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RESEARCH ARTICLE

Clinical and Laboratory Analysis of the Efficiency of Hirudotherapy in Complex Treatment of Endodontal Diseases

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Abstract

To date, teeth with inflammation in the periapical tissues, including destructive ones, often give non-abating exacerbations by conservative treatment, and they are removed. The article displays the clinical and laboratory results of a study of the effectiveness of hirudotherapy in combination therapy of inflammatory diseases of the endodontic. In recent years, a number of new drugs and methods for the treatment of chronic apical periodontitis have been proposed. The arsenal of new drugs for endodontic practice is extensive, but the result of treatment is often not optimistic. The purpose of the study was to conduct a clinical and laboratory analysis of the method of hirudotherapy to increase the effectiveness of treatment of chronic apical periodontitis. Based on clinical and laboratory data, the local course of the inflammatory process was studied, including cases when using hirudotherapy in patients with acute exacerbation of chronic apical periodontitis. Hirudotherapy allows one to stop inflammatory syndrome in earlier periods and reduce the time of treatment. The use of hirudotherapy in the combination with conventional endo therapy of patients with chronic apical periodontitis result in pronounced anti-inflammatory, analgesic, and vasoconstrictor effect that increases the effectiveness of treatment of chronic apical periodontitis.

Keywords: *Inflammation, Apical periodontitis, Hirudotherapy, Cytomorphology, Protocooperation.*

Introduction

A wealth of experience has been gained in the treatment of chronic apical periodontitis. Conservative, conservative surgical and surgical are some of the many methods used in the treatment of apical periodontitis. The choice of method and drugs used is determined by the etiology of chronic apical periodontitis and the clinical picture of the course of the disease.

Conservative treatment is indicated when there is sufficient opportunity to influence the inflammatory process in the periapical region through a root canal. Among alternative methods of treatment there are physiotherapeutic procedures (potassium iodide electrophoresis, copper-calcium depophoresis, etc.).

These procedures are sufficiently able to stop the inflammatory process in the periodontium, as well as stimulate reparative processes in bone tissue [1, 2].The number of contraindications, such as benign and malignant neoplasms, hypertension, pregnancy, cardiovascular insufficiency of 1-2 degrees, and, finally, intolerance to physiotherapy procedures limit or make the use of these methods impossible.

Conservative-surgical methods allow saving the entire tooth or part of it, to create conditions for the functioning of the tooth, followed by prosthetics. Surgical methods are indicated in cases of failure of conservative treatment. But in these cases, there is a

violation of the anatomical and functional integrity of the dentition, caused by turning off part of the teeth from the function [3, 4].Endodontic treatment is aimed at eliminating the inflammatory focus and qualitative obturation the canals in order to preserve the tooth and its functional ability.

The complex anatomy of the endodontic, the high virulence of microorganisms and the diversity of microflora often do not allow 100% to prevent its impact. For the treatment of chronic apical periodontitis in modern dentistry, various methods and medicines are used, the choice of which is primarily determined by both the etiology and the clinical picture of the disease. The methods and medicines used are not always sufficiently effective. Despite the quality endodontic treatment, there is often no complete restoration of bone tissue in the zone of destruction.

The use of antibiotics, immunomodulators, steroid and non-steroidal anti-inflammatory drugs, physiotherapy methods for the treatment of the chronic apical periodontitis is limited due to individual intolerance, concomitant, and allergic diseases. In this regard, the traditional treatment methods and, in particular, hirudotherapy is attracting attention of more and more dentists, because it has a systemic and multicomponent effect on the course of the inflammatory process, which is due to the presence of biologically active substances in the composition of the salivary gland secretion of the medicinal leech [5, 8].

Until recently the use of hirudotherapy and in dentistry, until recently, was minimal. Currently, there are reliable data on the effectiveness of hirudotherapy in various dental diseases [9, 11]. The secretion of the salivary glands of the medicinal leech includes hirudin.

