



IJPPR

INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals

ISSN 2349-7203





Human Journals

Review Article

March 2020 Vol.:17, Issue:4

© All rights are reserved by Pradnya Janardhan Bhagat et al.

Efficacy of Jalaukawacharana Therapy in Acne- A Literature Review and Meta Analysis

	
<p>Pradnya Janardhan Bhagat*¹, Savita Udayshankar Sharma²</p> <p><i>¹Associate Professor, Dept. of Shalyatantra, Shivalik Institute of Ayurveda and research, Dehradun, Uttarakhand.</i></p> <p><i>²Research Officer (R.O. Ayu), RARIMCH, NIT Complex, Near Gharkul Parisar, Nandanvan, Nagpur, Maharashtra.</i></p> <p>Submission: 22 February 2020 Accepted: 29 February 2020 Published: 30 March 2020</p>	

Keywords: Acne, *Jalaukawacharana*, Leech therapy, *Mukhdushika*

ABSTRACT

Acne is a very common medical condition and more than 90 percent of world population has been affected by acne in their life. Among various types of acne, acne vulgaris is the most common, with prevalence in 99 percent of the acne cases. *Mukhdushika* described in Ayurveda texts resemble acne vulgaris of modern science, it is caused by vitiated *Kapha*, *Vata* and *Rakta*. *Raktmokshana* (bloodletting) is mentioned in Ayurvedic texts as the preferred way of treatment in *Raktaja* as well as dermatological disorders. Acharya Susruta stated *Jalaukawacharana* is the preferred method of bloodletting in *Sukumara* (tender nature person). *Jalaukawacharana* being a bio-purificatory method removes deep seated toxins by letting out blood, clearing *Srotasa* and pacifying vitiated *Doṣha*. *Jalaukawacharana* possesses high efficacy in both cleansing and healing properties. Leech's saliva contains a complex mixture of different biologically and pharmacologically active substances which gets secreted into the site. Destabilase, Bdelin, Histamine, Hyaluronidase, Fibrinases and collagenase are important component of leech secretions which has been proved for normalization and improvement of capillary as well as collateral blood circulation, Expressed anti-inflammatory effect, Analgesia and anesthesia effect through saliva, Immunostimulation and immuno-modulating effect. In this review, an attempt has been made for reviewing the potentials of leech therapy in treating *Mukhdushika* w.r.t. acne vulgaris. For this purpose, various researches of *Jalaukawacharana* carried out at different Ayurveda centers as a monotherapy or in combination with other therapy or drugs were evaluated for their acne treating potential.



HUMAN JOURNALS

www.ijppr.humanjournals.com

INTRODUCTION

Acne is a very common medical condition and more than 90 percent of world population has been affected by acne in their life. Approximately 85% of people between the ages of 12 and 24 years experience at least minor acne.¹ Although acne is a skin disease, it can lead to prominent emotional and psychological impact on any patient, regardless of the severity or the grade of the disease.² Adolescent acne usually begins with the onset of puberty, when the gonads begin to produce and release more androgen hormone, and it is then further exacerbated by genetic and other factors. Though it is a common disease of adolescent, it can occur at any stage of life and may continue into one's 40s. There are various types of acne, such as acne vulgaris, acne rosacea, acne cosmetica, acne mechanica, and acne fulminans. Among them all, acne vulgaris is the most common, with prevalence in 99 percent of the acne cases. Acne can run in families, and those with family history of acne often develop acne at an earlier age. If both the parents have acne, there is 75% chance that a given child will develop acne. Plugged follicle, increased sebum production, propionibacterium acnes, and inflammation are thought to promote acne. Acne lesions are divided into inflammatory and non-inflammatory lesions. Non-inflammatory lesions consist of open comedones (blackheads) and closed comedones (whiteheads). Inflammatory lesions consists of red papules, pustules, nodules and cysts.³ Acne vulgaris typically affects the areas of skin with the densest population of sebaceous follicles (i.e. face, upper chest, back). Local symptoms of acne vulgaris may include pain, tenderness and or erythema.⁴

Ayurveda approach in *Mukhadushika* (acne)

