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(54) **Title:** OBTAINING VITAMIN/MINERAL/DRUG/HORMONE/ENZYME/BIO-MATERIAL COMPOSITION BY MEANS OF THROMBOCYTE CONCENTRATE

(57) **Abstract:** The invention relates to the in vitro preparation of PRF in tubes whose inner surface is coated with, but not limited to this, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins; to the release of said vitamin/mineral/drug/hormone enzyme/biomaterial compositions in a certain concentration within a certain period of time in the PRF during centrifugation and to in vivo application to the damaged area of the bone and soft tissue for use in in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and children's clinics. The main object of the invention is to provide a tube in which the PRF is prepared in a tube covered with local biological methods that accelerate tissue healing.

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OBTAINING VITAMIN/MINERAL/DRUG/HORMONE/ENZYME/BIO-MATERIAL COMPOSITION BY MEANS OF THROMBOCYTE CONCENTRATE

TECHNICAL FIELD

The invention relates to forming a product preparation for preparing a vitamin/mineral/drug/hormone/enzyme/biomaterial composition with thrombocyte concentrate.

PRIOR ART

In the field medicine and dentistry, reengineering of lost tissues has been a professional and social problem for hundreds of years, and studies are still ongoing. Tissue engineering includes applications for the regeneration of defects in the bone and soft tissue caused by degeneration, surgery or trauma. For this purpose autogenous bone and soft tissue grafts used as gold standard have problems such as donor region morbidity, blood loss and limited amount of acquisition. Studies are ongoing to solve these problems and the use of various local/systemic biological products is investigated. Bone morphogenetic proteins, bisphosphonates, biomaterials, vitamins, minerals and hyaluronic acid are used in these applications.

In this context, platelet-rich treatments (PRT), which are widely used in medicine and dentistry as an autogenous graft option since they are obtained from the patient's own blood, are a newly developed treatment option under the name of "orthobiologicals" in modern medical research. Today, it is of great interest as a biological treatment area in osteoarthritis and musculoskeletal repair as a reliable, noninvasive (medical procedure that does not require entering under the skin or can be performed without entering the body by opening an unnatural path). Orthobiology is a newly developed science and it is aimed to stimulate bone and tissue healing by applying the materials obtained from biological means.

The platelet-rich treatments are obtained by centrifuging the patient's own blood and separating the active, platelet-rich part. The resulting platelet-rich plasma fraction is administered to the damaged tissue for example by injection. The α-granules of platelets are rich in a variety of growth factors that play a key role in tissue repair, such as platelet-derived growth factor, transforming growth factor β, insulin-like growth factor, vascular endothelial growth factor and epidermal growth factor. These growth factors have the potential to stimulate cell proliferation, matrix remodeling and angiogenesis.

In the known state of the art, as it is obtained from the patient's own blood, the use of PRF (platelet rich fibrin) as an autogenous graft continues to be widely used in medicine and dentistry. In most of the publications in the literature, it is emphasized that in clinical applications of PRF, besides an effective development of neovascularization, rapid skeletal tissue maturation and increased tissue repair, almost no infection development.

PRF plays an active role in angiogenesis, immune control and epithelial closure, which are crucial in healing and soft tissue maturation. Since it is taken from the patient, it has many advantages such as not causing allergic reactions, preparation in a short time and easily, no risk of disease transfer, control of inflammation and suppression of infection thanks to the leukocyte therein and cytokines secreted by them.

Obtaining platelet rich fibrin in glass-coated plastic or glass tubes poses several problems. Various studies state that glass and glass-coated plastic tubes disrupt the quality and quantity of the fibrin matrix formed, the silica particles in the glass have a detrimental effect by disrupting the autogenicity of the preparation. These problems bring to mind the idea that the tubes used will be coated with agents that will not damage the PRF matrix structure, but rather have synergistic effects and increase matrix density.

BRIEF DESCRIPTION OF THE INVENTION

The invention relates to the in vitro preparation of PRF in tubes whose inner surface is coated with, but not limited to this, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins; to the release of said vitamin/mineral/drug/hormone enzyme/biomaterial compositions in a certain concentration within a certain period of time in the PRF during centrifugation and to in vivo application to the damaged area of the bone and soft tissue for use in in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and children's clinics.

The main object of the invention is to provide a tube in which the PRF is prepared in a tube covered with local biological methods that accelerate tissue healing.

