains unclear, psoriasis is fundamentally an autoimmune disease whereby CD4- positive, and a lesser extent CD8- positive, T lymphocytes are stimulated to evoke a type- 1 inflammatory response. Central to this response is the liberation of pro-inflammatory cytokines such as TNF-alpha which have the net result of producing local tissue damage and increasing the turnover rate of the epidermis. Where normal squamous epithelial cells require roughly 15 days to traverse from the basal layer to the stratum corneum, in affected epithelium, this rate is reduced to 1-3 days, This rapid turnover of affected squamous epithelium results in the silver scale that is characteristic of psoriasis.

5.3.3] Causes of Psoriasis: The cause of psoriasis is unknown, but there is some evidence of disordered arachidonic acid metabolism, Include an auto-immune disorder, stress, environmental factors, hormones, drugs, infections and sunlight.

5.3.4]Symptoms:

- Over 65% of patients complain of itching, Dry, thin, silvery-white scales variable in amount and thickness
- Patients may report that their disease worsens in the winter and improves in the summer
- Skin on epidermis layer red patches show blister various parts in body.

5.3.5] Treatment of Psoriasis:

Topical Treatment- Corticosteroids: They are commonly first-line therapy in mild to moderate psoriasis and in sites such as the flexures and genitalia, where other topical treatments can induce irritation. Improvement is usually achieved within 2 to 4 weeks, then maintenance is achieved by use in the weekends only where other topical treatments can induce irritation topical steroids under of recalcitrant psoriasis of the scalp, hands, feet and other areas .to avoid systemic effects of glucocorticoid, a maximum of 50 g ointment may be used per week. Vitamin D: Potent topical corticosteroids are superior to Calcipotriene. The efficacy of calcipotriene is not reduced with long-term treatment. Calcipotriene is applied twice daily. Salicylic acid inactivates calcipotriene.

Coaltar: Coal Tar is the dry distillation product of organic matter heated in the absence of oxygen Coal tar, in concentrations 5- 20% can be compounded in creams, ointments, shampoos and and pastes. It is often combined with salicylic acid (2-5%), which by its keratolytic action leads to better absorption of the coal tar. Disadvantages include: allergic reactions, folliculitis, it has an unwelcome smell and appearance and can stain clothing and other items. Coal tar is carcinogenic.

Tazarotene: It is a third-generation retinoid. It reduces mainly scaling and plaque thickness, with limited effectiveness on erythema. It is available in 0.05 percent and 0.1 % gels, and a cream. When used as a mono therapy, a significant proportion of patients develop local irritation (especially with the 1% formulations).

Phototherapy treatment Natural ultraviolet light from the sun and artificial ultraviolet light are used to treat psoriasis.A combination of a drug that makes skin more sensitive to light and ultraviolet light.

6.] Research Gap Analysis

Many research studies regarding *Dushivisha*(Cumulative poison) and management of it are carried out in context of *Agadtantra*,But not a single research studies in the field of *Ayurveda*iscarried out to explore the *DushivishariAgad*and its effect in psoriasis like dermatitis.

Psoriasis is a complex multifunctional inflammatory skin disease characterized by T-cell activation, local vascular changes, abnormal keratinocyte proliferation and neutrophil activation. Psoriasis is a dreadful disease affecting physical, mental and social status of the victims. A new understanding of this complex disease has catalyzed the development of targeted biological treatments. These revolutionary therapies are not without potential risk, however alternative natural therapies provides some options for increasing safety and efficacy in the management of psoriasis[14].

In the present days, the use of chemical agents as medicine for treating different disorders and ailments had provoked other serious side effects for human beings. Hence it is need of hour to develop medicine with no side effects .In this regard the herbal medicine provides the best solution.

7] AIM AND OBJECTIVES-

7.1] Aim-

Study the efficacy of *DushivishariAgad* on Imiquimod- Induced psoriasis like dermatitis in Balb/c Mice with reference to kitibhakushtha.

7.2] Objectives:

- To study physicochemical & phytochemical properties of *DushivishariAgad.*
- To study efficacy of *Dushivishariagad*on Imiquimod-Induced psoriasis like dermatitis in Balb/c mice with reference to*kitibhakushtha*.
- To study concentrations of IL-17 and IL-23 in the serum and skin tissue of mouse.
- To study the Histopathological changes in dorsal surface of skin.