In Ayurveda, *Mukhadushika* has been elaborated as one of the *Kshudra Rogas* (minor ailments) and the symptoms of *Mukhadushika* resemble acne vulgaris as per modern science. Acharya Vagbhata defined *Mukhadushika* as, the eruptions like *Shalmali* thorn, on the face during adolescence caused by vitiated *Kapha*, *Vata* and *Rakta*.⁵ *Mukhadushika* is also termed as *Yuvanpidika* or *Tarunyapitika* by the some Acharyas as the disease is seen in adolescent and young age group prominently. Acharya Charaka has mentioned that the vitiated *Pitta* in *Tvaka* (skin) & *Rakta* produces a reddish swelling called *Pidika* (pimple or pustule).⁶ According to Acharya Vagbhata, this *Pidika* is filled with *Meda*, which can be explained with the term *Medogarbhita* (filled with oil/sebum). We can correlate this with modern science which explains the similar pathology. Obstruction of follicular ducts due to impacted

sebaceous and keratin secretions in accompaniment with *Propionibacterium acnes* colonization contribute to the pathogenesis of acne vulgaris.⁷

Ayurveda texts such as *Sushruta Samhita*, *Ashtanga Hrudaya* (by Acharya vagbhata), *Sharangadhara Samhita*, *Chakradatta* etc., have stated the pathophysiology and treatment of acne. According to Acharya Sushruta, vitiation of *Kapha dosha*, *Vata dosha* and *Rakta Dhatu* (blood) lead to the formation of acne.⁸ Acharya Kashyapa mentioned that *Shukra Dhatu* (*Semen*) development occurs in the young age and there is changes in sexual characters along with formation of acne.⁹ In *Sharangadhara Samhita*, it has been mentioned that acne is caused due to *Shukradhatumala* (waste products of semen).¹⁰

Regarding the treatment, Ayurveda texts have mentioned *Shamana* and *Shodhana* therapy in the treatment of *Mukhadushika*. *Shamana* therapy (conservative treatment) includes use of internal and external medication. Many formulations for external application such as *Lepa* (paste) and *Taila* (oil) have been mentioned in Ayurveda classics. *Shamana* therapy aims towards the balancing of vitiated *Doshas*. Several Ayurveda formulations and herbs have been used to treat acne. These herbs help to eliminate various toxic elements and functions as topical antiseptic too. These formulations are mentioned in table 1.¹¹

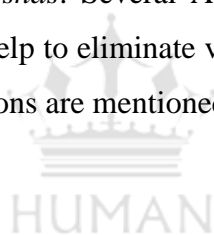


Table No. 1: Ayurveda formulations and herbs described in *Ayurveda* texts to treat acne.

S N	Herbs	<i>Lepa</i> (Pastes)	<i>Taila</i> (Oil)	Internal
1.	Lodhra <i>Symplocos racemosa</i>	<i>Marichadi Lepa</i>	<i>Kumkumadi Taila</i>	<i>KhadirastakaKashya</i>
2.	Yashtimadhuka <i>Glycyrrhiza glabra</i>	<i>Varunadi Lepa</i>	<i>Majishthadi Taila</i>	<i>Mahamanjishtadi kwatha</i>
3.	Dhanyaka <i>Coriandrum sativum</i>	<i>Vatankuradi Lepa</i>	<i>Haridradi Taila</i>	<i>Sarivadyasava</i>
4.	Vacha <i>Acorus calamus</i>	<i>Siddharthadi Lepa</i>	<i>Kanaka Taila</i>	<i>Guduchyadighanavati</i>
5.	Shalmali <i>Salmalia malabarica</i>	<i>Shalmali Kalkadi Lepa</i>	<i>Sarshapa Taila</i>	
6.	Manjishtha <i>Rubia cordifolia</i>	<i>Arjunadi Lepa</i>		
7.	Jatiphala <i>Myristica fragrans</i>	<i>Manjishthadi Lepa</i>		
8.	Daruharidra <i>Berberis aristata</i>	<i>Lodhradi Lepa</i>		
9.	Nimba <i>Azadirachta indica</i>	<i>Jatiphaladi Lepa</i>		
10.	Khadira <i>Acacia catechu</i>			
11.	Sariva <i>Hemidesmus indicus</i>			
12.	Kakamachi <i>Solanum nigrum</i>			
13.	Guduchi <i>Tinospora cordifolia</i>			

Shodhana therapy (cleansing or purification) includes *Vamana Karma* (stated by Sushruta and Vagbhata), *NasyaKarma* (stated by Vagbhata) and *Raktamokshana* (stated by Vagbhata and Chakradatta). Almost in all texts *Vamana* is the main treatment for *Mukhadushika*.

