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Leeches and Their Use in Medicine (Annellida: Hirudinea: Rhynchobdelliformes)

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Abstract: Leech therapy relieves pain and inflammation, and there are explanations for its success. In leech saliva, in addition to substances that prevent blood clotting, there are also analgesic and anesthetic compounds that reduce pain. This article aims to study the characteristics of the leech (Annellida: Hirudinea: Rhynchobdelliformes) and its use in Medicine. This is a narrative review of the literature, which is indicated to explain and discuss a certain subject from a theoretical or contextual perspective, to allow the reader to ask questions and update knowledge on a specific topic. The search for scientific articles that made up this review was carried out on Google Scholar, Biological Abstract, HAL, Qeios, ResearchGate, Scielo, and SSRN. The following descriptors in Health Sciences (DeCS) were used: biological therapy, larva, wound, debridement, and healing. The following inclusion criteria were considered: original articles and reviews, published nationally and internationally in full, available electronically, and published in Portuguese, English, and Spanish. The exclusion criteria were dissertations, theses, monographs, and conclusion work, as well as duplicates and those that require payment to access the content during integration.

Keywords: Debridement, Medicine, Pain, Surgery, Therapy.

RESEARCH PAPER

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1. INTRODUCTION

The leeches are animals belonging to the Phylum Annellida, class Hirudinea, order Rhynchobdelliformes, they live in humid environments or fresh water. They are well known because some species are temporary ectoparasites of other animals. Some species of leeches found in the tropics are arboreal. In general, leeches live in freshwater, but marine species and others live in damp clay (Figure 1) (Adams, 1988; Abramczyk, 2005; Bordoni, 2005).

Figure 1: Leeches need blood to grow and reproduce
Sources: Pixabay and https://theconversation.com/curious-kids-why-do-leeches-suck-our-blood-117316

1.1. Objective

This article aims to study the characteristics of the leech (Annellida: Hirudinea: Rhynchobdelliformes) and its use in Medicine.

2. METHODS

This is a narrative review of the literature, which is indicated to explain and discuss a certain subject from a theoretical or contextual perspective, to allow the

reader to ask questions and update knowledge on a specific topic. The search for scientific articles that made up this review was carried out on Google Scholar, Biological Abstract, HAL, Qeios, ResearchGate, Scielo, and SSRN. The following descriptors in Health Sciences (DeCS) were used: biological therapy, larva, wound, debridement, and healing. The following inclusion criteria were considered: original articles and reviews, published nationally and internationally in full, available electronically, and published in Portuguese, English, and Spanish. The exclusion criteria were dissertations, theses, monographs, and conclusion work, as well as duplicates and those that require payment to access the content during integration.

3. MORPHOLOGY

Its length varies from 6 to 10 cm; The largest leech in the world is *Haementeria ghilianii* Filippi, 1849 (Rhynchobdelliformes: Glossiphoniidae) from the Amazon, which measures 30 cm. The most common colors are black, brown, olive green, and red. Gas exchange is carried out over the entire general surface of the body; A type of hemoglobin can be found in the coelomic fluid of some species. Excretion occurs through the nephridia deep within the connective tissue. They have a cylindrical body covered with a cuticle and, unlike other annelids, they do not have bristles. Leeches are coelomates and their coelom is divided into segments or metamers for a total of 34 ringed segments on their body. The clitellum through which mucus is secreted, a structure, is not visible except during the breeding season (Figure 2) (Budzynski *et al.*, 1981; Adams, 1988; Fields, 1991; Bush *et al.*, 2001; Bordoni, 2005; Brusca *et al.*, 2007; Munshi *et al.*, 2008; Kutschera and Elliott, 2014; BBC News Brazil, 2016; Yapici *et al.*, 2017; Babenko, 2020; Jueg and Zettler, 2022; Pacievitch, 2024; Rutkauskaite-Suciliene, 2024).