This is a highly specific inhibitor of the enzyme thrombin, a direct-acting anticoagulant. The effect of blocking blood coagulation at the stage of fibrinogen into fibrin is associated with its action. According to Walsmann P. et al., 1981, the presence of hirudin slows thrombin activation of coagulation factors V, VIII, XIII. Hirudin prevents thrombosis, increases the total clotting time, and thrombin and thromboplastin times of blood plasma.

In addition, Wallis R., 1988, studies have shown that hirudin inhibits platelet release and aggregation by inhibiting the binding of thrombin to platelet receptor proteins, which distinguishes it from other thrombin inhibitors. Rapid penetration of the complex through the cell membrane, attachment to the damaged area of the vascular wall and to the wall thrombus, followed by lysis of the fibrin clot.

The destabilizing complex also interferes with thrombosis due to the blockade of thrombin, kallikrein (kininogenase) of plasma, platelet aggregation, and adhesion [12, 13, 14].Anti-inflammatory effect of hirudotherapy and provided by the presence in the medicinal leech secretions proteinase inhibitors - bdellins that inhibit trypsin, acrosin, plasmin, and Eglin, inhibitory alpha-chymotrypsin, chymosin, subtilisin and neutral protease of human granulocytes [15, 19].

Hirustasin, which has thrombolytic and anti inflammatory effects, was isolated in 1994. The composition of the secretion of the leech includes the highly active hyaluronidase. It provides the efficiency of diffusion of other biologically active substances of the secretion into the tissue, increasing their permeability, increasing tissue metabolism, and ensuring the anti-ischemic effect of hirudotherapy [20, 24].Despite the vast amount of materials and methods in modern endodontic practice, the effectiveness of endodontic treatment and tooth preservation has not been sufficiently solved. [24].

Improving the quality of treatment and preventing complications, both in the process of endodontic treatment and after, remains an urgent task. The goal of this study was to appreciate the effectiveness of hirudotherapy in complex treatment of chronic apical periodontitis with exacerbations and non-exacerbations based on dynamic observation with using clinical, laboratory, and functional methods research.

Materials and Methods

This work was done at Sechenov University with supported by the "Russian Academic Excellence Project 5-100". The material of the study was the data of examination and treatment of 49 patients with chronic apical periodontitis in the acute stage.

Divided into 2 groups: the main (25) and comparison (24). All research methods under this article have been conducted in accordance with the relevant guidelines and regulations. All experimental protocols were approved by the Local Ethics Committee of I. M. Sechenov First Moscow State Medical University (Sechenov University) protocol number № 10-16 from 09.11.2016. Prior to the study, informed consent was obtained from all patients for the upcoming study.

The control group consisted of patients (10) without the endodontic disease. The study was performed in double-blind. We used clinical, laboratory, and statistical methods. Clinical methods included data from anamnesis, general somatic pathology, hygienic condition of the oral cavity, periodontal status, and severity of the endodontic inflammatory process. The secretion of unstimulated mixed saliva in the morning hours on an empty stomach was determined by collecting into a measuring tube for 10 minutes.

The absolute number of cells in 1 ml of mixed saliva was counted. In smears stained by Romanovsky- Giemsa, the number of neutrophils and epithelial cells (per 300 cells) was counted; the data obtained were reduced to the sialogram. To determine the content of lysosomal-cationic proteins in neutrophils, smears of mixed saliva were stained with intense green.

In the combination therapy of patients with chronic apical periodontitis, medical leeches (Hirudo medicinal is) grown on the biofactory of JSC "Rosfarmaciya" M3 RF according to uniform standards. The use of the medicinal leech (MP) in the complex treatment of chronic apical periodontitis was carried out according to the method developed by the authors.

Before the start of the session, the hirudotherapy and the patient were acquainted with the upcoming procedure and received consent for treatment. After that, the medicinal leech was applied to the mucous membrane of the alveolar process in the projection area of the root of the causal tooth from the vestibular side, Figure 1. The aspiration method was used. The procedure lasted an average of 15-20 minutes, Figure 2.