Vamana procedure is specially indicated in *Kaphaja* diseases in which *Doshas* are eliminated through mouth. *Virechana* procedure is also practiced by Ayurveda physicians as it is choice of treatment in *Raktaja* and dermatological disorders. *Nasya* therapy is mostly indicated in *Urdhwajatrugata* (disease occurring above clavicle) diseases in which medicines are introduced through nasal route. It is another type of *Shodhana Chikitsa* elaborated by Vagbhata for the treatment of acne.¹²

***Jalaukawacharana* Approach in *Mukhadushika* (Acne)**

Raktmokshana (bloodletting) is mentioned in Ayurvedic texts as the preferred way of treatment in *Raktaja* (blood born disease) as well as dermatological disorders. *Rakta Dhatu Dushti* is one of the main pathogenic factors of *Mukhadushika* formation. In *Mukhadushika*, Acharya Vagbhata and Chakradatta have indicated *Siravedha* (phlebotomy) as the preferred *Raktamokshana* method. But considering adolescent age group of acne patients, the major age group in which this disease occurs belongs to *Sukumara* category (tender nature) and *Siravedha* being a *Shastra Karma* (surgical method) is not accepted easily. *Vamana* Karma also is an exhaustive and time consuming procedure due to its pre and post therapeutics implementations. Acharya Susruta stated *Jalaukawacharana* is the preferred method of bloodletting in *Sukumara* (tender nature).¹³ Therefore for *Raktamokshana*, *Jalaukawacharana* (leech therapy) can be more suitable *Shodhana* procedure as compared to *Vamana* and *Siravedha* in acne patients. *Jalaukawacharana* is much safer, less complicated and an almost painless procedure.

Ayurveda believes in treating the root cause by *Shodhanachikitsa*. *Raktamokshana* removes the vitiated blood and clears the *Strotovarodha* (blockage in channels), thus breaking the *Samprapti* (pathogenesis). *Jalaukawacharana* being a bio-purificatory method removes deep seated toxins by letting out blood, clearing *Srotasa*(channels) and pacifying vitiated *Doṣha*. *Jalaukawacharana* possesses high efficacy in both *Shodhan* (cleansing) and *Ropana* (healing), without producing any adverse effects. It is cost effective and easy to apply. Hence, it can be used as an alternative approach for management of acne. Therefore, *Jalaukawacharana* being the easiest and almost painless method can be used as a preferred way of *Raktamokshana* in acne.

Hirudotherapy; The treatment of disease conditions with medicinal leeches (*Jalaukawacharana*) is termed as Hirudotherapy, a Latin term.¹⁴ Hirudotherapy (Leech

therapy) is one of the oldest practices in medicine; In July 2004, the FDA approved leeches as a medical device in the area of plastic and reconstructive surgery.¹⁵ Hirudotherapy is used in dermatology for treating diseases of the epidermis, such as psoriasis, lichen planus and scleroderma. Local oxygenation was increased in patients receiving leech therapy for these disorders. In patients with lichen planus derma, decreased inflammation and lesion number found with hirudotherapy.¹⁶

Many studies have found that leeches have various bioactive molecules in their secretions. More than 20 molecules and their modes of action have been identified. The components of these secretions are thought to play important role in breaking pathogenesis of acne. Destabilase is a component in leech secretions which has glycosidase activity and shows both antibacterial and fibrinolytic actions.¹⁷ Bdellin is an enzyme that inhibits activities of trypsin, chymotrypsin, and plasmin, it can be used as plasmin inhibitor to control bleeding.¹⁸ It also have anti-inflammatory role. Histamine by its vasodilatation property improves microcirculation replacing old stagnant blood with fresh blood. Hyaluronidase which is also known as spreading factor facilitates the penetration and diffusion of pharmacologically active substances into the tissues and has antibiotic properties.^{19,20} Fibrinases and collagenase in saliva reduces the density of scar tissue and it helps to reduce fibroblast formation in scars.²¹