Another object of the invention is to obtain a product-preparation with a better fibrin structure than that obtained by the classical method, with the preparation of PRF in the tube covered with local biological agents that accelerate the designed tissue healing.

Another object of the invention is to provide a product-preparation that provides equal application of the dose and release of the agents in the PRF and its composition.

Another object of the invention is to provide a product-preparation which is useful by showing synergistic effect in accelerating and/or supporting the healing process of wounds, burns, chronic wounds with PRF and the agents of its composition.

Another object of the invention is to increase the release time and shorten the resorption time of the biological/biocompatible agent in the tissue to be applied to the site by means of the vitamin/mineral/drug/hormone/enzyme/biomaterial compounds covering separator tube and PRF with product-preparation.

Another object of the invention is to reduce the workload during the operation and accelerate the surgical procedure with the obtained PRF and vitamin/mineral/drug/hormone/enzyme composition product-preparation.

For achieving all of the abovementioned objects the present inventions relates to forming a product-preparation for preparing vitamin/mineral/drug/hormone/enzyme/biomaterial composition with thrombocyte concentrate. Accordingly, it is provided that vitamin/mineral/drug/hormone/enzyme/biomaterial compositions are obtained together with PRF (platelet rich fibrin=thrombocyte rich fibrin) which obtained from the patient himself/herself. The subject of the present invention relates to a product-preparation which can be applied in vivo to the damaged region in bone and soft tissue, which is biologically compatible with the tissue and enhancing the tissue regeneration with the synergistic effect of the biological products comprised therein, wherein biological/biocompatible agents covering the separator tube release in a certain concentration within a certain period of time in PRF, it comprises the following steps

i. Coating of 20 mL glass tube with vitamin/mineral/drug/hormone/enzyme/biomaterial composition,
ii. Taking 10 mL of blood from the patient to be treated and putting it in a tube covered with vitamin/mineral/drug/hormone/enzyme/biomaterial composition
iii. Centrifuging the tube in a particular centrifuge machine,
iv. Removing the tube from the centrifuge machine and obtaining the product-preparation
v. Using the obtained product-preparation in the patient.

Another preferred embodiment of the invention is that vitamin/mineral/drug/hormone/enzyme/biomaterial compositions such as, but not limited to these, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds are used as an inner surface coating material in the tube mentioned in step (i). Thus, the preparation-product is used in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and child clinics.

Another preferred embodiment of the invention is to provide a preparation-product which can be applied in vivo to the patient by obtaining a thrombocyte in a more intensive concentration, prepared in vitro after step (iv), related growth factors, and a composition comprising, but not limited to these, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins. This composition, which can be transplanted into the patient for the treatment of the indications described by the examples, will be mainly beneficial in accelerating tissue remodeling during the healing process by providing direct metabolization of the biological/biocompatible agents with which the separation tube is coated at the wound region, with a denser fibrin structure.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows the view of PRF preparation with the classical method.

Figure 2 shows a representative view of the empty separator tube coated with vitamin/mineral/drug/hormone/enzyme/biomaterial composition.

Figure 3 shows a representative view of platelet-rich plasma (PRF) + vitamin/mineral/drug/hormone/enzyme/biomaterial preparation obtained after centrifuging the 10 ml blood which is taken into the tube covered with vitamin/mineral/drug/enzyme/biomaterial composition.

REFERENCE NUMBERS PROVIDED IN THE DRAWINGS

10 Standard Tube

1 Vitamin/mineral/drug/hormone/enzyme/biomaterial tube inner surface coating composition

2 Venous blood

3 Red blood cells

4 Platelet-rich plasma(PRF)+vitamin/mineral/drug/hormone/enzyme/biomaterial

5 Platelet-poor plasma

6 Platelet-rich plasma fraction (PRF)

DETAILED DESCRIPTION OF THE INVENTION

In this detailed description, subject of the invention relates to production of the product-preparation (4) obtained by combining local biological agents (1) accelerating the tissue healing and PRF (6) for use in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and child clinics and it is only explained with examples that will have no limiting effect for a better understanding of the subject.

In the known state of art, as shown in Figure 1 platelet-rich treatments (PRT) are obtained by centrifuging the patient's own blood in a glass and plastic tube (10) and separating (6) the active, platelet-rich part. The resulting platelet-rich plasma fraction is administered to the damaged tissue for example by injection. The α-granules of platelets are rich in a variety of growth factors that play a key role in tissue repair, such as platelet-derived growth factor, transforming growth factor β, insulin-like growth factor, vascular endothelial growth factor and epidermal growth factor.