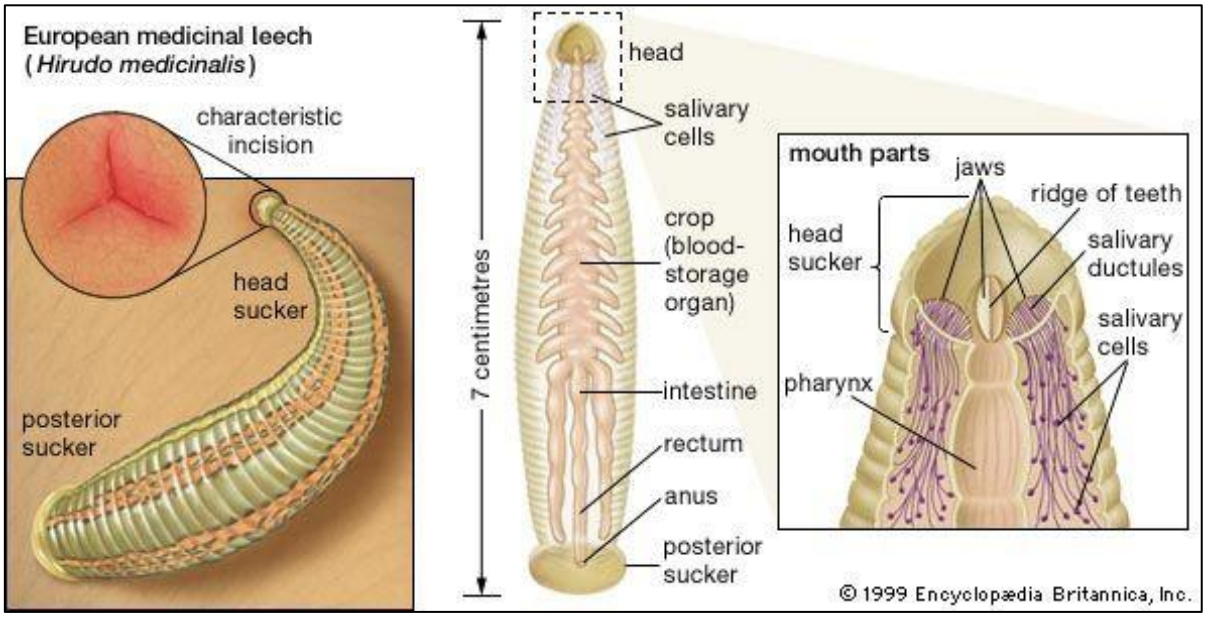

Figure 2: Anatomy of the European medicinal leech After attaching its head sucker to the skin, the European medicinal leech *Hirudo medicinalis* L., 1758, uses its three jaws with razor-sharp teeth to make a neat Y-shaped cut. Salivary ductules between the teeth secrete several pharmacologically active substances, including a local anesthetic and the potent anticoagulant hirudin

Source: https://www.britannica.com/animal/Protostomia

4. PARASITISM

Leeches attach themselves to the animal to be parasitized through their suction cups that pierce its skin. Digestion is extracellular through a complete digestive tract. Its muscular pharynx sucks blood, which will be stored in a crop. Generally, there is no pain in this process, and the presence of the salivary enzyme hirudin prevents blood clotting. In the morphology of these animals, it is common to find two suckers: A posterior one, for locomotion, and an anterior one, to suck the blood of parasitized beings. Digestive system and feeding in leeches: They have jaws and three blades placed at an angle to each other, they cut through the skin of their hosts and contain a Y-shape behind the blades there is a mouth located ventrally on the anterior end of the body, then there is pharynx, esophagus, crop, gizzard and finally intestine. They can be scavengers, predators, and parasites (Figure 3) (Amaral and Migotto, 1980; Budzynski *et al.*, 1981; Adams, 1988; Fields, 1991; Mory *et al.*, 2000; Bush *et al.*, 2001; Bordoni, 2005; Brusca *et al.*, 2007; Munshi *et al.*, 2008; Kutschera and Elliott, 2014; News Brazil, 2016; Yapici *et al.*, 2017;

Babenko, 2020; Jueg and Zettler, 2022; Pacievitch, 2024; Rutkauskaite-Suciliene, 2024).

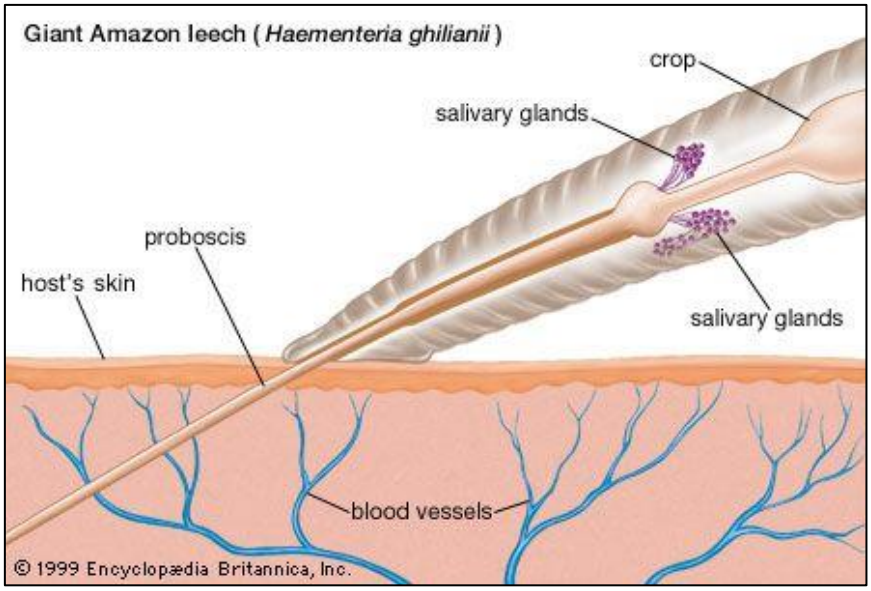

Figure 3: Head of the giant Amazon leech *Haementeria ghilianii* Filippi, 1849. The retractable proboscis is used to pierce the skin and suck blood from the host
Source: https://www.britannica.com/animal/Protostomia

Leeches are bloodsuckers, they are divided into two large groups: Those that pierce the host's skin with the help of their jaws to obtain blood, and leeches that introduce a structure that we could compare to a proboscis or a straw, into the pores of the skin sucking blood directly from a blood vessel. They feed mainly on vertebrates, such as cattle, horses, reptiles, fish, humans, and other species. With large suckers on the back of the body and small suckers on the front of the body, near the mouth, leeches move in palm-sized movements, that is, they shrink and crawl (Figure 4) (Amaral and Migotto, 1980; Budzynski *et al.*, 1981; Adams, 1988; Bush *et al.*, 2001; Bordoni, 2005; Brusca *et al.*, 2007; Munshi *et al.*, 2008; Kutschera and Elliott, 2014; Yapici *et al.*, 2017; Babenko, 2020; Jueg and Zettler, 2022).