Procedure of *Jalaukawacharana*¹³

Leech is kept in solution of turmeric for a 45 min till they regained their motility. This Leech is applied on roughened desired spot. If Leech refuses to stick on the desired spot, then drop of milk is applied on the spot. On sticking to the affected part from the suckers, Leech assumes the shape of a horseshoe in the raised and arched position. Leech then is covered with a piece of thin and wet cotton and drops of water were frequently dropped on cotton piece. An attachment period lasts 20 to 45 minutes, during which the leech sucks between 5 and 15 ml of blood.²⁷ Generally, after 30-40 minutes, leech automatically detaches from the site. Turmeric powder is then sprinkled over the leech's mouth for inducing vomiting. Sometimes gentle squeezing of the leech is required from its posterior end toward anterior end to expel out the sucked blood. After satisfied vomiting of sucked blood, leech becomes active again and stored in freshwater.

WHY THIS REVIEW

In this modern era, beauty is the soul of physical appearance. Acne is affecting large population of world at the age, of adolescence, when they are most conscious about their looks. Treating acne with antibiotics and modern treatment sometimes is not enough. In Ayurveda literatures, various treatments are available for acne, but still not authenticated scientifically. In this review, an attempt has been made for reviewing the potentials of leech therapy in treating *Mukhadushika* w.r.t. acne vulgaris. For this purpose, various researches of *Jalaukawacharana* carried out at different Ayurveda centers were evaluated for their acne treating potential. *Raktamokshana* through leech therapy in acne needs to explore with more studies as a monotherapy or in combination with other therapy or drugs. This review helps new researcher to select their topic of interest about leech therapy in acne.

MATERIALS AND METHODS

In this review work, the clinical studies carried out at different Ayurved centres and published in the indexed and referred (ISSN numbered journals) were included. The research articles and case reports were searched in Google, PubMed, DHARA online and AYUSHPORTAL (A research portal of Govt. of India.) in the month of February 2020. In these review keywords *Mukhadushika*, *tarunyapidika*, acne, *Jalaukawacharana*, leech therapy were used for searching the research data on acne.

RESULT AND DISCUSSION

Previous Research Work

1. Role of *Jalaukawacharana* in *Mukhadushika* (Acne Vulgaris)²²

In this clinical study 30 patients of 16-30 years, ages were taken and treated with *Jalaukawacharana* weekly once for four weeks. Here author concluded that anticoagulant, anti-inflammatory, analgesic property of leech saliva helps in relieving the symptoms such as inflammation, pain, papules, pustules, comedones, nodules etc. Results were highly significant and encouraging and *Jalaukawacharana* proved to be an easy and less invasive treatment without causing any adverse effect.

2. A clinical study to evaluate the efficacy of *Jalaukawacharana* and *Siddarthakadi Lepa* in the management of *Mukhadushika* w.s.r. acne vulgaris²³

In the study, 15 patients of Acne Vulgaris were treated with *Jalaukawacharana* of four sittings with seven days of interval. *Siddarthakadi Lepa* was also applied daily in between four sittings of *Jalaukawacharana*. Assessment was done on the basis of subjective parameters i.e. *Pidika* (Acne lesions) grading and associated complaint grading, as well as objective parameters i.e. count of lesions. Intervention was found to be significantly effective ($p < 0.05$) in reducing Acne grading as well as associated symptoms i.e. *Vedana* (Pain), *Daha* (Burning), *Paka* (Inflammation), *Kandu* (Itching), *Vaivarnyata* (Discolouration) and *Snigdhata* (Oily). Intervention also showed significant result in reducing count of lesions too. However, no significant effect was seen on the number of scars. After follow-up period, only 13.33% showed relapse in grading of acne.

3. Clinical study to evaluate the efficacy of *Jalaukawacharana* and *Sarivadyasava* in *Yuvanpidika* (acne Vulgaris) ²⁴

A study by Rathod Motilal *et al* in 2012, total no. of 20 patients of acne were subjected to *Raktamokshana* with *Jalauka* weekly once and *Sarivadyasava* in dose of 25 ml three time a day orally for a period of 28 days, established the efficacy of leeching in acne with a $p < 0.001$ in symptoms of Size, Tenderness, swelling and severity of lesions, which shows very significant results. 50% patients showed complete relief and 40% patients showed moderate relief in symptoms.