The subject of the invention is to obtain PRF (4) in the tube (10) coated with local biological agents that accelerate tissue healing. Resorption time is reported to be 3-15 days in the site where PRF (6) is applied. This indicates that there is a need for studies for a product accelerating the regeneration such as PRF (6) to benefit from its effectiveness for a longer period, to increase the resorption time and to increase the effectiveness of the material in which it is mixed until it is resorbed in the field where it is applied. "Preparation in which PRF is combined with local biological agents accelerating the tissue healing" obtained by the procedure of the invention provides that it both shows longer activity in the receiving site and shows synergistic effect with the agent with which it is mixed by eliminating the short resorption time emerged as disadvantage during clinical use of the PRF (4). Even if there is a combined use in previous studies, for example, vitamin C, vitamin D, hyaluronic acid concentrates are applied to the tissue separately by the obtained thrombocyte concentrate and/or after being prepared separately, it is mixed in ex

vivo and transplanted into the relevant region. However, there are shortcomings in these methods such as difficulty in preparation, long bedside time, high cost, short release time of the additional agent in the tissue and low dose.

The present invention relates to coating (1) of the inner surfaces of the separator tubes with these agents such that while preparing the PRF (6), as shown in Figure 2, but not limited to these, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds will provide that certain amount of concentration of the toxin agents will be present within the PRF in certain period of time. It will enable the acceleration of the healing process and the remodeling of the tissue by providing direct metabolization of agents at the wound region which are known to have positive impact on the wound healing such as vitamin C, vitamin D, hyaluronic acid, bisphosphonate, antibiotic, anti-inflammatory, analgesic, corticosteroid, calcium, parathormone.

The composition to be obtained by the method of the invention is an easy-to-prepare, time-saving at the bedside and relatively low-cost product. Further, the preparation to be obtained by the procedure according to the invention makes it possible to reduce the workload during the operation, accelerate the surgical procedure and increase the therapeutic effectiveness by ensuring (4) that the release and the dose of the PRF and the agents in the composition are applied evenly.

It is expected from the platelet concentrate and compositions obtained by the method of the present invention to be useful in accelerating and/or supporting the healing process of wounds, burns, chronic wounds, to successfully heal the wounds which conventional treatments have failed, to reduce the infection risk, to

accelerate the patient recovery, to improve the patient comfort, to reduce the medical care costs and to provide better aesthetical results.

The preparations obtained by the method of the present invention will be useful in wound and tissue healing, regeneration treatments, treatment of traumatic and surgical wounds, application of grafts, dental implants, treatment of osteoradionecrosis or ostecrosis. Bone defects or bone-related diseases will be beneficial in the healing of bone grafts and bone fracture treatment. It will be useful in the treatment of periodontal disease, in the need for periodontal tissue regeneration, in increasing connective tissue formation.

Preparations obtained by the method of the present invention will be useful in providing pharmaceuticals for tissue regeneration or cosmetic compositions for skin regeneration.

While the compositions obtained by the method of the present invention will be prepared to be locally applicable, they will guide the preparation of preparations in injectable or subcutaneously injectable form into the wound or into the bone defect or adjacent region thereof.

The preparations to be obtained by the method of the invention can be prepared in drop form or thread form by preparing them in a solid carrier, fixing this carrier to the defect, or mixing it with a cream or emulsion or incorporating it into a hydrogel carrier.

The preparation obtained by the method of the invention can be stored by freezing at -70°C and can be used at a later date.

With the products obtained by the method of the invention, by developing an alternative local application technique instead of systemic usage of the agents such as vitamin C, vitamin D, hyaluronic acid, bisphosphonate, antibiotic, anti-

inflammatory, analgesic, corticosteroid, calcium, parathormone, less damage to organs such as liver and kidney undergoing drug excretion and synthesis will be achieved.

The product obtained by the method of the invention allows it to be properly shaped according to the missing organ or field by being turned into a model with three-dimensional scanners.

