



RESEARCH ARTICLE

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Therapeutic Effectiveness of using Papaya Leaves in the Treatment of Dental Diseases

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ABSTRACT

The Purpose of the Study is to study the therapeutic - pharmacological effect of using papaya leaves (*Carica papaya* L) grown in a solar greenhouse in an arid ecosystem in dental practice.

Materials and Methods: Prepared solution from the leaves of the melon tree (*Carica papaya* L) examined 23 patients in the clinic for the treatment of dental diseases.

Results: The resulting solution from papaya leaves can reduce the treatment period for purulent infections in the oral cavity by two to three days, which in turn is important for the timely prevention of abscesses and other therapeutic side effects. Agricultural technology has been developed for the possibility of cultivation and technology for obtaining proteolytic enzymes from the leaves and fruits of the melon tree in the conditions of Turkmenistan using renewable energy sources to create a microclimate.

Conclusion: The advantage of the proposed product compared to the known ones is that the treatment uses an organic, natural composition of plant substances, which has effective properties in the treatment of dental inflammatory diseases and wounds.

ARTICLE HISTORY

Received December 01, 2023

Accepted December 21, 2023

Published January 05, 2024

KEYWORDS

Melon Tree, Papaya Leaves, Proteolytic Enzymes, Purulent Wounds, Dentistry, Treatment, Technology

Introduction

Relevance of the problem. One of the main objectives of the State Program "Health" developed by the President of Turkmenistan Gurbanguly Berdimuhamedov is to provide the country's population with medicines using domestically produced medicines, studying the possibility of growing valuable medicinal plants in the conditions of Turkmenistan, developing agricultural cultivation technologies and providing the country with medicines and valuable raw materials for industry [1].

In the modern world, much attention is paid to the use of biologically active drugs of plant origin in medical practice. World medicine is limited from the use of antibiotics, as the immune system is reduced and leads to other complex consequences. Scientists believe that in the future, antibiotics can be replaced by super antibodies, for which the cell wall will not be an obstacle, which will be able to penetrate inside cells and destroy pathogenic bacteria, viruses and toxins. They are testing technology for modifying antibodies, which allows them to freely enter and leave cells [1-10].

Biotechnological Features of Melon Tree Leaves

Botanical description. Melon tree or papaya (*Carica papaya* L) is a

perennial tropical palm-like plant up to 4–6 m high in the papaya family (*Caricaceae*). The fruits hang on cuttings under the crown, are juicy, very large (up to 10-length 30 cm, up to 1-weight 4 kg), resembling a melon in size and shape. Ripe yellow fruits are edible. Inside there is a cavity filled with black seeds. The trunk is green, herbaceous, not woody, has no branches, at the top of the crown there are large palmate dissected 5-7 lobed leaves with very long petioles. The leaves and fruits of the melon tree are used to improve the digestibility of tough meat. See figure and photograph 1 [2-9].

Medicinal raw materials. Leaves, seeds and dried milky juice - latex - are used as medicinal raw materials. Coagulated lumps of latex are crushed and dried in the sun or with gentle artificial heating (in the latter case, higher quality papain is obtained). The resulting latex is dissolved in water and precipitated with alcohol to purify papain. Papain is also found in smaller quantities in other parts of the plant, in particular in the leaves (*Folia Caricae Papayae*) [2-9]. Of some -economic importance are the seeds of the melon tree, which contain fatty oil, which includes oleic (75.50%), linoleic (2.13%), palmitic (11.38%), stearic (5.25%) -and arachidic (0.31%) acid.

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Figure1 and Photography. Melon tree (*Carica papaya* L): 1 - general appearance of the plant; 2 – female center of branches; 3 – male flower; 4 – stamen; 5 - longitudinal section of the fetus; 6 – longitudinal section of the seed.