4. Role of *Jalaukawacharana* and *Mahamanjishtadi Kwatha* in the Management Of *Mukhadushika* w.s.r to Acne Vulgaris- A Case Study.²⁵

Author selected a case of 25-year-old female acne patient; she was given four sittings of *Jalaukawacharana* with 7day interval along with the 10 ml *Mahamanjishtadi Kwatha* internally after meal continuously for 28 days. This remedy was given for 3 months. During complete duration of treatment, patient was instructed to follow *Pathya* and *Apthya* as advised strictly. After four setting of *Jalaukawacharana* there was complete disappearance of acne, complete relief in itching, burning sensation and pain. Significant improvement was observed in discoloration over face from grade 3 to 1.

5. The efficacy of *Jalaukawacharana* (leech therapy) in *Mukhadushika* (acne) ²⁶

A study by Asutkar *et al* in 2015 established the efficacy of leech therapy in acne with a $p < 0.05$ and a reduction in symptoms of Size, Tenderness, Redness which was significant at

5% significance. It was a clinical study and leech application was done every fourth day for total five settings. Out of 30, 29 patients got satisfactory results,

6. Efficacy of *Jalaukawacharana* in the Management of *Yuvanapidaka* w.s.r. to Acne Vulgaris- A Pilot Study.²⁷

In this study, a total 17 patients of age group between 14-40 years were registered out of which 15 patients completed the whole trial. Statistically Extremely significant results ($p < 0.001$) were found in tenderness (68.91%), Itching (77.77%), burning (55.96%), size (82.11%), extent of lesion (72.42%), significant result ($p < 0.05$) was found in discoloration (91.66%). Statistically nonsignificant results ($p > 0.05$) was found in discharge (81.82%). Author concluded that *Jalaukawacharana* alone is very significant in relieving the associated complaints of acne like itching, burning, pain due to *Sroto Shodhana* effect, due to improving local blood circulation, due to various bioactive substances released by leech.

7. Management of *Mukhadushika* (Acne vulgaris) with *Jalaukawacharana* (Leech Therapy): A Case Study²⁸

A 20 year's old female patient of acne on both cheeks associated with itching, burning sensation for 2 years. Three sittings of *Jalaukawacharana* were done (on Day 1, Day 5, and Day 10). Results were encouraging with its simple methodology achieved by *Sthanik Shodhana* (local cleansing effect).

8. Leech therapy in Dermatology,²⁹

In this paper, author concluded that skin disorders primarily of autoimmune kind make the dermis thick with poor tissue perfusion. The leech saliva prevents inflammatory mediators, decreases tissue thickness, accelerates reperfusion and increases permeability of the tissue making it more acceptable to therapeutics. Hence making leech therapy an ideal mode of treatment in dermatology.

The prevalence of acne vulgaris globally was 681.2 million in 2016. This was an increase of 10 percent from 612 million in 2006. By the end of 2026, the number of people affected by acne in India is estimated to reach 23 million at a compound annual growth rate (compound average growth rate) of 0.5 percent.³⁰

In Ayurveda texts, acne has been described as one of the *Kshudra Rogas* (minor ailments) caused by the vitiation of *Vayu*, *Kapha* and *Rakta* in the pathology of the disease. Ayurveda believes in expelling the root causes of acne by advising *Shodhana Chikitsa*. *Raktamokshana* among the *Shodhana* may give better relief particularly when *Rakta* is vitiated. *Raktamokshana* not only purifies the channels of the area of application but also lets the other parts of the body become free from disease and action is fast than other shaman remedies. As in *Yuvanapidaka* vitiated *Dosha*, *Dhatu* and *Mala* (basic bio-elements) gets accumulated in *Srotasa* (channels), causing blockages and leads to *Pidaka* formation. *Jalaukawacharana* being a bio-purificatory method removes deeply seated toxins by letting out blood, clearing *Srotasa* and pacifying vitiated *Dosha*. Among the *Anushastras* Vagbhatta considers *Jalaukas* (leeches) as the best as all places of the body for all the persons. Leech application not only removes blood from the site but also injects biologically active substances which help to manage various ailments. Leech's saliva contains a complex mixture of different biologically and pharmacologically active substances which gets secreted into the wound. So *Jalaukawacharana* has been proved for normalization and improvement of capillary as well as collateral blood circulation, Expressed anti-inflammatory effect, Analgesia and anesthesia effect through saliva, Immuno-stimulation and immuno-modulating effect.

