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Potential Combination of Hesperetin and Vitamin C based on PLGA (Poly Lactic-co-Glycolic Acid) on Healing Fragility Fracture in Osteoporosis Patients

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Abstract

Background: Osteoporosis is a degenerative bone disease that occurs around 15.3% in Southeast Asia. Osteoporosis patients are very vulnerable and often experience fragility fractures. Until now, the treatment of fragility fractures, namely pharmacological and operative therapy, has not been optimal in restoring bone density and allowing recurrence.

Purpose: The aim of this research is to find the modality of therapy fragility fracture that can prevent and improve the condition.

Methods: This literature was prepared using literature review methods being sourced from valid medical journals such as published in PubMed, Research Gate, and Google Scholar.

Results: Hesperetin in citrus fruit can inhibit RANKL-induced osteoclastogenesis, reduce the amount and size of F-actin, and accelerate fracture healing in vivo. Vitamin C, which is also present in citrus fruit, has an important role in the synthesis of the triple helix collagen and shows better fracture healing at week 4. PLGA has a role in more specific drug delivery, preventing rapid clearance, biodegradable, increasing calcium deposition, and extending drug release time.

Conclusion: Combination of Hesperetin and PLGA-based vitamin C can be promising therapies in the healing and prevention of fragility fractures. Suggestion: The author suggests conducting further research to determine side effects, proper dosage, and drug administration.

Keywords: Fragility Fracture, Hesperetin, PLGA, Vitamin C

Introduction

Osteoporosis is a condition where bone resorption increases faster than bone degeneration [1]. Osteoporosis can be found all over the world and is still a health problem, especially in developing countries. Based on data from WHO, worldwide there are 200 million people suffering from osteoporosis [2]. The prevalence of osteoporosis in Southeast Asia is estimated to be around 15.3% [3] and based on data from the Indonesian Osteoporosis Association in 2007, 32.3% occurred in women and 28.8% occurred in men of the total population of those aged over 50 years [4].

As life expectancy increases, the risk of osteoporosis sufferers is increased, which is generally experienced at the age of 50 years and over [5]. In conditions of osteoporosis, sufferers tend to experience fractures before being diagnosed. The lack of follow-up examinations for fractures caused by osteoporosis has resulted in many cases of osteoporosis being underdiagnosed, resulting in a fragility fracture. Fragility fractures occur as a result of impact to bone with minimal energy. The bones most commonly fractured are the pelvis, spine, and wrist [6]. Among all fractures due to osteoporosis, fractures of the pelvis have the greatest morbidity and mortality [7].

Until now, the handling of fragility fractures due to osteoporosis in the world and in Indonesia has been carried out by providing pharmacological and operative therapy. However, the choice of therapy has not been optimal in restoring bone density and there is still the possibility of recurrence. Osteoporosis is more common in elderly women who have menopause due to loss of estrogen function [8]. Therefore, handling fragility fractures that is easy, simple, safe and of good effectiveness is needed.

In the last decade, several studies have shown the role of hesperetin and vitamin C in the healing process of fragility fractures due to osteoporosis. Hesperetin

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is a flavonoid glycoside from the family flavonoid found in citrus fruit [9]. Hesperetin is known to have activity against osteogenic differentiation of MSCs (mesenchymal stem cells) which are the main cells that play a role in fracture healing. Mesenchymal stem cells have the potential to differentiate into several cell types such as chondrocytes, osteoblast, and adipocytes [10]. In addition, hesperetin is known to prevent bone resorption with an inhibitory effect on RANKL which plays a role in osteoclastogenesis [11].

Citrus fruits also have high vitamin C concentrations. Vitamin C is an essential element in fracture healing. Deficiency of vitamin C can cause the fracture to heal more slowly. Specifically, the function of vitamin C is in the hydroxylation of amino acids required in the formation of triple helix collagen which is essential in callus formation, and acts as an antioxidant and anti-inflammatory in the fracture healing process [12]. Considering that fragility fracture in osteoporosis is a degenerative bone disease that is vulnerable to the elderly population, administration of hesperetin and vitamin C has the potential to be a holistic therapeutic innovation. Holistic is meant to intervene in overall health in addition to maintaining bone health but also can increase immunity, prevent inflammation, and help the healing process of fractures due to osteoporosis [13].

Looking at the various mechanisms associated with hesperetin and vitamin C for fragility fractures due to osteoporosis, raises new hope in the treatment of fragility fractures in the elderly population. As therapy for fractures, hesperetin and vitamin C require transporters or carriers which are important in reaching the target and as nanoencapsulation so that the half-life of the drug is longer. One that has the potential to become a biotransporter in the bone repair process is PLGA. PLGA (poly lactic-co-glycolic acid) with nanoparticles can increase the calcium deposition process through osteoblasts [14]. Better understanding of the role of hesperetin and PLGA-based vitamin C will open the possibility of finding other interventions in the treatment of fragility fractures due to osteoporosis in the future. Therefore, the authors are interested in investigating more deeply the potential of hesperetin and PLGA-based vitamin C for healing fragility fractures in osteoporosis.