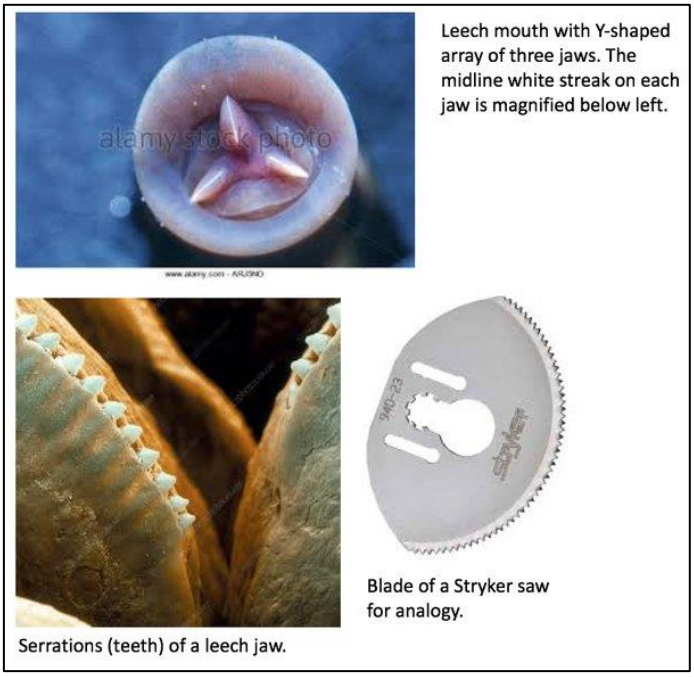

Figure 4: A leech mouth has three jaws, arranged in a Y pattern. Each jaw is shaped somewhat like a wedge of a circular saw blade, with teeth corresponding to the saw teeth. These jaws don't rotate in a full circle like a circular saw, but rock back and forth, sawing away somewhat like a serrated steak knife, to lacerate the skin. Stryker saws for cutting the bone and cutting casts off patients operate similarly—oscillating rather than spinning
Source: https://www.quora.com/Do-leeches-have-300-teeth

5. RESPIRATION AND REPRODUCTION

Respiration is cutaneous and carried out by a network of capillaries below the epidermis. However, some species have branchiae. They are protrandric hermaphrodites, the testes mature before the eggs. They have 32 brains, nine pairs of testicles, and a jaw with three rows of one hundred teeth each. The cocoon receives the eggs as it passes through the female gonopore. Fertilization is internal. Sperm transfer occurs between gonopore (Figure 5) (Adams, 1988; Mory *et al.*, 2000; Bordoni, 2005; Brusca *et al.*, 2007; Munshi *et al.*, 2008; Kutschera and Elliott, 2014; Jueg and Zettler, 2022).

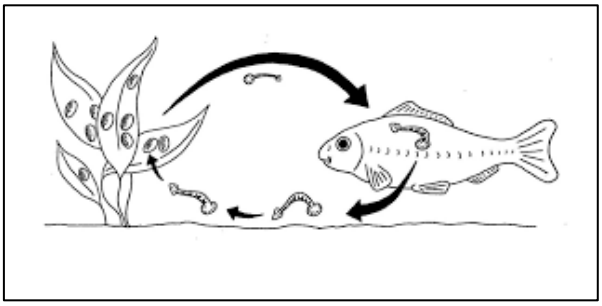

Figure 5: The life cycle of a leech
Source: chrome-extension://efaidnbmnnnibpcajpcglclefindmkaj/https://ifm.org.uk/wp-content/uploads/2021/08/Leeches.pdf

6. DISEASE TRANSMISSION

In Iraq, they consider vectors of protozoa such as *Trypanosoma* Gruby, 1843, and *Haemogregarina* Danilewsky, 1885, in fish, amphibians, reptiles, cattle, man, and other domestic animals to cause anemia for them, they can serve as hosts, both intermediate and final, they demonstrate both pathogenic and beneficial effect on humans and animals. Leeches are capable of transmitting *Trypanosoma evansi* Evans, 1880 (Trypanosomatida: Trypanosomatidae), a parasite of horses that causes great damage to livestock in the Pantanal region of Mato Grosso. *Trypanosoma evansi* is the protozoan that causes a disease also known as surra, endemic to the Pantanal and other swampy areas. Leeches are mechanical vectors of *T. evansi*. In this form of transmission, unlike biological transmission, the parasite does not use the vector's cellular machinery to complete part of its life cycle. But as the trypanosome survives a few hours in the vector's mouthparts, it is capable of transmitting it from one animal to another the penis. In some, without a penis, hypodermic impregnation occurs (Figure 6) (Adams, 1988; Nehal, 1994; Mory *et al.*, 2000; Bush *et al.*, 2001; Abramczyk, 2005; Bordoni, 2005; Munshi *et al.*, 2008; Alharbi, 2015; Giordani *et al.*, 2016; Yapici *et al.*, 2017; Aregawi, 2019; Zichao *et al.*, 2019; Kim *et al.*, 2022; Jueg and Zettler, 2022; Fapesp, 2024).