The invention relates to a product-preparation accelerating the tissue regeneration with the synergistic effect of the biological products contained therein, which is biologically compatible with the tissue, in vivo applicable to the damaged region in the bone and soft tissue wherein the PRF (platelet-rich fibrin=thrombocyte-rich fibrin) is prepared in vivo in tubes (1) coated with, but not limited to this, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins for use in in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and children's clinics, wherein said agents performs releasing in a certain concentration within a certain period of time in the PRF (4) during centrifugation, it comprises the following steps

i. Coating of 20 mL glass tube (10) with vitamin/mineral/drug/hormone/enzyme/biomaterial composition (1),

ii. Taking 10 mL of blood (2) from the patient to be treated with the standart method and putting it in a tube (10) covered with vitamin/mineral/drug/hormone/enzyme/biomaterial (1)

iii. Centrifuging the tube in a particular centrifuge machine,

iv. Removing the tube (10) from the centrifuge machine and obtaining the product-preparation (4)

v. Using the obtained product-preparation in the patient.

As shown in Figure 3, mentioned in step (i) and according to the invention coating agent in the separator tube (1) whose inner surface is coated, vitamin/mineral/drug/hormone/enzyme/biomaterial compounds (2) are, but not limited to these, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins.

After step (iv), the preparation-product (4) containing vitamin/drug/biomaterial is obtained with PRF. The obtained preparation-product is transplanted to the patient.

EXAMPLES

1. Preparation-Product

Use of the preparation containing PRF+ vitamin D (4) obtained by the method of the invention to accelerate regeneration in tooth extraction and cyst cavities

A sample of 10 ml whole blood (2) from the patient is taken into a tube (10), the inner surface of which is prepared according to the method of the present invention, coated with vitamin D (1) as shown in Figure 2. The separator tube (10) containing blood and vitamin D agent is centrifuged (Figure-3). The obtained PRF+vitamin D (4) is taken from the tube with the help of forceps. The platelet-rich part of the obtained concentrate is separated from the erythrocyte-rich part. After tooth extraction under local anesthesia, the wound is closed primarily by placing it

to the bone defect where the cyst is removed. In various studies, it has been determined that vitamin D is very important for absorption of sufficient amount of calcium and phosphorus, it positively affects bone development, increases the growth rate of animals and improves calcium absorption. This process is supported by peptide hormones such as parathyroid, calcitonin, insulin and growth hormone; 25-hydroxyvitamin, 1,25 dihydroxyvitamin D, glucocorticoids, sex hormones and thyroid hormones. Vitamin D, having active metabolite of $1,25[OH]_2D_3$, finds itself a wide range in almost all human metabolism. In addition to all these effects, Vitamin D also plays an important role in the mineralization of growth plates, in the mineralization of osteoids on trabecular and cortical bone.

In this example, the use of vitamin D (4) together with the PRF membrane allows to shorten the healing time of the bone defect at the surgical site 4 months after the operation and to implant the same session without requiring a second operation.

2. Preparation-Product

Use of the preparation containing PRF + biomaterial (4) obtained by the method of the invention in the regenerative treatment of tooth extraction socket

A sample of 10 ml whole blood (2) from the patient is taken into a tube (10), the inner surface of which is covered with biomaterial (1), as shown in Figure 2 prepared according to the method of the present invention. The separator tube containing whole blood (2) is centrifuged (Figure-3). The wound is closed primarily by placing the obtained PRF+biomaterial (4) to the bone defect after tooth extraction under local anesthesia. Bone grafts are used to fill the defect region, provide structural support and correct the defective appearance.

The purpose of the preparation-product obtained is to allow the osteoblasts to provide bone healing by keeping the cells away from the region that will adversely affect healing in the area where healing is expected by using a barrier membrane. In addition to these used materials, it is known that growth factors also have an effect on the induction of bone formation. Bone regeneration is faster when growth

factors are added to bone grafts. Growth factors play a role in cell proliferation, differentiation, chemotaxis and extracellular matrix synthesis.

The product-preparation obtained by the invention is sutured primarily by covering the bone defect that occurs in a patient with small molar extraction with the PRF membrane+collagenous allograft (4). It is observed that bone healing in the surgical site occurs without any problem 6 months after the operation.

With this example, the preparation of the PRF membrane in the tube (10) inner surface of which is covered with the biomaterial (1) and its application to the site (4) facilitates the manipulation of the graft, provide its stabilization and shortens the 6-month period required for bone healing.