Biologically active substances of leaves. By electrophoresis in an acidic environment, 7 proteins were identified in the latex of *Carica papaya* L: lipase, chitinase, lysozyme and a complex of proteolytic enzymes: Papain (EC 3.4.22.2) - monothiol cysteine endoprotease. Due to the nature of its enzymatic action, it is called “plant pepsin”. But, unlike pepsin, papain is active not only in acidic, but also in neutral and alkaline environments (pH range 3–12, optimum pH 5); Chymopapain (EC 3.4.22.6) is a monothiol cysteine proteinase [2-10]. Due to its substrate specificity, it is similar to papain, but differs from it in electrophoretic mobility, stability and solubility; Proteinase IV - cysteine proteinase, the main proteinase of latex, makes up about 30% of the protein present in it (Buttle DJ) [2-10]. Shows a high degree of homology with papaya proteinase III (81%), chymopapain (70%) and papain (67%). Very close to chymopapain in molecular weight and molecule charge. Contamination of chymopapain by this enzyme is responsible for its heterogeneity across studies. M. P. Thomas et al. (1994) classify this enzyme as a fraction of chymopapain M; Carikain (EC 3.4.22.30) - the most alkaline among the cysteine proteinases of papaya latex [2-10]. Like papain, it is first produced as the inactive zymogen procaricain, which contains an inhibitory proregion of 106 N-terminal amino acids. Activation of the enzyme involves the cleavage of the proregion of the molecule without its subsequent conformational changes. The structure of papaya proteinases was studied using X-ray analysis (Maes D); Proteinase w (endopeptidase A, peptidase A) is a monothiol cysteine proteinase [2-10]. It is a polypeptide containing 216 amino acid residues and 3 disulfide bonds. For the manifestation of its enzymatic activity, the presence of a free cysteine residue in the active center is important (Dubois T). Shows a high degree of homology with papain (68.5%). In terms of the specificity of the enzymatic action, it resembles papain, since it binds to the

substrate in areas where disulfide bonds are localized. Cleavage occurs when the next position is leucine, valine or threonine; Peptidase II - alkaline monothiol cysteine proteinase. Contains a dithioacyl group in the catalytic center.

The latex of unripe papaya fruits also contains inhibitors of proteolytic enzymes: cystatin (a proteinase inhibitor with a molecular weight of 11,262 Da) and a protein with the properties of a cysteine proteinase inhibitor, the molecule of which consists of 184 amino acid residues, contains 2 disulfide bonds and 2 carbohydrate residues in positions Asp84 and Asp90 (Odani S) [2-10].

The ripe fruits of the melon tree contain 8–12% sugar, a significant amount of vitamins A, B1, B2, C and D, and tonic substances.

Pharmacological properties. Proteolytic enzymes of the melon tree have anti-inflammatory, anticoagulant, degradative, analgesic, bactericidal, hemolytic properties. Considering these properties, proteolytic enzymes of plant origin are widely used in medical practice: ophthalmology, surgery, neurosurgery, orthopedics, urology, gastroenterology, etc. They help destroy semipeptide proteins and amino acids, dissolve dead cells, without affecting normal ones.

The pharmacological industry of foreign –countries produces more than 100 drugs using papaya milky juice (lecozyme, lecopaine, caricase, carcazyme, carpain, wobenzyme, super fat burner No. 1 and many others), widely used in various fields of medicine [3-9].

Based on this, taking into account the biological characteristics of papaya and the natural and climatic conditions of Turkmenistan, we came to the conclusion that it was necessary to select the design of greenhouses for its cultivation. For this purpose, in 1981, on the basis of the NPO "Solntse" of the Academy of Sciences of the TSSR, a trench-type solar greenhouse was built. Then a combined-industrial combined greenhouse was developed using thermal waste from the plant and solar energy to create a microclimate. Currently, agricultural technology and technology for obtaining proteolytic enzymes from the melon tree have been developed.

Materials and Methods

Material research. As a result of scientific research, free phenolic compounds, tannins, organic acids, steroidal and triterpene saponins, flavonoids, lipids, coumarins, glucose, alkaloids, which are widely used in modern medical practice, were discovered in the leaves of the melon tree in place with proteolytic enzymes [2-10].

The results of chemical and spectral analysis of melon tree leaves are shown in Tables 1, 2.

