A HISTOPATHOLOGICAL STUDY ON LEECH APPLICATION IN THE MANAGEMENT OF VICARCIKA (ECZEMA)

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INTRODUCTION

Skin is an extraordinary structure. It is frequently damaged because it is directly in the 'firing line' and, for this reason, skin diseases are very common. There are more than 3000 known diseases of the skin.1 Although most of the chronic skin conditions, such as eczema, psoriasis, vitiligo and leg ulcers, are not immediately life-threatening, they are recognized as a considerable burden on health status and quality of life, including physical, emotional and financial consequences.

Eczema or dermatitis is a pattern of inflammatory response of the skin, which is characterized by erythema, edema, vesiculation, exudation and crust formation.2 Acute eczema presents with weeping, crusting and vesicle formation and sub acute or chronic with dryness, scaling, fissuring and lichenification.

Histologically, dermatitis is characterized by inflammation of the
epidermis i.e. perivascular lymphohistocytic infiltrate. The hallmark of acute dermatitis is spongiosis (intraepidermal vesicles) and exocytosis of lymphocytes. In sub acute dermatitis, apart from the features of acute dermatitis, parakeratosis (increase in thickness of stratum corneum) and acanthosis (thickened epidermis) are prominent. As eczema becomes more chronic, there is tendency for it to become more acanthotic and less spongiotic\(^3\) accompanied by T-helper lymphocytic infiltration in the upper dermis.\(^4\)

Management of eczema involves topical applications like emollients, antipuritic lotions, steroids, coal tar and zinc paste etc, oral antibiotics, sedative antihistamines, corticosteroids and immunosuppressive agents.\(^5\) Even after their use relapses, recurrences and other complications are very common. Indiscriminate use of these drugs can cause vomiting, diarrhea, stomach upset, allergic reaction, peptic ulcer, neuropsychiatric reactions, diabetes, diminution of libido, Cushing’s syndrome etc.

In maximum cases, failure of contemporary medicine in the treatment of eczema push them to chronic state and hence report to Ayurvedic system in the later stage. *Vicarica*ka has been mentioned in Ayurvedic classics under *Kshudra Kustha*\(^6\) and also under *Kshudra roga*.\(^7\) According to Ayurveda, *Rakta dushti* (vitiation of blood) is one of the prime causes of skin diseases, among the *Shodhana karma* (purificatory treatments); bloodletting is considered as the best treatment for skin diseases. According to *Sushruta*, if a person regularly undergoes bloodletting, he can develop resistance against all types of skin diseases.\(^8\) In the present study, among the various methods of bloodletting, leech application was selected as it is specifically indicated for *Tvak dosha* (skin disease)\(^9\) and biochemicals released from leech saliva have antinflammatory, antibiotic, thrombolytic actions etc., further leeches can be easily administered with less complication and are cost effective. Many clinical studies have already proved the efficacy of leech application in skin diseases especially in eczema, but still effect of leech application on histological level was not explored. Hence this study was designed to prove the efficacy of leech application in eczema at the tissue level through Histopathological examinations.

**MATERIALS AND METHODS**

**Selection of Patients:**

Ten patients having the classical symptoms of Eczema were randomly selected from O.P.D. & I.P.D. sections of Dept. of *Shalya*, S.V. Ayurvedic Hospital, Tirupati. The inclusion criteria included patients of age group 16 to 70 yrs (as bloodletting is indicated for this age group).\(^10\) The exclusion criteria included, patients having secondary infections, bleeding disorders, Diabetes Mellitus, Heart diseases, Tuberculosis, Leprosy and Generalized eczema. A specific case sheet was prepared and the patients of the present study were examined in detail as per case sheet. Routine hematological investigations and urine examinations were carried out before and after treatment to rule out the other associated pathology. The patients were included for study after taking written consent for ethical clearance.

**Treatment:** Minimum 4 sittings of leech application therapy were carried out with seven days interval. Number of leeches applied was decided on the basis of the size
of lesion. Procedure of leech application was undertaken in the following stage:

**Pre operative procedure:** Non poisonous Leeches were identified and collected from fresh water sources; they were stored in mud pots. On the day of application, the patient was placed on proper position according to the site of lesion. Leeches (no. as required) were kept in turmeric water to make them active, increase appetite and free from natural urges, after keeping the leeches for 45 minutes they were put in fresh water.

**Operative procedure:** Lesion is cleaned by water and dried with cotton. After this, rubbing of skin was done as per need. For this purpose, either sterilized cotton bandage pad or thick thread of jute was used. The activated leeches were applied over the lesion site. Leeches become elevated like a horse shoe or raised in arched position from its neck which indicates that they are sucking the blood vigorously or strength fully. After catching the site, leeches were covered by wet cotton pad and regularly sprinkled with cold water. The leeches usually detach from the site after sucking the vitiated blood or have to be removed by sprinkling salt over it when the patient complains pain or itching.