8.] MATERIAL AND METHODS:

8.1] Pharmaceutical study:

In this study *DushivishariAgad*will be prepare its standardization will be done on the basis of its physicochemical & phytochemical study will be done.

8.1.1] Procurement of Raw materials: All required raw materials will be purchase.

8.1.2] Authentication of Raw materials: All Raw drugs will be verified and authentified from dravyagun department of MGAC.

8.1.3] Classical method of preparation of *Dushivishariagad*[†]-

Table- 2 Preparation methods of DushivishariAgad

Pippali (Piper longum)
Jatamansi (Nordostachysjatamanis)
Lodhra (Symplocusracemosa)
Ela (Elettariacardamomum)
Musta (Cyporusrotundus)
Suvarchika (Gynandropsispentaphylla)
Dhyamak (Cymbopogon martini)
Tagar(Tabernaemontanadivaricata)
Kushtha (Saussurealappa)
Yashtimadhu(Glycyrrhizaglabra)
Chandan(Santalum album)
Gairik Fe2 o3(Red oxide of iron)

The contents of *DooshivishariAgad*will betaken in equal parts.

8.2] Analytical Study:

8.2.1) Description (Organoleptic characters of *DushivishariAgad*)
- *Rupa*(Colour)
- *Rasa*(Taste)
- *Gandha*(Odour)
- *Sparsha*(Touch)

8.2.2)Physico-chemical Parameters for *DushivishariAgad*-
- Ash value
- Total Ash
- Acid insoluble ash
- Waterl soluble Ash
- Extractive value a. Water soluble b. Alcohol soluble c. Methanol soluble
- pH Value
- Conductivity
- HPTLC

8.3] Experimental Study-
Sampling procedure:
8.3.1] Type of Study: Experimental study.
Animals required for this study i.e. Eight- to eleven-week-old BALB/c male mice weighing approximately 20 g each will be taken from Animal house for the study Animal will be acclimatized for 7 days prior beginning of study. Animals will be fed with food supplied from animal house and tap water.

Prior to and during the experimental period, all mice were assessed for their health status, including food and water intake, bodyweight, behavioral signs, respiratory patterns, and cardiovascular signs. Experiments were conducted according to international and national guidelines for ethical conduct in the care and use of Animals and were approved by the Animal Ethics Committee. The dorsal skin of the mice was shaved and a commercially available 5% IMQ cream was applied. The mice were randomly divided into 5 groups of 6animals and received the treatment on day 1 and continuing for 16 consecutive days, mice in all groups except for the normal group received a daily topical dose of 62.50mg of the IMQ cream on the shaved area of their backs. This translates into a daily dose of 3.125mg of the active ingredient[15]. A test substance *Dushivishariagad* and a standard reference (acitretin) were freshly prepared in water before being given to the mice. Mice received the treatment once a day in the morning for 10 days(7 to16days) by oral gavage (sterile disposable plastic needles).

Dosage: Human dosage of *DooshivishariAgada* mentioned in classics is 12 gm[16] and Animal Dose will be calculated by dose conversion table of Paget and Barnes (1964)-12 multiplied 0.018=0.216gm/20gmweight of Balb/c mice that is 0.0108gm means 10.8mg per mice.After experiment the animals will be sacrificed for histopathological examination.

8.3.2] Study Design: Experimental

9.3.3] Experimental Animal:
Table- 3 Details of Experimental Animal

Animal species to be used	Balb a/c mice
Sex of animals to be used	Male in each group will be taken.
Average weight of animals	20-22 gm.
No. of animals	06 mice for each group
No. of groups	05