Table 1: Results of Chemical Analysis of Papaya Leaves

Chemical element name	Method of determination	Element composition
Ash content % at 450 °C	Gravimetric	17.5
Potassium (K ₂ O), mg/kg	Photometric	54881
Sodium (Na ₂ O), mg/kg	Photometric	4600
Phosphorus (P ₂ O ₅), mg/kg	Photometric	6848
Sulfur (S), mg/kg	Gravimetric	7934
Calcium (Ca), mg/kg	Titrimetric	14626
Magnesium (Mg), mg/kg	Titrimetric	21601
Iron (Fe), mg/kg	Photometric	129.2
Zinc (Zn), mg/kg	Atomic absorption	1.8

Treatment methods and clinical examination. The work makes an analytical analysis of methods for treating the disease in dental practice using proteolytic enzymes. The most commonly used drugs and proteolytic enzymes :

- Trypsin is an enzyme of animal origin, used in the form of a solution, prepared ex tempore;
- Chymotrypsin - an enzyme of animal origin, use like trypsin;
- Chymopsin - an enzyme of animal origin (a mixture of trypsin and chymotrypsin), use a 0.1% solution of the drug;
- Terrilitin - used in the form of a solution containing 40-50 proteolytic units (PU) in 1 ml;
- lysoamidase is a proteolytic enzyme that also has a bacteriolytic effect. It is used topically in the form of a solution in 0.01 M sodium phosphate buffer (pH = 8.0) in concentrations of 5, 10, 25 PE per 1 ml. E.V. Borovsky et al. (1989) suggest using a 1% enzyme solution, injecting it into periodontal pockets on turundas;
- Profezim is a proteolytic drug immobilized on aminocellulose, which has a prolonged effect, has increased resistance to temperature factors, denaturing agents, and pH changes, and does not have a locally irritating effect.

Table 2: Results of Spectral Approximate Analysis of Papaya Leaves (In Terms of Plant Dry Matter)

Chemical element name	Element composition, mg/kg	Chemical element name	Element composition, mg/kg
Vari (Ba)	21	Tungsten (W)	< 1
Strontium (Sr)	35	Scandium (Sc)	0.53
Molybdenum (Mo)	0.35	Cadmium (Cd)	<3
Lithium (Li)	3.5	Hafnium (Hf)	<3
Chromium (Cr)	0.35	Waist (Tl)	<0.3
Bismuth (Bi)	0.26	Cobalt (Co)	<1
Tin (Sn)	0.18	Indium (In)	<5
Titanium (Ti)	1.75	Platinum (Pt)	<0.5
Vanadium (V)	0.35	Tantalum (Ta)	<50
Beryllium (Be)	0.17	Series (Ce)	<20
Silver (Ag)	0.02	Silver (Au)	<0.5
Surma (Sb)	<0.5	Zirconium (Zn)	7
Germanium (Ge)	<0.3	Gadolinium (Gd)	<10
Gallium (Ca)	0.2	Thorium (Th)	<30
Lead (Pb)	0.35	Arsenic (As)	<2
Manganese (Mn)	17.5	Uranium (U)	<1
Nickel (Ni)	0.53	Lanthanum (La)	<1
Copper (Cu)	5.2	Ytterbium (Yb)	<0.1
Niobium (Nb)	<1	Yttrium (Y)	<1

We have conducted research using papaya leaves on diseases of the oral cavity and teeth. Considering that the leaves of the melon tree contain papain, an enzyme of plant origin that is capable of dissolving denatured areas of collagen and other insoluble proteins, without losing its activity in the slightly acidic environment of the source of inflammation.

The essence of the results of use is as follows: a solution is prepared from papaya leaves. The infectious focus is treated with a remedy consisting of a 5%–20% solution of a substance isolated from the leaves of a melon tree in distilled water and saline.

When treated with the above methods, the agents are mainly administered intramuscularly or intravenously and contain formulations containing chemical products, which does not allow their use for all patients due to possible allergic reactions and the insensitivity of the human body to some substances used in the treatment of dental diseases. In addition, known methods of treatment are based on the use of complex technologies and equipment for producing these enzymes, which affects the availability of treatments on a mass scale.

Considering the shortcomings of the above-described means and methods of treatment, the task was set to research and find new, effective means and methods for the treatment of purulent infections in dental practice, and methods based on the use of products, mainly of organic origin.

Some of them are known from [10-12].