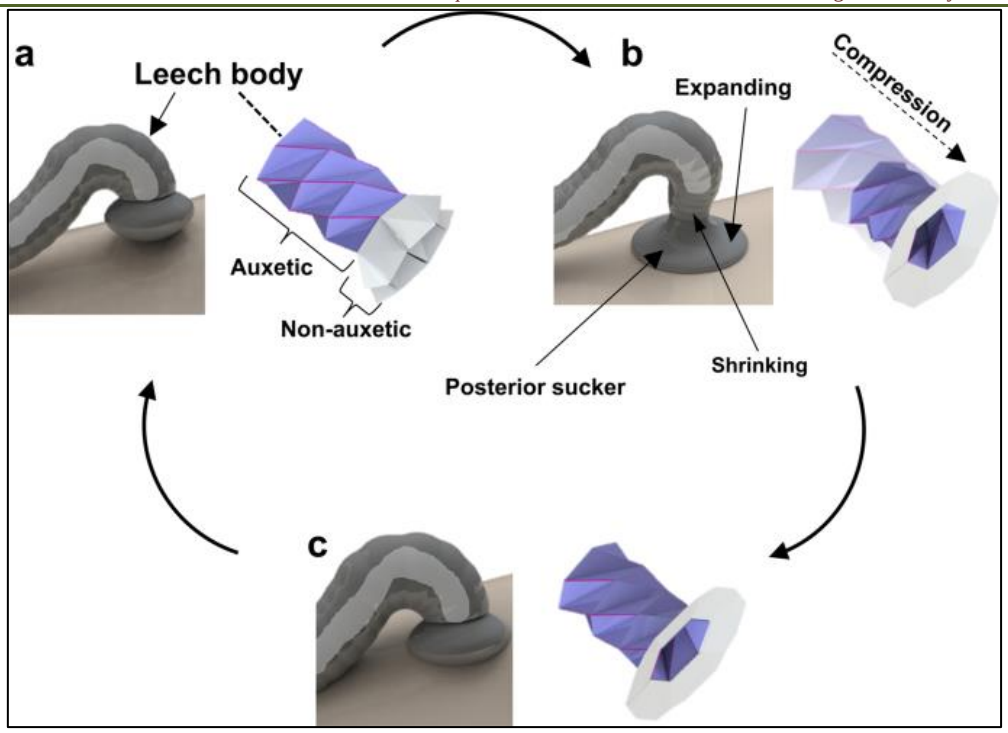

Figure 6: Schematics of the sticking mechanism of the posterior leech sucker. (a) Before the attachment, the shrunk sucker is positioned to target the skin surface. (b|) the sucker part of the leech is stretched out along the radial direction and shrunken the body to attach to the skin. (c) the leech's body expands for the pumping to the victim's blood. The body trunk of the leech was mimicked by auxetic design, and the sucker was mimicked by non-auxetic origami design. The 3D origami composed of a non-auxetic tip and the auxetic trunk is proposed in this study

Source: https://doi.org/10.1038/s41528-022-00139-x

7. THERAPY AND DRUG PRODUCTION

The BioPharm laboratory in Wales breeds tens of thousands of these dark, slug-like annelids for use in hospitals in various parts of the world. This is its role as a fundamental tool in contemporary reconstructive surgery. Therapy with these animals is used to relieve pain and inflammation, and there are explanations for its success. In leech saliva, in addition to substances that prevent blood clotting, there are also analgesic and anesthetic compounds that reduce pain. The technique was also used in cases of toothache and ear infections (Budzynski *et al.*, 1981; Carter, 2001; Eroglu *et al.*, 2001; Fort, 2001; Abramczyk, 2005; Bordoni, 2005; Koh and Kini, 2009; Amorim, 2014; Lauche *et al.*, 2014; Alharbi, 2015; Mattos, 2018; Babenko, 2020; Iwama *et al.*, 2021; Iwama *et al.*, 2022; Kim *et al.*, 2022; Pacievitch, 2024).

In Europe and the United States, leeches are being used in plastic and reconstructive surgeries, as they can cause a small hemorrhage that imitates venous circulation, helping to reestablish blood circulation in the delicate area where the graft was applied. In this way, leeches are used to assist in the transplantation of fingers, ears, or any parts that have been seriously damaged in accidents. These annelids effectively combat gangrene, decongest blood vessels remove excess blood, and restore normal blood pressure and circulation. Leeches have been proven to be effective against inflammatory problems such as arthritis they could cure everything from local pain (something proven) and inflammatory processes to obesity, gout, tumors, and mental disorders, Leeches can be used in skin treatment, and nephritis (Figure 7) (Eroglu *et al.*, 2001; Fort, 2001; Abramczyk, 2005; Bordoni, 2005; Koh and Kini, 2009; Lauche *et al.*, 2014; Alharbi, 2015; Yapici *et al.*, 2017; Mattos, 2018; Babenko, 2020; Pacievitch, 2024; Rutkauskaite-Suciliene, 2024).