A forced stop was not performed, since, as the leech quenches its hunger, it releases from the patient, as shown in Figure 3. Bleeding in patients stopped on its own, as in Figure 4. One procedure involved the application of 1-2 medical leeches. With exacerbation of the chronic apical periodontitis, 3-5 MPs were used for a treatment course. In the case of a pronounced destructive process in the periapical region, treatment was prolonged, and 7-15 medical leeches were used. X-ray quality control of treatment was carried out. Before treatment and 6 months after treatment (Figure 5-6).

Fig. 1: Leech insertion at treatment site

Fig. 2: Leech beginning to feed itself

Fig. 3: Leech releasing the patient

Fig. 4: End result after the leech release the patient

Fig. 5: Exacerbation of chronic periodontitis

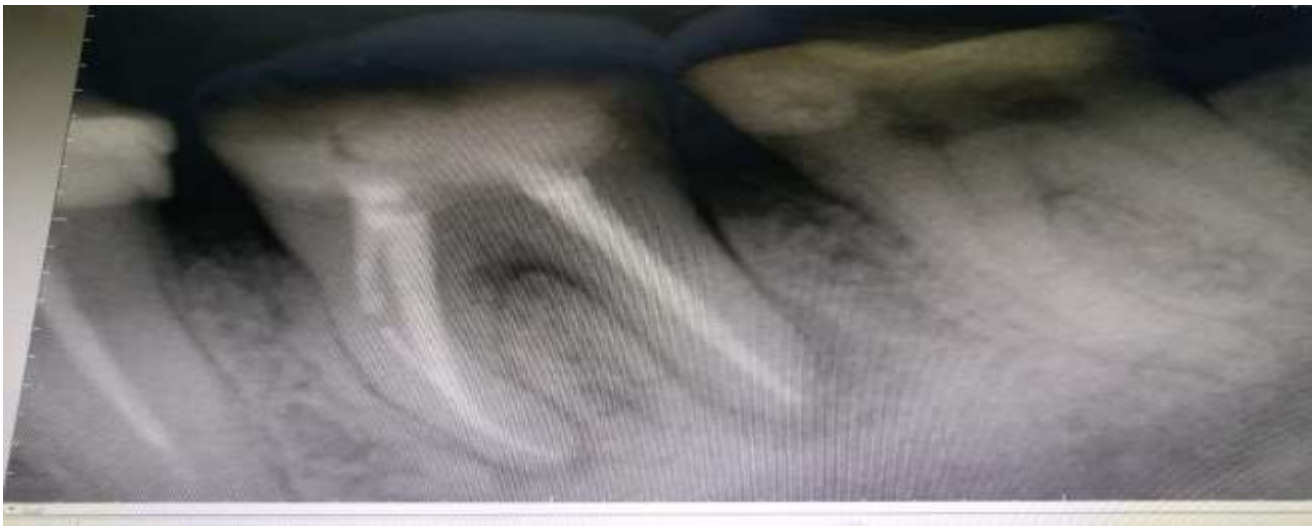

Fig. 6: Long-term results of treatment after 6 months

Results and Discussions

The distribution of teeth (54) treated for exacerbation of chronic apical periodontitis is presented in Table 1.

Table 1: The distribution of teeth treated for exacerbation of chronic apical periodontitis.

№	Observation groups	Numbers		Total
		Single rooted	Multiple or Multi rooted	
1	Core group	15	13	28
2	Comparison group	14	12	26
	Total	29	25	54