9.3.4] Grouping[15]:
Table-4 Plan of the Study

Group	No of animals	Drug Given	Dose	Duration	Route of administration
Group A (Normal Control group)	06	Apply Vaseline on shaved dorsal skin surface.	_	For 16 days.	Local
Group B (positive control Group)	06	5% IMQ cream on shaved back.	3.125mg	For 16 days.	Local
Group C (Standard control group)	06	IMQ cream on shaved back plusacitretin orally.	IMQ Cream 3.125mg and Acitretin 5.14mg once daily	for 10 days (from 7-16 day)	IMQ cream-Local Acitretin-Oral
Group D (Test group 1)	06	IMQ cream on shaved back plus DVA with plain water will be given.	IMQ Cream 3.125mg and DVA 10.8mg per mice suspension	For 10 days.	IMQ cream-Local DVA-Oral
Group E (Test group 2)	06	IMQ cream on shaved back plus DVA with Honey will be given.	IMQ Cream3.125 mg and DVA 10.8mg per mice with honey.	for 10 days.	IMQ cream-Local DVA-Oral

8.3.5] Sample size- 30Balb a/c mice

8.3.6] Study Duration- 16days.

8.3.7] Study Centers:
- The study will be conducted at the following concerned centres.
- Animal house of DMIMS (DU), Sawangi, Wardha.
- Mahatma Gandhi Ayurved college, Hospital and Research Institute, Salod(H) Wardha.
- Histopathological lab of JNMC, Sawangi ,Wardha.
- As per the necessity the study will be conducted at other renowned institute.

8.3.8] Assessment Parameters:
- All animals from all groups will be screened for following parameters on day 1 & throughout the experiment upto day 16 of study.
- Psoriasis area and severity index (PASI) scores were determined by evaluating the degree of erythema, thickening, and scaling on the affected dorsal skin surface.
- PASI for eachwas measured on a four-point scale (0 = none; 1 = slight;2 = moderate; 3 = marked; 4 = very marked). The severity of skin inflammation was measured by the combined scores(erythema plus scaling plus thickening) giving a range ofscores of 0–12.

Table-5 Assessment Scores:

PASI Score Assessment on dorsal skin surface	0=None	1=slight	2=moderate	3=marked	4=very marked
Erythema					
Thickening					
Scaling					

Mice were then sacrificed on day 17 at the laboratory. Central dorsal skin tissue (approximately 1cm2) from all the groups were excised for histological studies.

3. To measure cytokine levels in serum, mouse blood will be collected at 24 h after the final treatment .The concentrations of IL-17 and IL-23 in the mouse serum and skin tissue were measured.

8.3.8] Analysis plan (Statistical test):
Statistical analysis will be done by applying suitable tests. (ttest,One way ANOVA test)

8.3.9] RESULTS
Results will be drawn from the observations of statistical test, biochemical parameters and histopathology reports.

8.3.10] CONCLUSION
Conclusion will be drawn on the basis of results of Statistical Analysis and histopathology reports.

9.] Scope Of The Proposed Study:
Present study is an attempt to discover a new cost effective formulation as the ingredients of *Dushivishariagad*are easily available and the method of preparation does not require much time and manpower, hence if this formulation showed significant efficacy then this will open a new gateway for further researches involving management of chronic disorder like psoriasis.If desired effects occur due to ingestion of *Dushivishariagad* then it will generate collateral evidence for the said statement of *Charakaacharya.*As a result of above, it will also provide the preventive measure for side effects due to modern medicine.

10.] Ethical Clearance:
This study will be conducted after obtaining due Clearance from

Institutional Ethical Committee and I.A.E.C of DatttaMeghe Institute of Medical Sciences, Sawangi, Wardha,

REFERENCES:

1. AmbikaDuttaShashtri,SushrutSamhita,Published by Chaukhamba Sanskrit Sansthan Varanasi, 2013, Kalpasthan, 2/24- 29.
2. KashinathShastri, Charaksamhita (2011) reprint, Chikitsasathana, 23/31, Varanasi, ChaukhambhaBhartiAkadami, Pg.630
3. https://en.wikipedia.org/wiki/Psoriasis
4. BramhanandTripathi Editor, Ashtanghrudayam of Shrimadvagbhata edited with 'Nirmala Hindi Commentary, Chaukhamba Sanskrit pratishthan, Delhi, Reprint 2014, Uttarsthana, Vishapratishediyadhyaya, chapter 35, verse 38-39.
5. Sharma RK,DashB.English translation on charakasamhita of Agniveshas Reprint 2014 chikitsasthana,kushthachikitsa: chapter 7,verse12-14:Varanasi(India):chaukhamba Sanskrit series office,2014;p.324-29.
6. Agnivesa. Chikitsasthana chapter 23/31. In: Charaka, Dridhbala, Chakrapanidutta (eds.) CharakaSamhita with Ayurveda Dipika Commentary. Varanasi: ChaukhambhaOrientalia; 2015. p. 573.
7. Jangid SK, Jadhav DK. "ayurvedic management of cumulative toxicity (dushivisha)": a review.
8. AmbikadattaShastri, Sushrutsamhita-purwardha, kalpasthana (2003) reprint, Varanasi, Chukhambha Sanskrit Sansthan, Pg.566
9. AmbikadattaShastri, Sushrutsamhita-purwardha, kalpasthana (2012) reprint, Chapter-2/28,Varanasi, ChukhambhaSanskritSansthan, Pg.33
10. AmbikadattaShastri, Sushrutsamhita-purwardha, kalpasthana (2012) reprint, Chapter-2/30-32, Varanasi, Chukhambha Sanskrit Sansthan, Pg.33
11. KashinathShastri, Charaksamhita (2011) reprint, Chikitsasathana, 23/31, Varanasi, ChaukhambhaBhartiAkadami, Pg.630
12. AmbikadattaShastri, Sushrutsamhita-purwardha, kalpasthana (2012) reprint, Chapter-2/25, Varanasi, Chukhambha Sanskrit Sansthan, Pg.32.
13. Priyavatsharma, DravyagunVidnyan Part II- page no. 276, 31, 616, 716, 715, 370, 64 572.8.Acharya priyavat Sharma /Dr. Guruprasadsharma, DhanvantariNighantu, chaukhambaOriyantaliya, Varanasi 2005, 78, 79, 30.
14. PapolaVibhooti,Fight Psoriasis Naturally Through Ayurveda, Indo American Journal of Pharmaceutical Research.2016:6(07). ShriDevBhoomi Institute Of Education Science & Technology, Dehradun, Uttarakhand, India.
15. Mingkwan Na Takuathung**,** Antipsoriatic Effects of Wannachawee Recipe on Imiquimod-Induced Psoriasis-Like Dermatitis in BALB/c Mice,Hindawi Evidence-Based Complementary and Alternative Medicine Volume 2018.
16. YadavajiTrikamaji, editor. Commentary: nibandasangraha of DalhanaAcharya on Sushrutasamhita of Sushrutakalpastana, chapter 2, verse no.25, 10th edition, Varanasi: Chowkambha Sanskrit Sansthan;2012;p.565.
17. N. Jayachandra Reddy, D. NagoorVali, M. Rani, S. SudhaRani,Evaluation of antioxidant, antibacterial and cytotoxic effects of green synthesized silver nanoparticles by Piper longum fruit, Department of Biochemistry and Molecular Biology, School of Life Sciences, Pondicherry University, Pondicherry, India
18. E S Sunila,GKuttan**,**Piperlongum inhibits VEGF and proinflammatory cytokines and tumor-induced angiogenesis in C57BL/6 mice,International Immunopharmacology,6(5):733-41,June 2006, PubMed.
19. Biswajit Bose, DebabrataTripath,Secondary metabolite profiling, cytotoxicity, anti-inflammatory potential and in vitro inhibitory activities of Nardostachysjatamansi on key enzymes linked to hyperglycemia, hypertension and cognitivedisorders, Phytomedicine 55:58-69,10.1016/j.phymed.2018.08.01.
20. IndraD.Bhatt ,PreetiDauthal,Characterization of essential oil composition, phenolic content, and antioxidant properties in wild and planted individuals of *Valerianajatamansi* Jones, ScientiaHorticulturae Volume 136, 1 March 2012, Pages 61-68.