**Post operative procedure:** After detachment of leeches, wound was cleaned with anti septic solution and after that tight bandage was applied for prevention of secondary hemorrhage. After falling off, the leeches were dusted with rice powder and turmeric powder was applied on the mouth to make it vomit the sucked blood (otherwise it may cause death of leeches). Leeches after vomiting were put in fresh water and when they move freely without sinking down were considered as properly vomited. Used leeches were kept in a separate pot containing pure water and used for the same patient after a gap of 7 days. The frequency of leech application will vary according to the severity of disease. Generally leeches should be applied once in a week up to 4-6 sittings. Leeches should be reserved for a particular patient to avoid cross infection.

**Contra indication of leech therapy**
2. Severe anemia.
3. Allergic reaction to active substances of the leech such as hirudin, hyaluronidase, egline, apyrase, destabilase, etc.
4. Human with weak constitution.
5. Pregnancy.

**Precautions during leech application**
1. Bleeding and clotting time of the patient should be normal.
2. Gentle handling of the leech.
3. Cover the leech with wet cotton.

**Histopathological analysis**

Skin biopsy (Incisional biopsy) was taken from the lesion site before and after the treatment. Sample was transported to the pathology lab in a fixative (10% formalin solution). For the histological examinations, paraffin-embedded tissue sections of the skin (5 µm) were stained with hematoxylin-eosin (HE). The tissue samples were then examined and photographed under a light microscope (Olympus CX31, Olympus, Japan), for observation. The Histopathological parameters were fixed as follows: Acute dermatitis – Spongiosis and exocytosis of lymphocytes; Sub acute dermatitis – Parakeratosis, acanthosis, Spongiosis and exocytosis of lymphocytes; Chronic dermatitis - Irregular acanthosis, papillary dermal fibrosis and focal superficial epidermal necrosis.
RESULTS

Effects of Leech application in Eczema in Histopathological alterations

Figure 1.A (40X mag)  Figure 1.B (10X mag)
Figure 1.A (before treatment histopathology photo of Case 1) shows hyperkeratosis, spongiosis, irregular acanthosis with blunting of the rete ridges and presence of lymphocytes in the epidermis. Figure 1.B (after treatment histopathology photo of Case 1) shows decrease in acanthosis, mild hyperkeratosis, decreased inflammatory component and decrease in spongiosis.

Figure 2.A (40X mag)  Figure 2.B (40X mag)  Figure 2.C (40X mag)
Figure 2.A (before treatment histopathology photo of Case 2) shows hyperkeratosis and irregular acanthosis of epidermis with focal areas of spongiosis with irregular rete ridges (with widening of retephages) and prominent presence of melanocytes. Shows collection of non specific inflammatory cells in the sub epidermis and dermis reveals fibrocollagenous tissue with few adnexa. Figure 2.B (after treatment histopathology photo of Case 2) shows decrease in the hyperkeratosis, melanin pigmentation is decreased, irregular widening of rete ridges is decreased and the inflammatory component is reduced. Figure 2.C (after treatment histopathology photo of Case 2) shows the presence of granulation tissue in the dermis.
Figure 3.A (40X mag)
Figure 3.A (before treatment histopathology photo of Case 3) shows hyperkeratosis increase in collection of inflammatory cells and spongiosis.

Figure 3.B (40X mag)
Figure 3.B (after treatment histopathology photo of Case 3) shows decrease in hyperkeratosis, spongiosis and collection of inflammatory cells.

Figure 4.A (40X mag)
Figure 4.A (before treatment histopathology photo of Case 4) shows the presence of hyperkeratosis with keratin plug.

Figure 4.B (40X mag)
Figure 4.B (after treatment histopathology photo of Case 4) shows decrease in hyperkeratosis, spongiosis and collection of inflammatory cells.

Figure 5.A (10X mag)
Figure 5.A (before treatment histopathology photo of Case 5) shows hyperkeratotic epidermis with irregular rete ridges and mild spongiosis. Dermis reveals collagenised fibrous tissue and collection of inflammatory cells.

Figure 5.B (10X mag)
Figure 5.B (after treatment histopathology photo of Case 5) shows hyperkeratotic epidermis with no irregular rete ridges and decrease in spongiosis. Dermis reveals granulation tissue.
Figure 6.A (10X mag)
Figure 6.A (Case 6) shows that a focal part of epidermis is removed probably by leech bite.

Figure 6.B (10X mag)
Figure 6.B (Case 6) shows the gap being filled with healthy cells in the latter stage.

Figure 7.A (40X mag)
Figure 7.A (before treatment histopathology photo of Case 7) shows hyperkeratosis, spongiosis and mild acanthosis with collection of leucocytes.

Figure 7.B (40X mag)
Figure 7.B (after treatment histopathology photo of Case 7) shows hyperkeratosis, decrease in spongiosis and acanthosis. The sub epidermal zone reveals formations of granulation tissue.

Figure 8.A (40X mag)
Figure 8.A (before treatment histopathology photo of Case 8) shows irregular pattern of epidermis.