Before treatment (day 1), semi-quantitative assessment of the severity of pain in patients with exacerbation of chronic apical periodontitis, the main group, and the comparison group was not significantly different. In the second visit, the intensity of pain was significantly reduced in patients of the main group, and in the third - the pain practically disappeared, i.e., it noted a pronounced analgesic effect hirudotherapy. In the comparison group, the severity of pain on the 3-5th day decreased, but on the 7-10th day remained significantly higher than with hirudotherapy and. conceivably analgesic effect of hirudotherapy due to the fact that under the influence of salivary gland secretion of the medicinal leech reduced levels of bradykinin and kallikrein, which, together, with a substance P mediates pain. Along with the analgesic effect, hirudotherapy had a pronounced vasoconstrictor effect. The edema and hyperemia severity indexes in the patients of the main group decreased in the dynamics of

treatment, and the edema completely disappeared by the 3rd visit. By 7-10 days in the comparison group, hyperemia and edema decreased but did not disappear. The anti-edematose effect of hirudotherapy may be associated with improved microcirculation in the oral mucosa and periodontal disease manifested in an increase in vascular tone, blood flow efficiency, and the area of functioning capillaries. Indicators of the total score (in points) of the severity of clinical manifestations of acute exacerbation of chronic apical periodontitis in the main group and the comparison group in the dynamics of treatment (Table 2).

Table 2: Indicators of the total score (in points) of the severity of clinical manifestations of acute exacerbation of chronic apical periodontitis

Visit day number	Groups		Significance of Differences
	The main	Comparisons	
1st	8.70 ± 0.47	8.63 ± 0.50	> 0.005
3-5th	1.50 ± 0.40	5.63 ± 0.68	<0.001
7-10th	0.30 ± 0.15	5.25 ± 0.75	<0.001

In the main group of patients with exacerbation of chronic apical periodontitis, who used hirudotherapy, on the 3-5th day of treatment, the total indicator of clinical manifestations decreased 5.8 times, and on the 7-10th day - 29 times. In the comparison group, the decrease in the total index was less pronounced. On the 3-5th day of treatment, the index decreased 1.5 times, and on the 7-10th day-1.6 times.

Thus, according to clinical data, the use of hirudotherapy in patients with exacerbation of chronic apical periodontitis revealed clinical improvement in the patients' condition associated with the rapid resolution of the inflammatory process and due to the pronounced analgesic and decongestant effect of the secretion of the medicinal leech secretion. Indicators of the amount of secretion of unstimulated mixed saliva in the compared groups did not significantly change during the treatment process.

Indicators of cytosis and absolute neutrophil content in mixed saliva in patients with exacerbation of chronic apical periodontitis with conventional endodontic treatment and combined it with hirudotherapy were significantly higher than in healthy if u (Table. 3). In the compared groups, the cytosis and absolute neutrophil counts did not significantly differ in the compared groups.

Table 3: Results of cytological and cytochemical study unstimulated mixed saliva of patients with exacerbation of the chronic apical periodontitis and the main group and the comparison group

Groups observations Options	Control group	The main			Comparative		
		Dates (visits)			Dates (visits)		
		1	2	3	1	2	3
Cytosis (x10 ^{6} in 1 ml)	0,68±0,08	1,60±0,21	1,68±0,41	1,14±0,21	1,33±0,02	1,03±0,32	2,18±0,31
Neutrophils (x10 ^{6} in 1 ml)	0,09±0,01	1,28±0,23	0,43±0,13	0,24±0,09	1,09±033	0.84±0,04	0,61±0,22
Lysosomalnation proteins (SCS)	0,67±0,01	0,27±0,09	0,55±0,13	0,78±0,14	0,33±0,02	0,42±0,05	0,45±0,09

Notes: * - p < 0.05; ** - p <0.01 - reliability of differences in the indices of the compared groups

In the main group, in the third visit, the index of cytosis of mixed saliva decreased significantly, in the comparison group-significantly increased. The absolute neutrophil content in mixed saliva in the compared groups decreased during the

treatment process (Table 3). However, with hirudotherapy in the third visit, the absolute neutrophil content was 5.3 times lower than before treatment, and in the comparison group, respectively, 1.8 times. Thus, during hirudotherapy, there is a more pronounced tendency to a decrease in the content of neutrophils in mixed saliva, which reflects the anti-inflammatory effect of the medicinal leech.