3. Preparation-Product

Use of the preparation containing PRF+hyaluronic acid (4) obtained by the method of the invention in skin regeneration

A sample of 10 ml whole blood (2) from the patient is taken into a tube as shown in Figure 2, the inner surface of which is covered with hyaluronic acid (1) prepared according to the method of the present invention. The separation tube containing whole blood is centrifuged (Figure-3). The resulting PRF+hyaluronic acid (4) is injected subcutaneously under the skin.

While application fields of PRF in dermatology is chronic wounds, ulcers and burns, also it has been successfully used in the field of cosmetic dermatology based on the principle of wound healing. Thanks to the growth factors it contains, PRF stimulates the production of collagen, fibroblast and other matrix components. Therefore, it can be used to increase the effect of cosmetic procedures. Also, since PRF is an autologous product, it does not carry an allergic reaction risk.

4. Preparation-Product

Use of the preparation containing PRF+diclofenac (4) obtained by the method of the invention in the treatment of muscle injury

A sample of 10 ml whole blood (2) from the patient is taken into a tube (10), the inner surface of which is covered with diclofenac prepared according to the method of the present invention. The separation tube containing whole blood is centrifuged (Figure-3). The obtained PRF+diclofenac (4) is injected intramuscularly into the damaged muscle tissue. It is observed that the patient showed healing 4 weeks after the operation with intramuscular PRF application due to laceration occurring in the Achilles tendon.

5. Preparation-Product

Use of the preparation containing PRF+antibiotic (4) obtained by the method of the invention in the treatment of gingival recession

A sample of 10 ml whole blood (2) from the patient is taken into a tube (10), the inner surface of which is coated with antibiotic (1) prepared as shown in Figure 2 according to the method of the present invention. The separator tube (10) containing whole blood is centrifuged (Figure-3). After removing the half-thickness flap around the teeth under local anesthesia, the obtained PRF+antibiotic (4) is placed in the soft tissue defect, the flap is sutured by being slid to the crown. Mucogingival surgery defines procedures to protect the gingiva, eliminate abnormal frenulum or muscle joints, and increase vestibular depth. Citric acid, tetracycline HCl, tetracycline-HCl as well as chemical additives such as fibrin glue, and sodium hypochlorite applied to the root surface during mucogingival surgery have been tested in both animal and human studies. These agents have been used to remove the smear layer on the root surface, to remove collagen fibrils in the dentin in order to facilitate the formation of new connective tissue attachment, and to remove the cytopathic cement residues that are thought to prevent the growth of gingival fibroblast. It is known that PRF acts by thickening the keratinized gingiva in the treatment of gingival recession.

In this example, the preparation and application to the site (4) of the PRF membrane in the tube (10) inner surface of which is covered with antibiotic (1) increases the growth factors that contribute to the periodontal regeneration in the gingival groove fluid, allowing the keratinized gum gain to be achieved in a larger amount and in a shorter time.

6. Preparation-Product

Use of the preparation containing PRF+bisphosphonate (4) obtained by the method of the invention in cyst enucleation surgery

10 ml of whole blood (2) sample is taken from the patient into a tube, the inner surface of which is covered with bisphosphonate (1) prepared according to the method of the present invention. The separator tube (10) containing whole blood is centrifuged (Figure-3). The wound site is closed primarily by placing the obtained PRF+biomaterial (4) to the bone defect under local anesthesia after cyst enucleation. Bisphosphonate-derived drugs inhibit bone resorption by disrupting the function of osteoclasts and increase bone density.

In this example, the preparation and application of the PRF membrane in the tube (10), whose inner surface is covered with bisphosphonate, increases bone density and accelerates the healing of bone tissue and soft tissue.

7. Preparation-Product

Use of the preparation containing PRF+biomaterial (4) by the method of the invention in sinus lift surgery

10 ml of whole blood (2) sample is taken from the patient into a tube (10), the inner surface of which is covered with biomaterial (1) prepared according to the method of the present invention. The separation tube containing whole blood is centrifuged (Figure-3). The wound is closed primarily by placing the obtained PRF+biomaterial (4) to the bone defect after tooth extraction under local anesthesia. Bone grafts are used to fill the defect site, provide structural support and correct the defective

appearance. These osteoconductive materials fill the bone defect with a passive roof function. In oral and maxillofacial surgery, especially in cases with extensive reconstructive surgery, fibrous healing appears as an undesired condition. In order to prevent these problems, a guided bone regeneration technique has emerged. The purpose of this technique is to allow the osteoblasts to provide bone healing by keeping the cells away from the region that will adversely affect healing in the area where healing is expected by using a barrier membrane. In addition to these used materials, it is known that growth factors also have an effect on the induction of bone formation. Bone regeneration is faster when growth factors are added to bone grafts. Growth factors play a role in cell proliferation, differentiation, chemotaxis and extracellular matrix synthesis.