21. Vijayabaskaran M, BabuG,In vitro antioxidant potential of ethanolic bark extract of symplocosracemosaroxb,International Journal of Pharmacy and Technology,2(3): 320-328,July2010
22. SrabanaMaitra et al ,Pharmacological Study of SymplocosracemosaRoxb, Ijppr.Human, 2017; Vol. 9 (1): 106-113
23. AnuradhaVaidya et al,An in vitro study of the immunomodulatory effects of Piper nigrum (black pepper) and Elettariacardamomum (cardamom) extracts using a murine macrophage cell line, American International Journal of Research in Formal, Applied & Natural Sciences, 8(1), September-November, 2014, pp. 18-27.
24. S K verma,vartikajain,blood pressure lowering, fibrinolysis enhancing and antioxidant activities of cardamom (elettariacardamomum), Indian J. Biochem. Biophys., Vol. 46, december 2009
25. F. Salah Akrayi,Antibacterial Effect of Seed Extracts of Cardamom (Elettariacardamomum) against Staphylococcus aureus and Proteus mirabilis, Tikrit Journal of Pure Science 17 (2) 2012
26. SabrinRagab Mohamed Ibrahim ,Anti-inflammatory terpenoids from Cyperusrotundus rhizomes, Pak. J. Pharm. Sci., Vol.31, No.3(Suppl), July 2018, pp.1449-1456.
27. Kilani-Jaziria,b, W. Bhouri,Phytochemical, antimicrobial, antioxidant and antigenotoxic potentials of Cyperusrotundus extracts , South African Journal of Botany 77 (2011) 767–776
28. Kapil Lawrence, Reena Lawrence, Anioxidant activity of palmarosa essential oil (Cymbopogon martini) grown in north indian plains, Asian Pacific Journal of Tropical Disease (2012)S765-S768
29. BrunaFernanda,*Cymbopogonmartinii essential oil and geraniol*at noncytotoxic concentrations exerted immunomodulatory/anti-inflammatory effects in human monocytes, 2014 Royal Pharmaceutical Society, Journal of Pharmacy and Pharmacology, 66, pp. 1491–1496
30. FatemehSaeidienik,The Effects of Iron Oxide Nanoparticles Administration on Depression Symptoms Induced by LPS in Male Wistar Rats, Basic ClinNeurosci. 2018 May-Jun; 9(3): 209–216.
31. Ali TalhaKhalil,Biosynthesis of iron oxide (Fe2O3) nanoparticles via aqueous extracts of Sageretiathea (Osbeck.) and their pharmacognostic properties, Green chemistry letters and reviews, 2017 vol. 10, no. 4, 186–201.
32. RajandeepKaur,Glycyrrhizaglabra: a phytopharmacological review, *IJPSR, 2013; Vol. 4(7): 2470-2477.*
33. Syed LuqmanShah,Inhibitory Effects of *Glycyrrhizaglabra and* Its Major Constituent Glycyrrhizin on Inflammation-Associated Corneal Neovascularization, Hindawi Evidence-Based Complementary and Alternative Medicine,Volume 2018.
34. Md. AtaurRahman,Antiproliferative properties of *Saussurealappa Clarke root extract in SH-SY5Y neuroblastoma*cells via intrinsic apoptotic pathway, Animal Cells and Systems, 19:2, 119-126.
35. Shirishkumar D, Ambavade, Anxiolytic activity of root extracts of *Saussurealappa Clark. inmice,Journal of Natural Remedies, Vol. 6/2 (2006) 103 – 108.*
36. *SunkaraYashvanth,* Anti-inflammatory and cytotoxic activity of chloroform extract of roots of *Saussurealappa Clarke, Journal of Pharmacy Research 2010, 3(8),1775-1778.*
37. Eslammelakawe,Antimicrobial activity of methanolic *Nigella sativa* seeds and *Saussurealappa* roots extracts on multidrug resistant Gram negative bacteria, African journal of Biological Science Article 14, Volume 13, Issue 1, 2017, Page 155-163.
38. RakeshKumar,Phytochemistry and Pharmacology of Santalum album L.: A Review, World Journal of Pharmaceutical Research · October 2015
39. SaurabhChhatre ,Phytopharmacological overview of *Tribulusterrestris,*Pharmacogn Rev. 2014 Jan-Jun; 8(15): 45–51.
40. AmandeepKaur, Medicinal uses of nahush (*valerianawallichii dc.): A review article* , *wjpmr, 2019,5(6), 155-156*