The results of a study of the content of lysosomal-cationic proteins in neutrophils of mixed saliva in patients of the main group and the comparison group show that under the influence of hirudotherapy, normalization of the state of the oxygen-independent bactericidal system of neutrophils is observed, which reflects the activation of local nonspecific immune defense mechanisms.

According to clinical and laboratory studies, it has been revealed that hirudotherapy during exacerbation of apical periodontitis relieves an edematous, painful, inflammatory syndrome and reduces the treatment time. The use of hirudotherapy in apical periodontitis prevents the development of an exacerbation of the disease after endodontic treatment.

Conclusions

Application hirudotherapy in the combination therapy of patients with chronic apical periodontitis has a pronounced anti-inflammatory, analgesic, and decongesting, detoxification effects. Hirudotherapy allows one to stop inflammatory syndrome in earlier periods and reduce the time of treatment.

The use of hirudotherapy is advisable to prevent the activation of the inflammatory process in patients with endodontic diseases during and after the endodontic intervention. Hirudotherapy of patients with chronic apical periodontitis normalizes the independent oxygen system of lysosomal-cationic proteins in neutrophils and activates the mechanisms of local non-specific immune defense. Thus, the analysis of the method of hirudotherapy in the combination therapy of diseases of endodontics showed its effectiveness. However, according to the cytological study revealed earlier clinical recovery, compared with morphological.

References

1. Sedigh-Rahimabadi M, Shams M, Fani M, Chijan MR (2016) Gingivitis from the Viewpoint of Traditional Persian Medicine. Iran J. Med. Sci., 41(3): S35.
2. Sirak SW, Entschladen F, Shchetinin EW, Grimm WD (2015) Low-level laser irradiation (810 nm) with toluidinblue photosensitizer promotes proliferation and differentiation of human oral fibroblasts evaluated in vitro. Journal of Clinical Periodontology, T. 42. № S17. 328a-328.
3. Ergesheva EV, Davidyants AA, Dorofeev AE, Sevbitova MA, Timoshina MD (2018) Evaluation of effectiveness of an alternative method in complex treatment of diseases of endodont. Asian J. of Pharm., 12(S3):933-936.
4. Grimm WD, Arnold WA, Sirak SW, Vukovich MA, Videra D, Giesenhagen B (2015) Clinical, radiographic, and histological analyses after transplantation of crest-related palatal-derived ectomesenchymal stem cells (paldscs) for improving vertical alveolar bone augmentation in critical size alveolar defects. Journal of Clinical Period ontology, T. 42(S17):366b-366.
5. Hyson JM (2005) Leech therapy: a history. J. Hist. Dent., 53(1):25-7.
6. Ling He, Juan Zhong, Qimei Gong, Sahng G. Kim, Samuel J. Zeichner, Lusai Xiang, Ling Ye, Xuedong Zhou, Jinxuan Zheng, Yongxing Liu, Chenyu Guan, Bin Cheng, Junqi Ling, Jeremy J. Mao (2017) Treatment of Necrotic Teeth by Apical Revascularization: Meta analysis. Scientific Reports, 7: 13941.
7. Shetinin EV, Sirak SV, Ignatiadi ON, Sirak AG, Demurova MK, Dygov UA (2014) Experimental and laboratory substantiation of the choice of antibacterial agents for the treatment of periodontitis. Medical Bulletin of the North Caucasus, T. 9. № 4. C.349-351.
8. Platonova VV, Sevbitov AV, Shakaryants AA, Dorofeev AE (2018) The experimental clinical substantiation of treatment of patients with odontogenic phlegmon of maxillofacial area using delargin in complex therapy. Klinicheskaya Laboratornaya Diagnostika, 63(5):293-296.

9. Abdualkader AM, Ghawi AM, Alaama M, Awang M, Merzouk A (2013) Leech therapeutic applications. Indian J. Pharm. Sci., 75(2): 127-37.