The enzymes obtained in the work were used in surgery. The proteolytic and anti-inflammatory properties of papain made it possible to use it for the sanitation of various fistulas, cleansing of necrotic tissue from wounds and preparing the surfaces of trophic ulcers for skin grafting in patients [2-9].

With wet necrosis after a burn, papain allows you to speed up the cleansing of the surface from necrotic tissue and preparing it for skin grafting. Enzymes were used in hospital military unit 2523 in the treatment of osteomyelitis, phlegmon, narrowing of the esophagus, mastitis, as evidenced by photographs and a certificate of use.

The use of domestic papain makes it possible to effectively treat patients, while limiting the use of scarce, expensive imported drugs such as lekozim, lecopain and others. (See the effect of the drugs lekozim, krpazim, wobenzym, etc.).

Results and Discussion

Research results. Having studied the possible use in dental practice of proteolytic enzymes of plant origin of the melon tree, the contents of chemical elements in the leaves of the melon tree were determined using chemical methods.

As a result, the following chemical substances were identified in the leaves of the melon tree:

- Using the gravimetric method, ash content at 450 °C was determined, which is 17.5% and sulfur - 7934 mg/kg;
- Using the photometric method, the content of chemical elements in the leaves was determined: potassium (K₂O) - 54881; sodium (Na₂O) – 4600; phosphorus (P₂O₅) -6848; iron (Fe) -129.2 mg/kg;
- The titrimetric method determined the content: calcium (Ca), - 14626 mg/kg and magnesium (Mg) - 21601 mg/kg; The zinc content (Zn) was determined by the atomic absorption method - 1.8 mg/kg.

As can be seen in the leaves, the quantitative content of substances is: potassium -54881 mg/kg; magnesium - 21601 mg/kg; calcium - 14626 mg/kg; sulfur - 7934 mg/kg and phosphorus - 6848 mg/kg; sodium – 4600 mg/kg; then comes iron - 129.2 mg/kg and zinc - 1.8 mg/kg. See photo of a solution made from melon tree leaf biomass.



Photo 1: Photograph of The Prepared Solution from The Biomass of Melon Tree Leaves

Having studied the biological and chemical properties of melon tree leaves, including testing for negative reactions in the human body, we came to the conclusion that substances obtained from melon tree leaves can be successfully used in the treatment of inflammatory purulent wounds in dentistry.

The Essence of The Research Results Is as Follows: The resulting biomass from the leaves of the melon tree is made into a 5% - 20% solution from distilled water or saline.

The infectious focus is treated with a product consisting of a 5% - 20% solution of a substance obtained from leaf biomass. The frequency of treatment of the source of infection is three times a day at regular intervals or with a frequency of treatment depending on the degree of inflammation of the organ. This method of treatment can be used in the treatment of other inflammatory processes in the human body.

Under our supervision in 2018, in Ashgabat clinics, preparations made from the biomass of melon tree leaves were used to treat purulent infections, ulcerative gingivitis, periodontitis, especially with suppuration from periodontal pockets and abscess formation; a total of 23 patients were treated.

During treatment, the infectious focus is treated with the drug three times a day at regular intervals or with a frequency of treatment depending on the degree of infection of the organ. The method allows you to expand the possibility of its use by using a catheter, endoscope and other medical instruments, with the help of which enteral penetration into the dental part of the human body is carried out. Treatment is carried out with a medicinal product, including a 5-20% solution of a substance crushed mass of fresh melon tree leaves in distilled water or saline solution. The infectious focus is treated with the drug three times a day at regular intervals or with a frequency of treatment depending on the degree of infection of the organ.

In addition, the proposed method makes it possible to expand the possibility of its use by using a catheter, endoscope and other medical instruments, with the help of which enteral penetration into the root part of a human tooth is carried out.

Moreover, the treatment can be applied to areas with purulent infection in the esophagus or stomach, as well as areas with purulent infection in the eyes, ear, and in the treatment of otitis media.

The wound was closed with gauze –pads soaked in a 0.25-0.5% papain solution, and in cases of a large amount of necrotic tissue, the papain concentration was increased to 1%. It is noted that when applied topically, papain does not penetrate deeply into the tissue, but exerts its effect mainly on the surface of the wound, lysing non-viable tissue. Therefore, in 25 cases, we infiltrated a papain solution into the necrotic tissue, which contributed to the rapid cleansing of the wound.