In this example, the preparation of the PRF membrane in the tube (10) covered with the biomaterial and its application to the field (4) facilitates the manipulation of the graft, enhances its stabilization and allows the shortening of the 6-month period required for bone healing.

CLAIMS

1. Product-preparation comprising therapeutical thrombocyte concentrate, it is **characterized by** in vitro preparation of PRF (6) with tubes (10) inner surface of which is covered with vitamin/mineral/drug/hormone/enzyme/biomaterial compositions (1), release of the said vitamin/mineral/drug/hormone/enzyme/biomaterial compositions (1) in a certain concentration within a certain period of time in PRF (4) during centrifugation and in vivo application thereof to the damaged region in bone and soft tissue.

2. Product-preparation comprising therapeutical thrombocyte concentrate according to the claim 1, **characterized in that** as an inner surface coating material, said tube (10) is one (1) of the vitamin/mineral/drug/hormone/enzyme/biomaterial compositions such as, but not limited to these, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins.

3. Product-preparation comprising therapeutical thrombocyte concentrate according to the claim 1, **characterized in that** it is used in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and child clinics.

4. Product-preparation comprising therapeutical thrombocyte concentrate according to the claim 1, **characterized in that** it is biocompatible with the

tissue and it increases tissue regeneration with the synergistic effect of the biological products it contains.

5. Obtaining a thrombocyte concentrate product-preparation used in dentistry, maxillofacial surgery, orthopedics, dermatology, plastic and reconstructive surgery, neurosurgery, sports medicine, ophthalmology, veterinary medicine, oncology, ear-nose-throat medicine, jaw facial prostheses, eye and children's clinics, **characterized in that** PRF (6) is prepared in vivo in tubes (10) inner surface of which is coated with, but not limited to these vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins, said agents perform releasing in a certain concentration within a certain period of time in PRF (4), it can be applied in vivo to the damaged region in the bone and soft tissue, it enhances the tissue regeneration with the synergistic effect of the biological products it contains wherein it is obtained by using the following process steps

i. Coating of 20 mL glass tube (10) with vitamin/mineral/drug/hormone/enzyme/biomaterial composition (1),

ii. Taking 10 mL of blood (2) from the patient to be treated with the standart method and putting it in a tube (10) covered with vitamin/mineral/drug/hormone/enzyme/biomaterial (1)

iii. Centrifuging the tube in a particular centrifuge machine,

iv. Removing the tube (10) from the centrifuge machine and obtaining the product-preparation (4)

v. Using the obtained product-preparation in the patient

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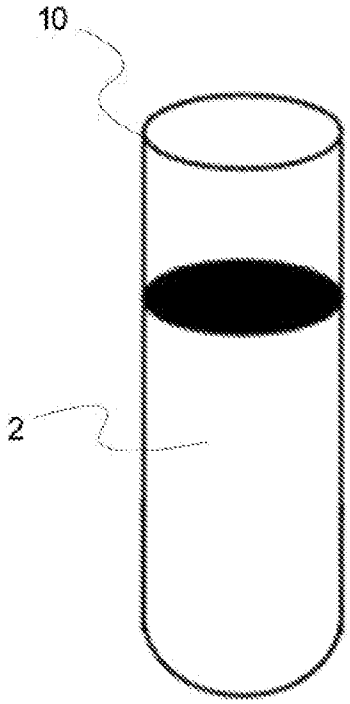
6. Method for obtaining a product-preparation comprising therapeutical thrombocyte concentrate according to the claim 5, **characterized in that** vitamin/mineral/drug/hormone/enzyme/biomaterial compounds (1) mentioned in step (i) are, but not limited to these, vitamins (vitamin C, vitamin D, vitamin A, vitamin E, vitamin B and its derivatives), minerals (e.g. calcium, iron, zinc), hyaluronic acid, synthetic grafts (biomaterials), bisphosphonate, antibiotic, antifungal, antiparasitic, antiviral, anti-inflammatory, analgesic compounds, aspirin, steroids, glucocorticosteroids (corticosteroid compounds), hormones (e.g. parathormone), enzymes, enzyme inhibitors, peptides, glycoproteins, lipoproteins, growth factors, immunomodulators, immunoglobulins, tumor-killing compounds, toxins.