Figure 7: Raktamokshana or bloodletting is one of the panchakarmas in Ayurveda. Bloodletting is of two types: Sashtrakrita using sharp instruments and Asashtrakrita without using sharp instruments. Leech therapy Jalookavacharana is one of the Asashtrakrita types of bloodletting. Jalouka leech has got its name since jal or water is its life. There are two main types of jalookas savisha poisonous and nirvisha. Nonpoisonous ones have specific characteristics, and they are carefully selected from freshwater ponds for leech therapy
Source: https://mauryaayurveda.com/leech-therapy-in-ayurveda/

Scientists have already identified several medicinal substances that this small invertebrate produces in its saliva every time it bites its host. Some of these substances, which are being studied, could become useful drugs for the treatment of cardiovascular diseases. The fact is that, to date, there is no tool as effective as these creatures to prevent blood from accumulating in the treated regions, reducing pressure on veins, and forming new blood connections. Leeches secrete peptides and proteins that work to prevent blood clots. This anticoagulant property keeps blood flowing to wounds to help them heal (Figure 8) (Adams, 1988; *Bush et al.*, 2001; Carter, 2001; Eroglu *et al.*, 2001; Fort, 2001; Bordoni, 2005; Koh and Kini, 2009; Amorim, 2014; Lauche *et al.*, 2014; Alharbi, 2015, Yapici *et al.*, 2017; Mattos, 2018; Babenko, 2020).

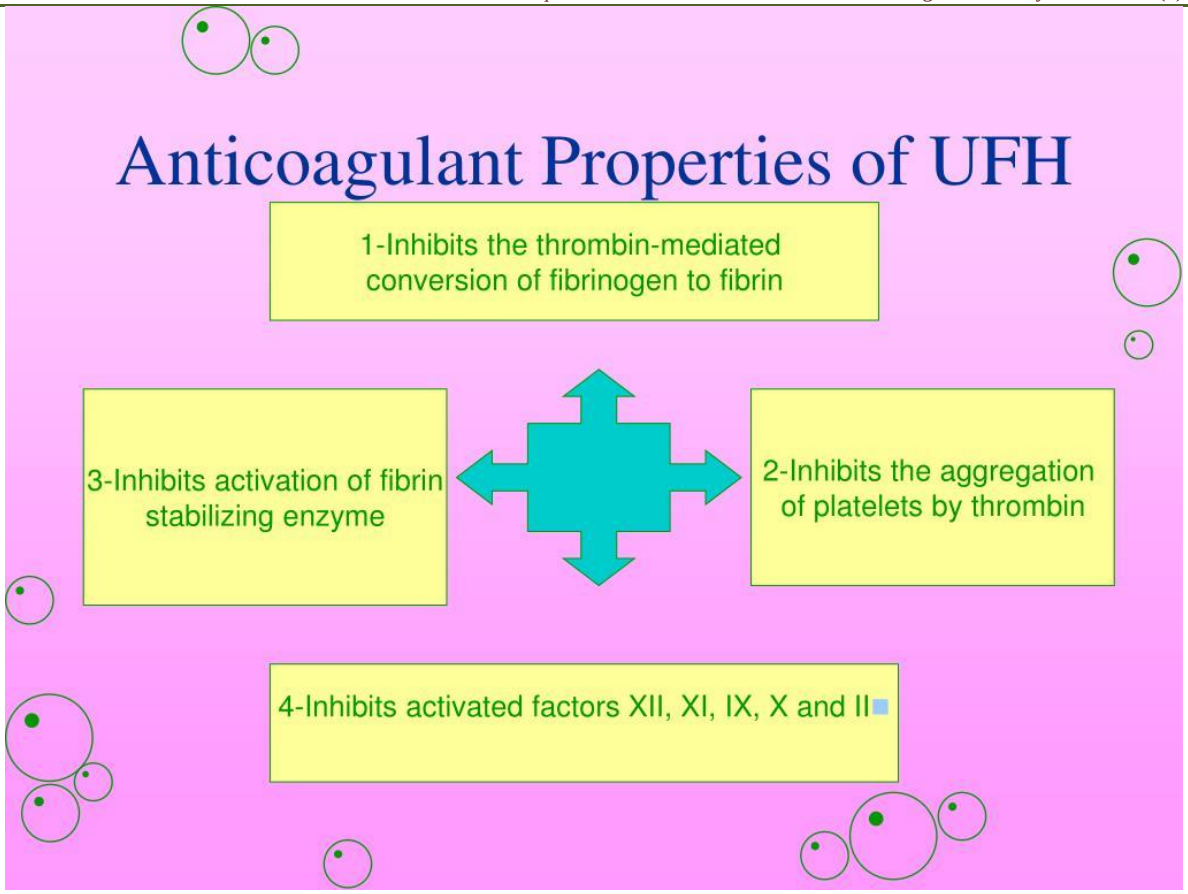

Figure 8: Anticoagulant Property
Source: https://www.slideserve.com/schuyler/parenteral-anticoagulant

Clinical trials suggest that leech therapy is a suitable treatment for the common joint disease. The anti-inflammatory and anesthetic properties in leech saliva reduce pain and tenderness at the affected joint site. On the other hand, people with heart disease can also use the method to improve inflammation and blood flow. The two bilateral pairs of *H. ghilianii* salivary glands are located at the base of the proboscis and encapsulated by a thin membrane. Both salivary glands of the leech contain an anticoagulant that not only inhibits the clotting of human plasma but also dissolves previously formed fibrin clots. This anticoagulant activity is attributable to an enzyme, for which the name hementin is proposed. Hementin catalyzes the proteolytic degradation of fibrinogen and fibrin, even in the presence of protease inhibitors that occur in human plasma (Bush *et al.*, 2001; Carter, 2001; Eroglu *et al.*, 2001; Fort, 2001; Bordoni, 2005; Koh and Kini, 2009; Amorim, 2014; Alharbi, 2015; Yapici *et al.*, 2017; Mattos, 2018; Babenko, 2020; Pacievitch, 2024; Rutkauskaite-Suciliene, 2024).