Complications of Hirudotherapy³¹

Excessive bleeding can occur with leech therapy; it can be controlled by applying direct pressure or topical thrombin. These properties permit the wound to ooze up to 50 ml of blood for up to 48 hours. Leech bites usually bleed for an average of six hours. Another serious complication of leech therapy is infection.

CONCLUSION

By thorough review of article related to use of *Jalaukawacharana* (leech therapy) in *Mukhadushika* (acne) it can be concluded that *Jalaukawacharana* as a monotherapy and combination with other Ayurveda *shamana* drugs gives satisfactory results and for its validation, RCTs on more number of patients is required.

REFERENCES

1. Vagbhatta, *AṣṭangaHṛudaya*, with the commentaries, Edited by Pt. Hari Sadashiva Shastri- Paradakara Bhisagacharya, Published by Chaukhamba Surbharati Prakashana, Varanasi, 2007, Uttara tantra, 31/5, Page 888

2. Agnivesha, Charaka, Charaka Samhita, Chakrapani Ayurved Dipika Vyakhya, edited by Dr. Lakshmidhara Divedi, reprint ed. Chaukambha Krishnadas Academy, Varanasi, 2013; Sutra Sthana, TrishothiyaAdhyaya, 18/24, page375.
3. Gollnick HP. From new findings in acne pathogenesis to new approaches in treatment. *J Eur Acad Dermatol Venereol* 2015;29 Suppl 5:1-7.
4. American Academy of Dermatology. Skin conditions by the numbers. March 26, 2019, Available at <https://www.aad.org/media/stats/conditions/skin-conditions-by-the-numbers>.
5. Kellett SC, Gawkrödger DJ. The psychological and emotional impact of acne and the effect of treatment with isotretinoin. *Br J Dermatol*. 1999 Feb. 140(2):273-82.
6. Thomas P. Habif, James G.H. Dinulos, M Shane Chapman, Kathrin A Zug, Skin disease: diagnosis and treatment, Elsevierinc. First South Asia edition, 2011, page101
7. Jaggi Rao, Medscape, Oct 23, 2019, Available from: <https://emedicine.medscape.com/article/1069804-overview>
8. Sushruta, Sushruta Samhita with “NibandhaSangraha” commentary by Dalhanacharya, and the NyayacandrikaPañjika of Sri Gayadasa Acharya, edited by Vd. Yadavaji Trikamaji Acharya and Narayan Rama Acharya, Eighth edition, Published by Chaukhamba Orientalia, Varanasi, (2005); Nidanasthāna 13/39 Pg. no. 323
9. Sharma H, Bhishgacharya S. Kashyapa Samhita of Vruddhajivaka. Varanasi: Chaukhamba Sanskrit Sansthan; 2012. pp. 79.
10. Sharangadhara, Sharangadhara Samhita with “Dipika” hindi commentary by Addhamala, edited by Dr. Brahmanand Tripathi, ChaukhambaSurbharatiPrakashana, Varanasi, 221001, Pūrva Khanda 5/14
11. Sachin Anil Upasani et al. Review on Yuvanpidika Vis-A-Vis Acne VulgarisIjppr.Human, 2016; Vol. 5 (3): 77-94.
12. Vāgbhatta, Aṣṭāṅga Hṛudaya, with the commentaries, Sarvaṅgasundarā of ArundattaArundatta and Āyurvedarasāyana of Hemādri, Edited by Pt. Hari SadashivaShastri-Paradakara Bhisagacharya, Published by Chaukhamba Surbharati Prakashana, Varanasi, 2007, Uttara tantra, 32/3, Page 890
13. Susruta, Susruta Samhita. with Nibandha Sangraha commentary by Vaidya Yadavaji Trikamaji Acharya, Reprint Chaukhamba Surbharati Prakashana, Varanasi, Sūtrasthāna, 2003; 13(3).