After the first applications of papain, nonviable tissues were clearly –demarcated from healthy tissues and softened, especially the surface layer, which resembled a mucoid mass. During dressings, there was abundant discharge from the wounds with noticeable rejection of necrotic tissue. Bright granulations appeared on the surface of the wounds, cleared of non-viable tissue, and a tendency toward epithelization was noted along the edges.

In patients with suppuration of postoperative –wounds, the disappearance of purulent fibrous plaque was noted after 3-4 applications, epithelization of wounds - after 6.

Good results were obtained in patients with phlegmons and abscesses of various localizations of the upper and lower extremities: after opening the abscess and applying 25 applications of papain, complete cleansing of the wound from non-viable –tissue was noted, after 9 - epigelization of the wound.

In patients with mastitis, after opening the abscess, wound cleansing was noted on the 4th day of application, on the 7th day - good epigelation and on the 11th day - complete healing by secondary intention.

After 4 applications of papain in patients with trophic ulcers in the foot area, –cleansing of the wound surface was observed, after 7 - good granulation, and therefore on the 8th day the patients underwent free skin grafting according to Trisch. The grafts are viable, and wound healing was noted on day 12. The tissue granulation and dermal scar are delicate.

Discussion

The main distinguishing feature of the method, remedy and method for treating purulent infections in dental practice is the use as a remedy of such a component as biomass of leaves from crushed melon tree in combination with distilled water or saline solution. In addition, a new method of treatment is proposed, including both the new claimed agent and its use with instruments designed to penetrate the human dental body.

The advantages of the proposed remedy over the known ones is that the treatment uses an organic, natural composition of substances of plant origin, which has effective properties in the treatment of purulent infections of various types. The other component, the “carrier” of the product, is distilled water or saline solution, the purity of which also cannot be doubted.

Moreover, compared to the known one, the remedy and method can reduce the treatment period for purulent infections by two to

three days, which in turn is important for the timely prevention of abscesses and other clinical side effects.

The choice of the percentage of the substance made from the biomass of melon tree leaves and the frequency of treatment of areas of purulent infection in the oral cavity were determined experimentally, taking into account the course of the purulent process and the phase of the wound process itself. When using 4% of the leaf biomass substance, the expected treatment effect was not observed. That is, the experimental dose of 5% did not affect the focus of purulent infection. The use of more than 20% of the substance in the product does not radically affect either the speed of treatment or its effect, compared with the use of, for example, 19 or 25% of the substance.

Thus, the optimal ratio was identified, ranging from 5 to 20% of leaf biomass from the total composition of the product.

The effective frequency of infection treatment has been established as three times daily. It has also been found that, depending on the phase of the wound process, the frequency of treatment can be two or more than three times a day.

Data from testing the use of a 20% solution from leaf biomass in a separate group of patients are attached. Also attached are data from testing the use of a 5% Papaya solution on another group of patients.

With regard to the compliance of the claimed method and means with the criteria of novelty, level of effectiveness and industrial applicability, the following can be established.

The analytical search conducted by the authors did not reveal any publications or patents that described means and methods for treating purulent infections using a substance from the biomass of melon tree leaves. Thus, the methodology and method are not part of a previously known level of technology and meet the criterion of scientific novelty.

A similar search did not reveal data on the percentage of leaf biomass substances established by the authors for the purposes of human treatment, in particular in the treatment of purulent infections in dental practice. Thus, the essence of the invention is not an obvious solution for a specialist in the field of medicine and meets the criterion of “inventive step”.

Since the proposed means and method can be practically used in such a branch of human activity as medicine, the claimed invention meets the criterion of “industrial applicability”.

Thus, an effective ratio was identified, ranging from 5 to 20% of the solution of papaya leaf biomass from the total composition of the product.

The effective frequency of infection treatment has been established as three times daily. It has also been found that, depending on the phase of the wound process, the frequency of treatment can be two or more than three times a day.

Testing data for the use of a 20% Papaya solution in the product were carried out on a group of 23 patients.

The search did not reveal publications or patents that describe means and methods for treating purulent infections using the substance of Papaya juice. Thus, the research is not part of a previously known level of technology and meets the "novelty" criterion.