Figure 8.B (40X mag)
Figure 8.B (after treatment histopathology photo of Case 8) shows mosaic pattern (normal pattern) of epidermis with normal intra epithelial component.
Figure 9.A (before treatment histopathology photo of Case 9) shows hyperkeratosis, irregular acanthosis, extensive spongiosis and collection of non-specific inflammatory cells in the epidermis. Dermis reveals bundle collagens with inflammatory components and intra epithelial collection of inflammatory cells.

Figure 9.B (after treatment histopathology photo of Case 9) shows decrease in hyperkeratosis, no spongiosis and acanthosis and absence of inflammatory cells.

Figure 9.C (after treatment histopathology photo of Case 9) shows normal mosaic pattern arrangement of epidermal cells.

Figure 10.A (before treatment histopathology photo of Case 10) shows hyperkeratosis, spongiosis and acanthosis with more collection of melanin pigments. Epidermis shows collection of inflammatory cells. Dermis reveals collagen bundles, congested vessels and a few collections of non-specific inflammatory cells.

Figure 10.B (after treatment histopathology photo of Case 10) shows decrease in hyperkeratosis, spongiosis and acanthosis, with decrease in melanin pigments. Normal mosaic pattern of epidermal cells is noted. Inflammatory components in the epidermis are decreased. Figure 10.C (after treatment histopathology photo of Case 10) shows normal pattern of epidermis and presence of granulation tissues.
DISCUSSION

In eczema or dermatitis the leech application gives excellent response by reducing the inflammatory component, hyperkeratosis, spongiosis, irregular acanthosis and by evoking a granulation tissue response in the dermis.

Spongiosis is mainly intercellular edema between the keratinocytes in the epidermis, and is characteristic of eczematous dermatitis. Leech saliva releases anti inflammatory components like Hylaurinadase, Eglins and Bdellins, they may reduce the intercellular edema and hence spongiosis was found to be remarkably reduced in most of the cases.

Hyperkeratosis is a thickening of the outer layer of skin, producing a tough, often dry area that may be discolored. Hyperkeratosis can trigger localized hyper pigmentation in the stratum corneum of the epidermis, along with an increased production of protective keratin. Hyperkeratosis can be treated by removing the thickened layer of skin (containing keratin) to be replaced with a normalized layer of cells. Hyper pigmentation associated with hyperkeratosis can be effectively treated by combining a strategy to stimulate melanocyte degeneration and promote skin repair through hydration and collagen production. Leech therapy induces granulation tissue formation which in turn results in collagen formation causing keratolysis. There is also remarkable decrease in local/focal pigmented lesion which causes considerable reduction in hyper pigmentation especially in case of Sebhoric keratosis.

The medicinal leech is a beautiful symbol of ‘give and take’ and is sustainable resource management. Hirudo medicinalis is one of the oldest surviving animals on earth. The first documented accounts of the use of Leeches for medicinal purpose is from the time of extreme antiquity, more than 2500 years before in Ayurvedic texts, then long later during the period of Hippocrates. Dhanvantari, the Indian God of Ayurveda hold a leech in one of his hand. This simply suggests the importance of Leeches in medicinal field by ancient Indian science. Leeches have and will always be thought of as the ‘wonder doctors’ of science.

Although leeches may not be safe for people with diseases that impair blood clotting or for those with compromised immune function, it is believed to be safe for most of other people. The saliva of leeches contain a variety of substances such as Hirudin, Hylaurinadase, histamine like vasodilators, Eglins, Bdellins, Collagenase, destabilize, inhibitors of Kallikerin, super oxide production and poorly characterized anesthetic and analgesic compounds.

These substances reach epidermis and dermis by the action of enzyme Hylaurinadase. It has been proved through Laser Doppler flowmetry that there is a significant increase in superficial skin perfusion following leech application, especially 16mm around the bitting zone. Therefore a regional antiphlogistic, thrombolytic and antibiotic effect by these substances enforced by hylaurinadase might be the possible reason of improvement by treatment with leeches. The jaws of the leech pierce the skin so that potent biologically active substances can penetrate in to the deeper tissues. Hylaurinadase (spreading factor), an enzyme in leech saliva, further facilitates the penetration and diffusion of these pharmacologically active substances in to the tissues.
Leech saliva contains variety of biochemicals which are anti inflammatory, thrombolytic, antibiotic, analgesic, vasodilator etc. These pharmacologically active compounds causes normalization and improvement of capillary as well as collateral blood circulation, anti-inflammatory effect, analgesia and anesthesia effect through saliva, Immune-stimulation and Immune-modulating effect and early wound healing effect.

In conclusion, the Histopathological study done from the samples of eczematous lesion before and after leech application therapy proves that there are significant changes at histological level specifically in reduction of inflammatory component, hyperkeratosis, spongiosis and irregular acanthosis. It was also found that there was a considerable formation of granulation tissue, which helps in formation of healthy new tissues. Hence the application of leeches in the established cases of eczema can be handy and effective measure of treatment from the ever relieving problems of eczema particularly in the chronic cases. The study proves the utility of leech application further in other inflammatory disorders can also be considered and further research is also required to know the exact protocol for complete cure.

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REFERENCES


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