The hementin cleaves human fibrinogen to produce characteristic high molecular weight fragments. *Haementeria ghilianii* prevents its host's blood from clotting through a fibrinogenolytic mechanism that is completely different from that of hirudin, a thrombin-inactivating mechanism. polypeptide present in the saliva of another leech, *Hirudo medicinalis* L. 1758 (Acanthobdellida: Hirudinidae). Leech *H. ghilianii* has an anticoagulant in its salivary glands that renders ingested blood incoagulable by thrombin. The mechanism of blood in coagulation is associated with the cleavage of peptide bonds in fibrinogen, and thus the active agent, called hementin, is a proteolytic enzyme (Figure 9) (Nehal, 1994; Wallis *et al.*, 1997; Bush *et al.*, 2001; Carter, 2001; Eroglu *et al.*, 2001; Fort, 2001; Bordoni, 2005; Koh and Kini, 2009; Amorim, 2014; Alharbi, 2015; Yapici *et al.*, 2017; Mattos, 2018).

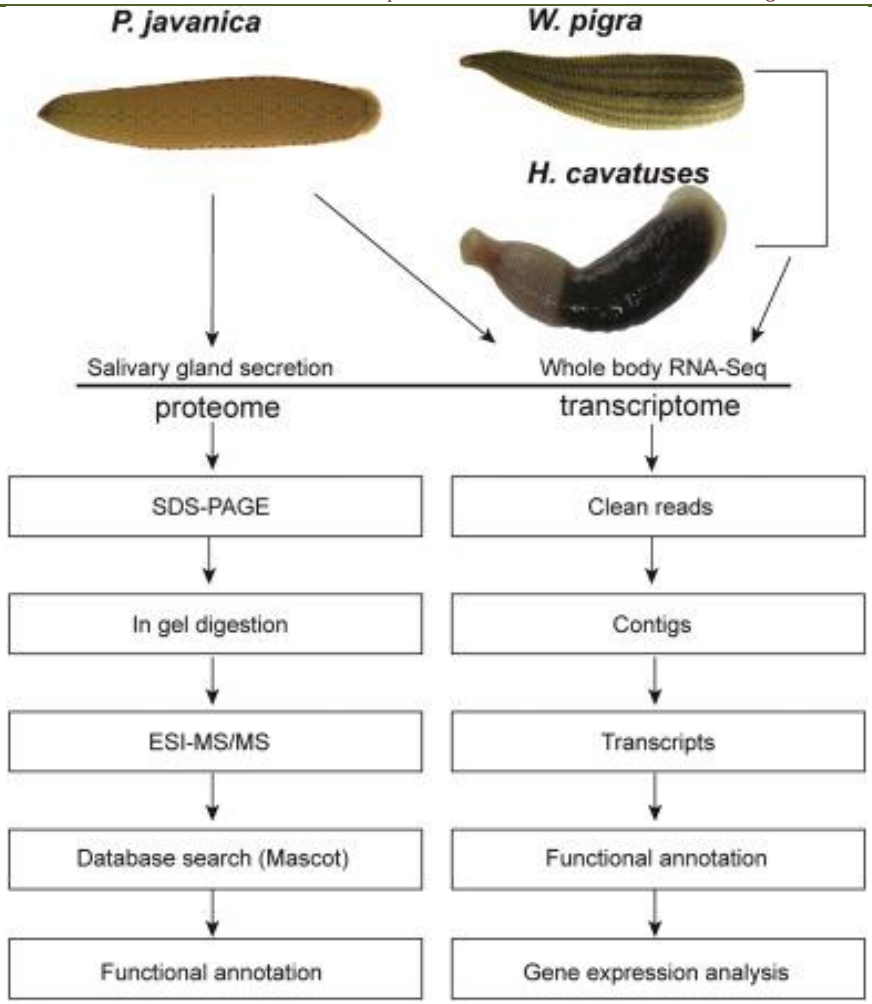

Figure 9: Medicinal leech therapy has been used for many centuries depending on the components of leech saliva secretions and active actions, but many components of the secretions were less known due to their low concentration and abundance. Determination of the profiles of leech salivary secretions is important to its medicinal application. Hereby, the molecular information provided by proteomic and transcriptomic analysis can be used to develop a more thorough understanding of leech sucking pathway and medicinal application. It provided a new foundation for performing novel investigations and discovering future pharmacological agents or targets in leech medicinal therapy

Source: https://doi.org/10.1016/j.jprot.2019.03.009

When a leech bites, it slowly sucks blood and injects compounds such as hirudin and calin present in its saliva that prevent blood from clotting. Leech saliva also contains histamine-like substances that dilate blood vessels and improve blood flow. Patients may develop skin infections in response to these treatments due to the *Aeromonas* bacteria, which lives in the guts of leeches and is found in their saliva (Bush *et al.*, 2001; Carter, 2001; Eroglu *et al.*, 2001; Fort, 2001; Bordoni, 2005; Koh and Kini, 2009; Mattos, 2018; Babenko, 2020).