A similar search did not reveal data on the established percentage of the substance in Papaya juice for the purpose of effective treatment in humans, in particular in the treatment of glandular organs. Since the proposed means and method can be practically used in such a branch of human activity as medicine, the research meets the criterion of "industrial applicability" [2-9].

The use of enzymes in clinical dental therapy makes it possible to actively and purposefully –intervene during suppurative processes, accelerating the cleansing of wounds from non-viable tissue. One of the advantages of using proteolytic enzymes is their necrotic effect in the absence of a damaging effect on living tissue. According to a number of authors, proteolytic enzymes of plant origin, unlike those of animal origin (trypsin, chymotrypsin), are capable of acting on denatured proteins, which served as a theoretical justification for their use in purulent surgery to remove necrotic tissue, dissolve fibrin accumulations in body cavities and even blood vessels, diluting wound discharge, as well as facilitating the penetration of medicinal substances through intercellular subtraction [1,6].

Thus, the results of treatment of patients with purulent wounds in the oral cavity indicate the effectiveness of –the use of proteolytic enzymes from the biomass of melon tree leaves.

Conclusions

The melon tree is a food product, but biologically active substances such as lipase, chitinase, lysozyme and a complex of proteolytic enzymes, including an inhibitor of cysteine proteinases, have been found in this plant.

In addition, the entire part of the melon tree contains proteolytic enzymes papain, chymopapain, proteinase, etc. These plant enzymes are widely used in medicine and various industries. For example, the proteolytic enzyme "Papain" is capable of breaking down proteins into polypeptides and amino acids, and hydrolyzes any peptide bonds, with the exception of proline bonds and glutamic acid bonds with a dissociated carboxyl group. It has a greater ability to break down proteins compared to most proteases of animal and bacterial origin. Although the potency of papain preparations varies depending on the method of preparation, it has the ability to break down lean meat in an amount 35 times its own weight. High quality papain digests egg albumin, the amount of which is 300 times its own weight. From the analysis of the milky juice obtained –from the fruits of the melon tree grown in the conditions of Turkmenistan, for the content of protein activity according to Lowiy (81%) and proteolytic activity according to Kunitz (1700 units/mg) [4,5].

Using the method of ion exchange chromatography, –the separation of the milky juice of the melon tree was obtained: the protein released in peak II has a proteolytic activity of 4000 units/mg, the remaining proteins came together without separating. According to the electrophoregram data, bands 1 and 5 are milky juice, band 7 is protein released in peak II, electrophoretically pure, without any impurities. Papain from " Serva " (Germany) was used as a marker.

The enzymes obtained from papaya milky juice differ from known enzymes of animal origin in that at certain concentrations –they exhibit more pronounced chondro-, fibrino- and thrombolytic properties, more actively lyse and hydrolyze young connective tissue and membrane formations [2].

A solution made from leaf biomass was used as a raw material for treatment. Since the milky tubes contain milky juice in all parts of the melon tree, which contains proteolytic enzymes.

The resulting biomass from the leaves of the melon tree was prepared as a solution containing a 5-20% solution of the substance, crushed mass of fresh leaves of the melon tree in distilled water or physiological solution.

As a result, the use of enzymes in clinical dental therapy makes it possible to actively and purposefully –intervene during suppurative processes, accelerating the cleansing of wounds from non-viable tissue. One of the advantages of using proteolytic enzymes from the melon tree is their necrotic effect in the absence of a damaging effect on living tissue. Proteolytic enzymes of plant origin, unlike those of animal origin (trypsin, chymotrypsin) are capable of acting on denatured proteins, which served as a theoretical justification for their use in purulent surgery to remove necrotic tissue, dissolve fibrin accumulations in body cavities and even blood vessels, dilute wound discharge, as well as facilitating the penetration of drugs through intercellular subtraction.

The accumulated experience of treatment with papain allows us to conclude that this enzyme –can be successfully used in the treatment of orthopedic pathology and the consequences of injuries. Enzyme therapy in orthopedics deserves further study and implementation in the practice of medical institutions. The method of enzyme therapy is simple, can be used on an outpatient basis, does not cause serious complications and in many cases is economical and effective.

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