10. Timoshin AV, Sevbitov AV, Drobot GV, Yumashev AV, Timoshina MD (2018) Use of bioresorbable plates on the basis of collagen and digestase for treatment of diseases of oral mucosa (review of clinical cases). International Journal of Green Pharmacy, 12(1): S290-S296.

11. Sevbitov A, Kuznetsova M, Dorofeev A, (...), Mironov S, Yusupova LL Dental anomalies in people living in radionuclide-contaminated regions. Journal of Environmental Radioactivity. 2020. 216: 106-190.

12. Jha K, Garg A, Narang R, Das S (2015) Hirudotherapy in Medicine and Dentistry. Journal of Clinical and Diagnostic Research, 9(12):05-7.

13. Suk Keun Lee, Sang Shin Lee, In Sun Song, Yeon Sook Kim, Young Wook Park, Jae Yong Joo, Heung Sik Um, Jin Woo Kim, Kyeong Yeon Kim, Seung Jin Choi, Kyung-Hwan Jung, Soo I Chung (2004) Paradoxical effects of elastase inhibitor guamerin on the tissue repair of two different wound models: sealed cutaneous and exposed tongue wounds. Experimental & Molecular Medicine, 36(3):259-267.

14. Grimm WD, Shchetinin EV, Bobrishev DV, Sirak SV (2015) Quantifying analysis of advanced glycosylation end products (ages) expression in periodontitis patients with diabetes type II. Medical Bulletin of the North Caucasus, T. 10 (2): 178-183.

15. Voloshina IM, Borisov VV, Sevbitov AV, Davidyants AA, Mironov SN, Kuznetsova M Yu, Ergesheva EV (2018) Distinctive features of micro crystallization of mixed saliva in children with different levels of activity of carious process. Asian J of Pharm., 12(S3):1017-1020.

16. Haycox CL, Odland PB, Coltrera MD, Raagi GJ (1995) Indications and complications of medicinal leech therapy. J. Am Acad. Dermatol., 33(6):1053-55.

17. Gröbe A, Michalsen A, Hanken H, Schmelzle R, Heiland M, Blessmann M (2012) Leech therapy in reconstructive maxillofacial surgery. J. Oral. Maxillofac. Surg., 70(1):221-7.

18. Makeeva IM, Nesvizhskiĭ Iu V, Butaeva NT, Turkina A Iu, Akimova IV (2009) Periodontal tissues contamination prevention in the process of teeth endodontic treatment. Dentistry, T. 88 (3): 38-40.

19. Sevbitov AV, Dorofeev AE, Davidiants AA, Ershov KA, Timoshin AV (2018) Assessment of pain perception of elderly patients with different levels of dentophobia during surgical dental appointment. Asian Journal of Pharmaceutics, 12(3): S1012-S1016.

20. Taneja P1, Rowson J (2011) National survey of the use and application of leeches in oral and maxillofacial surgery in the United Kingdom. Br J. Oral Maxillofac. Surg., 49(6):438-41.

21. Byrne P J, Bernstein PE (2001) The use of medicinal leeches to treat macroglossia secondary to blunt trauma. Otolaryngol Head Neck Surg., 125(6):649-50.

22. Hullett JS, Spinnato GG, Ziccardi V (2007) Treatment of an ear laceration with adjunctive leech therapy: a case report. J. Oral Maxillofac. Surg., 65(10):2112-4.

23. Makeeva IM, Byakova SF, Novozhilova NE, Adzhieva EK, Golubeva GI, Grachev VI, Kasatkina IV (2016) Detection of artificially induced vertical root fractures of different widths by cone-beam computed tomography in vitro and in vivo. International Endodontic Journal, T. 49 (10): 980-989.

24. Sevbitov AV, Dorofeev AE, Kuznetsova M Yu, Timoshin AV, Ershov KA (2019) Comparative characteristics of the crystallogram of the oral fluid in patients who use heroin and methadone. Periódico Tchê Química, 16(33) 94-101.