The salivary glands produce an active substance called hementerin. The impediment of plasma coagulation by recalcification is evident even without prior incubation with hementerin. Hementerin destroys fibrinogen, as systems rendered incoagulable do not clot by the addition of botropase or thrombin. Hementerin has fibrinogeniolytic action, requiring a blood cofactor that exists in plasma and serum. Hementerin interferes with the formation of prothrombin and thromboplastin in plasma but does not prevent the formation of thrombin in platelet-rich plasma. Thrombin is not affected by the action of hementerin, unlike the effect of hirudin, which is antithrombin. The worm bites and injects a complex cocktail of proteins into the host through saliva. This may be responsible for the therapeutic effects (Eroglu *et al.*, 2001; Fort, 2001; Bordoni, 2005; Koh and Kini, 2009; Alharbi, 2015; Yapici *et al.*, 2017; Babenko, 2020; Iwama *et al.*, 2021).

In the issue of Nature magazine, Andreas Michalsen and Gustav Dobos, from the University of

Duisburg-Essen, in Germany, have carried out clinical tests to study the effects of leech on osteoarthritis, a degenerative condition that causes inflammation and pain and affects 20% of people with over 65 years old. The treatment is painless. When applying them, patients feel a slight pinch but then do not feel pain or any other unpleasant sensation. The German researchers now want more conclusive results that indicate the reasons for the effect observed. "Leeches not only take, but they also give," said Dobos. "As the worm bites, it injects a complex cocktail of proteins into the host via saliva. This may be responsible for the therapeutic effects" (Adams, 1988; Carter, 2001; Eroglu *et al.*, 2001; Alharbi, 2015; Fapesp, 2024; Pacievitch, 2024; Rutkauskaite-Suciliene, 2024).

What is known, according to the scientist, is that the worm's saliva contains at least one anti-inflammatory molecule, called a "leech-derived tryptase inhibitor." Another known molecule produced by the animal is hirudin, discovered more than a century ago. It is a protein that prevents blood from clotting. A synthetic form of hirudin is sold in the form of the drug Refludan and researchers believe that many other leech-derived medicines will be discovered in the future (Figure 10) (Eroglu *et al.*, 2001; Fort, 2001; Zaidi *et al.*, 2011; Alharbi, 2015; Babenko, 2020; Pacievitch, 2024).

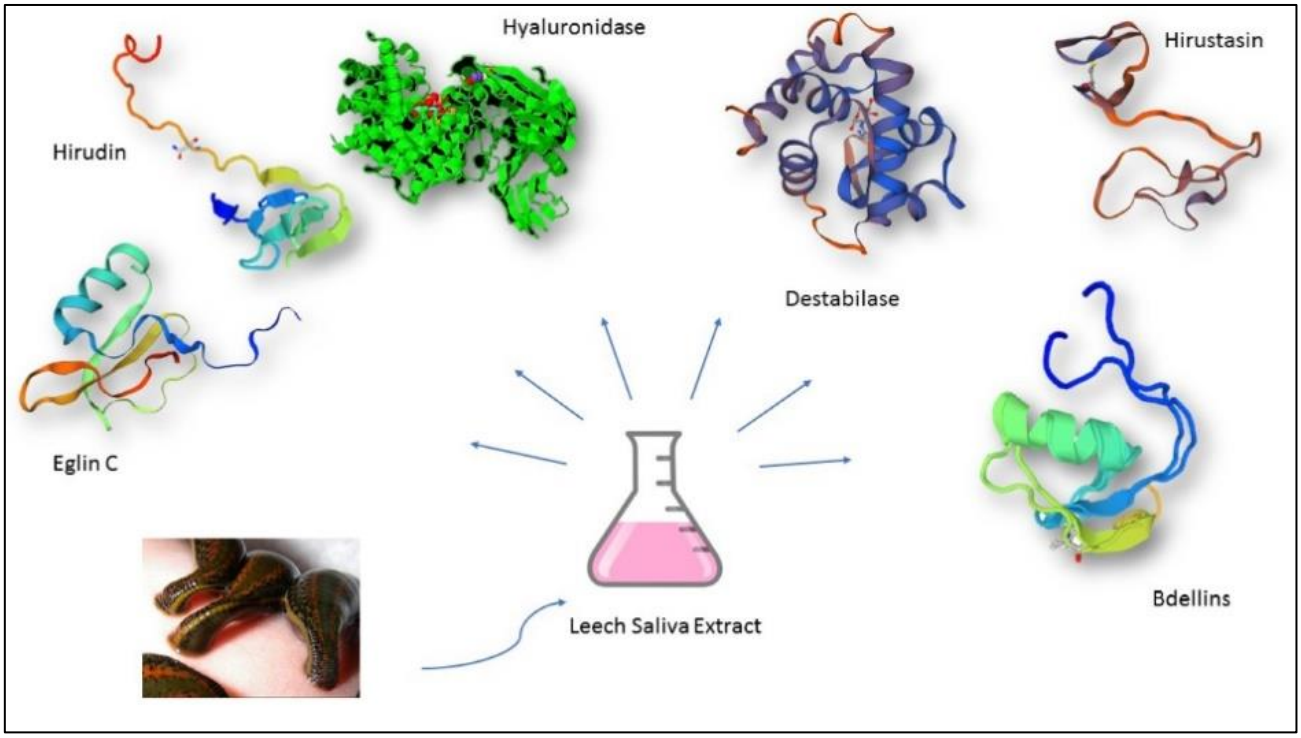

Figure 10: Medical leech saliva contains a wide range of proteins and enzymes that have multifunctional effects on diseases