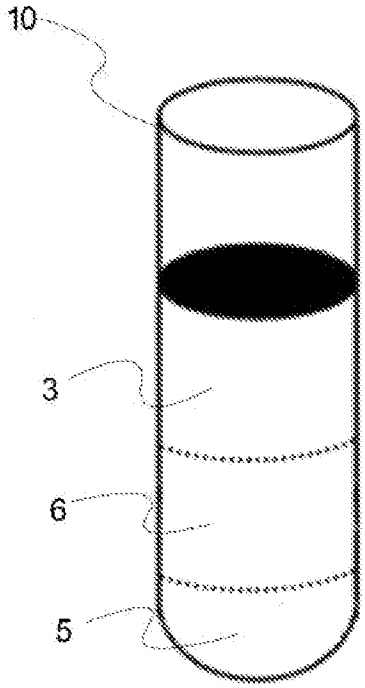


Figure 1

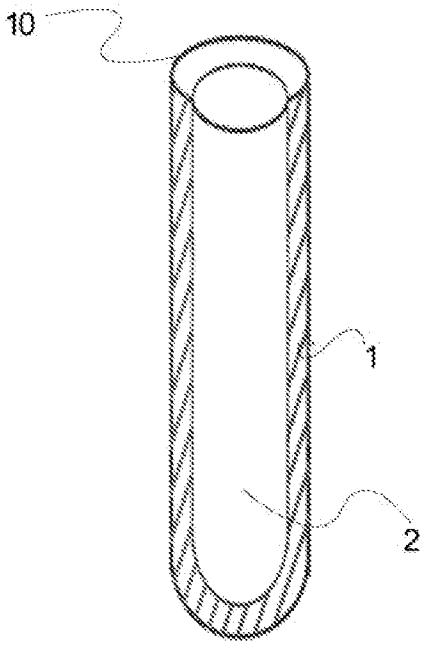


Figure 2

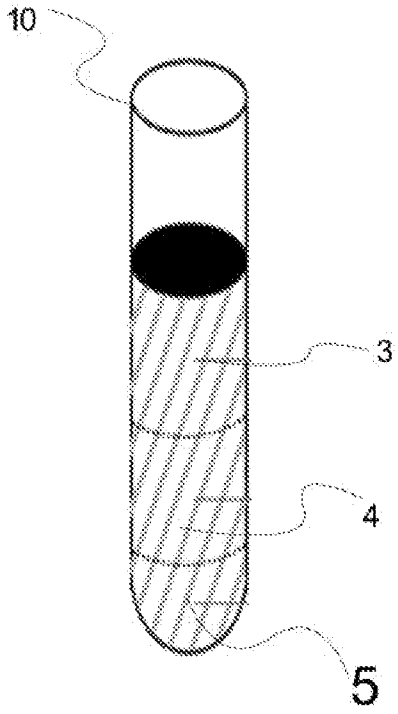


Figure 3

INTERNATIONAL SEARCH REPORT

International application No.
PCT/TR2020/050471

A. CLASSIFICATION OF SUBJECT MATTER

B01L 3/14 (2006.01)i; B01D 17/038 (2006.01)i; B04B 5/00 (2006.01)i; A61J 1/00 (2006.01)i

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

B01L; B01D; B04B; A61J

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

EPODOC, WPI

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2011110948 A2 (TURZI ANTOINE) 15 September 2011 (2011-09-15) Abstract; claims 6, 21 and 32; page 9	1-6
X	US 2014227241 A1 (LACZA ZSOMBOR; VACZ GABRIELLA; LACERTA TECHNOLOGIES INC) 14 August 2014 (2014-08-14) Paragraphs 2, 36-37, 40, 67 and 141	1-6
X	JP 05157747 A (NISSHO KK) 25 June 1993 (1993-06-25) Abstract; claim 1	1-4

☐ Further documents are listed in the continuation of Box C. ☑ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

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Date of the actual completion of the international search	Date of mailing of the international search report
30 September 2020	**30 September 2020**

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INTERNATIONAL SEARCH REPORT
Information on patent family members

International application No.
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