14. Whitaker IS, Rao J, Izadi D, Butler PE. Historical Article: Hirudo medicinalis: ancient origins of, and trends in the use of medicinal leeches throughout history. *Br J Oral Maxillofac Surg* 2004;42:133- 137
15. Whitaker IS, Izadi D, Oliver DW, Monteath G, Butler PE. Hirudo Medicinalis and the plastic surgeon. *Br J Plast Surg* 2004;57:348-353.
16. Glyova O. Modern Hirudotherapy -A Review. (Biotherapeutics, Education and Research Foundation). The (BeTER) LeTTER 2005;2:1-3,5.
17. Zavalova LL, Yudina TG, Artamonova II, Baskova IP. Antibacterial non-glycosidase activity of invertebrate destabilase-lysozyme and of its helical amphipathic peptides. *Chemotherapy* 2006; 52:158-160
18. Eldor A, Orevi M, Rigbi M. The role of the leech in medical therapeutics. *Blood Rev* 1996; 10: 201- 209.
19. Adams SL. The medicinal leech. *Ann Intern Med* 1988;109: 399-405.
20. A.H. Lone, T. Ahmad, M. Anwar, S. Habib, G. Sofi, H. Imam Leech therapy – a holistic approach of treatment in Unani (Greek-Arab) medicine *Anc Sci Life*, 31 (2011), pp. 31-35
21. Grumbine NA, Nicholas A. Feature: reviving an ancient therapy to manage chronic pain. *Podiatry Today*. 2003; 16:46-53.
22. Harmeet K. Gupta S., Role of Jalaukawacharana in Mukhadushika(Acne Vulgaris)] *IJAAR* Jan-Feb 2018 Vol:3, Issue:6, Page No:1045-1052
23. Pandey K.K., Tripathi A. K., Srivastava A.K. A clinical study to evaluate the efficacy of Jalaukawacharana and Siddarthakadilepa in the management of Mukhadusika w.s.r. to acne vulgaris- *International Journal of Herbal Medicine* 2017; 5(6): 22-29
24. Rathod M., Kamath S., Clinical study to evaluate the efficacy of Jalaukawacharana and Sarivadyasava in Yuvanpidika (Acne Vulgaris), *International Research Journal of Pharmacy*, 2012, 3(7):215-217
25. Dhalpe S.J., Chandurkar V.S., Role Of Jalaukawacharana and Mahamanjishtadi Kwath in the Management of Mukhadushika w.s.r to Acne Vulgaris- A Case Study. *International Journal of Research- Granthaalayah*, 6(8), 25-32. <https://doi.org/10.5281/zenodo.1403789>.

26. Gujjanwar S., Dwivedi S.K., Asutkar S.G. The efficacy of Jalaukawacharana (leech therapy) in Mukhdushika (acne) Asian Journal of Multidisciplinary Sciences. 2015; 3(6):213-218
27. Kumar V., Efficacy of Jalaukawacharana In The Management Of Yuvanapidaka W.S.R. To Acne Vulgaris- A Pilot Study. International Ayurvedic Medical Journal {online} 2016 {cited 2016 July} Available from: http://www.iamj.in/posts/images/upload/2933_2938.pdf
28. Samaranavake G.V.P., Peiris K. P. P., Pushpakumara A. A.J., Management of Mukhadushika (Acne vulgaris) with Jalaukawacharana (Leech Therapy): A Case Study, International Symposium on Ayurveda and Indigenous Medicine 2018, Kcc, Kandy, Sri Lanka28
29. Jahangir U., Akram M., Shaista U. Leech therapy in Dermatology. Journal of Pharmacognosy and Phytochemistry 2016; 5(3): 214-216
30. Reportlinker, December 2017, New York Available from: <https://www.reportlinker.com/p05251482>
31. Abdullah S., Mohammad Dar L., Rashid Wani A., Tiwari A. Hirudotherapy /Leech therapy: Applications and Indications in Surgery Arch Clin Exp Surg Year 2012, Vol:1, Issue:3, P.172-180