Material and Method

This literature is written using the literature review method. In writing literature, the author uses the Medical Subject Heading (MeSH) with the keywords Osteoporosis, Fragility Fracture, Hesperetin, Vitamin C, and PLGA. To get a specific journal, the author uses the Boolean logic "AND".

Initial search on the PubMed page, Research gate, Google Scholar, WHO, Indonesian Ministry of Health, and other journals against Indonesian and English language literature to get a research framework. A search was undertaken to identify literature in medical journals focused on fragility fractures in osteoporosis patients. After obtaining the research framework, then the authors identify the literature from the title and abstract. Literature related to the research framework will be included in the research inclusion, while literature that does not speak Indonesian and English will be excluded. All literature obtained will be read independently by the author, then the preparation of this paper is carried out.

In the literature search, a total of 132 literature was found after the duplicates were removed. Furthermore, it was filtered based on the title, abstract, and keywords so that there were 49 articles which would be reviewed as a whole in the literature. After a review of the contents of the entire literature, a total of 38 articles were used in the preparation of this literature review.

Table 1. RUST score analysis on second week and fourth week [31]

Variable	Second Week Median	P	Fourth Week Median	P
RUST score control	4.5 (4-8)	0,467	4.5 (4-6)	0.023
RUST score Vit C 5 mg	4.0 (0-4)		4.0 (4-6)	
RUST score Vit C 10 mg	4.5 (4-6)		4.0 (0-4)	
RUST Score Vit C 20 mg	6.0 (4-8)		8.0 (8-8)	

Results and Discussion

Osteoporosis

Osteoporosis is a clinical disorder characterized by low bone density, low bone mass, and bone structural defects. Osteoporosis occurs due to bone resorption is faster than bone formation so that the bones degenerate [1, 15]. The imbalance between matrix production and the mineralization process followed by decreased osteoblast activity or a lack of minerals needed in the bone-building process, such as calcium, magnesium and phosphate, can play a role in the formation of osteoporosis [16, 17]. Osteoporosis is defined by the WHO (World Health Organization) as a T-score of BMD (Bone Mineral Density) below -2.5 standard deviation (SD). BMD is most often examined using DXA (Dual-energy X-ray Absorptiometry) [18]. This combination causes the bones to be very brittle and have a greater risk of fracture than normal people. Osteoporosis is divided into 2, namely postmenopausal osteoporosis and osteoporosis senil. From the onset of menopause and 10 years after that, bone loss rates increase to 3% per year. From the age of 65 or 70 years the bone loss rate in women decreases gradually and by the age of 75 the rate becomes 0.5% per year [15].

The risk of fracture increases with age [15]. At the age of more than 50 years, it is estimated that 50% of women and 20% of men experience fractures associated with osteoporosis [19]. The term fracture that most commonly occurs in osteoporosis is called a fragility fracture . Fragility fracture is a fracture caused by low-energy trauma [20]. Hip fracture is the most common of all fragility fractures. Hip fractures cause the greatest morbidity, mortality, and social and financial costs [7].

Potential of Hesperetin Against Inhibition of Osteoclastogenesis and Healing of Fragility Fracture

Hesperetin (3 ', 5,7-trihydroxy-4-methoxyflavone) is a metabolite of hesperidin (hesperetin-7-O-rutinoside) which is the main flavonoid group and is abundantly found in citrus fruit. Hesperetin has several biological activities such as anti-inflammatory, analgesic, antioxidant, and lipid lowering effects [21-23]. Hesperetin (Hes) at high concentrations (100 and 200 μM) significantly decreased osteoclast survival. Hesperetin inhibits RANKL-induced osteoclastogenesis on various osteoclast precursors (BMMs, RAW 264.7, and Splenocytes), addition of Hes inhibits osteoclast maturation in a manner dose-dependent. Hesperetin is known to have activity against osteogenic differentiation of MSCs which are the main cells that play a role in fracture healing. This process occurs via the ERK and Smads signaling pathways. MSCs have the potential to differentiate into several cell types such as chondrocytes, osteoblasts, and adipocytes [10].

Hesperetin is also able to reduce the number and size of rings F-actin. Osteoclasts with normal function require the formation of normal rings F-actin , so that a reduction in the number and size of F-actin can interfere with osteoclast function [11,24]. Hesperetin in vivo showed inhibitory effect on osteoclastogenesis. Mice with model LPS-induced bone loss were given LPS without hesperetin as control and with hesperetin. In mice given hesperetin, the trabecular bone close to the growth plate had drastically fewer osteoclasts compared to models without hesperetin [25].