Source: Advanced Pharmaceutical Bulletin. 11(2):261-266. doi: 10.34172/apb.2021.038

The old leech of medical treatments in centuries past, which left patients sicker because of unrestrained bleeding, is once again arousing medical interest. In the journal "Molecular and Cellular Proteomics", Yanes explains that peptides and proteins present in leech saliva prevent the development of the cascade of blood reactions that lead to clotting. The research identified a protease inhibitor serine that indicates an important path for the development and research of new anticoagulant drugs. Leech therapy can be used as part of the treatment of various health conditions, such as:

People who are at risk of limb amputation due to the side effects of diabetes; Individuals diagnosed with cardiovascular diseases; Plastic surgery patients who are at risk of losing some of their soft tissue in the postoperative period; Treatment of blood clots and varicose veins. Furthermore, it is believed that treatment with leeches can offer benefits in cases of Migraine; Atherosclerosis; Alzheimer's; Infertility; Hepatitis; Cystitis; Sinusitis; Glaucoma; Chronic renal failure; Eczema; Arthritis; Hypertension; Varicose veins; Hemorrhoids (Figure 11) (Budzynski *et al.*, 1981; Whitlock *et al.*,1983; Adams, 1988; Carter, 2001; Fort, 2001; Bordoni, 2005; Koh and Kini 2009; Yapici *et al.*, 2017; Aregawi, 2019; Zichao *et al.*, 2019; Babenko, 2020; Iwama *et al.*, 2022; Pacievitch, 2024).

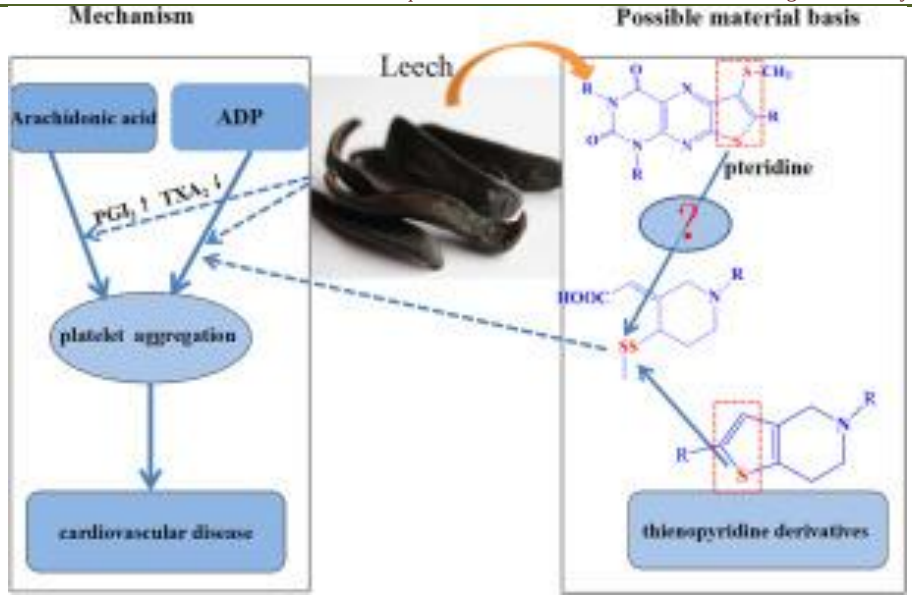

Figure 11: Where they were claimed to promote blood circulation and eliminate blood stasis. And have been used to prevent CVDs by exerting multiple effects when orally administered, one of which is the significant inhibition of platelet aggregation. Its ability to exert this effect has been extensively investigated in vivo and clinical practice
Source: https://doi.org/10.1016/j.jep.2018.12.010

According to research from 2004, there are several substances present in leech saliva, including around 60 different proteins that may be beneficial to human health. One study found that leeches can improve arterial function among the elderly. Additionally, other small research suggests that leeches' saliva may be used in preventing cancer metastasis and relieving cancer-related pain. Overall, leech treatment has few side effects. However, there is a risk of bacterial infection. Therefore, people immunocompromised by autoimmune diseases and environmental factors are not candidates for therapy (Bordoni, 2005; Koh and Kini, 2009; Alharbi, 2015; Aregawi, 2019; Zichao *et al.*, 2019; Babenko, 2020; Iwama *et al.*, 2021).

Conclusion

Leech therapy is used to relieve pain and inflammation, and there are explanations for its success. In leech saliva, in addition to substances that prevent blood clotting, there are also analgesic and anesthetic compounds that reduce pain.

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