Hesperetin, besides being able to inhibit osteoclastogenesis, also accelerates fracture healing in vivo. In a mouse model with osteotomy tibial compared with the composite hesperetin/gelatin-hMSC, hesperetin/gelatin scaffold, gelatin scaffold, and no treatment was used as a control. At the 8th postoperative week the fracture line was still visible and only minimal bone growth was found in the control and gelatin scaffold, whereas in the hesperetin / gelatin-hMSC group and the hesperetin / gelatin group, a bone bridge defect was found and at the final stage of endochondral ossification [26].

Potential of Vitamin C Against the Healing of Fragility Fracture

Citrus fruit besides having flavonoid hesperetin, also contain abundance of vitamin C or ascorbic acid. In ¾ glass of orange juice, you get 93 mg of vitamin C which fulfills 103% daily value (DV) [27]. Vitamin C plays an important role in collagen synthesis, which contributes to form organic bone [28]. The function of vitamin C specifically is in the hydroxylation of amino acids which is needed in the process of forming the triple helix of collagen which is essential in callus formation, and acts as an antioxidant and anti-inflammatory in the fracture healing process [12]. Vitamin C has a certain correlation with the risk of hip fracture. A meta-analysis of dietary vitamin C consumption showed that there was a positive association between dietary vitamin C intake and risk of hip fracture. The results showed that there was a reduction in the risk of hip fracture by 5% for every 50 mg / day increase in the dose of dietary vitamin C consumption [29]. A meta-analysis study conducted by Zeng, found a total of 13 articles with 19,484 identified subjects, that people with a higher frequency of vitamin C consumption had a 34% prevalence (95% CI, 6% - 53%) of less pelvic fractures. . Furthermore, statistical analysis of the association between vitamin C consumption and risk of osteoporosis obtained a relative risk (RR) of 0.66 (95% CI, 0.48 - 0.92) [30].

Based on the analysis of the RUST score (Radiological Union Scale for Tibia) on vitamin C supplementation, there was a statistically significant difference at week four. The higher the dose of vitamin C supplementation will provide better fracture healing, at the 20 mg supplementation dose of vitamin C has the greatest effect on the RUST score (Table 1). The RUST score is a score that is more valid and reliable when compared to conventional examinations [31].

The potential of PLGA as Nano-encapsulation Hesperetin and Vitamin C in the Healing of Fragility Fractures.

Hesperetin and Vitamin C in treating fractures in osteoporosis require drug delivery, so as to maximize its potential. Among the copolymers, PLGA emerged as a biocompatible and non-toxic polymer which is important in drug delivery, medical devices, tissue engineering, and surgical instruments [32-33]. PLGA has been approved by the US FDA in several therapeutic applications because of its sustained-release properties, biodegradability, and biocompatibility [34]. PLGA can prevent rapid clearance of the body, has a specific affinity for the cell surface and receptors to provide pinocytic uptake by target cells. It is good for selective targeting, non-immunogenicity, stable during drug release period, and biodegradable without leaving toxic residues [35]. In addition, PLGA with nanoparticles can enhance the calcium deposition process through osteoblasts [14].

PLGA nanoparticles or microspheres are connected to ligands target such as cytokines, vaccines, hormones, and chemotherapeutic agents [36]. PLGA is used to encapsulate drugs, thereby extending the drug release time. In Wang's study using different flavonoids, the use of nHAC / PLGA as nanoencapsulation can increase the half-life of these flavonoid compounds. The addition of PLGA can extend the release of nHAC / PLGA in the first 24 hours when compared to nHAC alone. The release of nHAC alone within 24 hours was obtained as much as 93% of the compound released, whereas in the use of nHAC / PLGA within 24 hours the percentage of release became 46% which was followed by constant release for 5 days [37]. Seeing the potential of PLGA as a nano-encapsulation in increasing the half-life of drugs, it can support the delivery of hesperetin and vitamin C in overcoming fragility fractures in osteoporosis patients. Apart from being able to treat fragility fractures in osteoporosis, these three elements give new hope to osteoporosis management. With the combination of the flavonoid hesperetin and vitamin C based on PLGA, it can make managing osteoporosis simpler. This can be estimated from the half-life of the substance which can be extended by the PLGA.

Conclusion

Hesperetin and PLGA-based vitamin C can be a promising therapeutic modality in healing fragility fractures in osteoporosis patients, especially in the process of bone matrix formation in the fracture healing process. A number of studies on hesperetin and vitamin C and their effect on bones can prevent and improve fragility fractures, besides that these two substances are easily found in citrus fruit. With PLGA nanoparticle-based administration, it can increase the potential of these two substances to reach specific targets and extend the half-life of hesperetin and vitamin C. However, these studies still have limitations in terms of the form of research that is still in thestages. in vitro and in vivo. Therefore the development of further research, both experimental and clinical trials regarding hesperetin PLGA-based and vitamin C needs to be carried out.

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