The Effect of Natural Products Extract on Stability of Orthodontic Implant an Experimental Study

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Abstract-This study was conducted to estimate any possible effect of the natural products extract on the stability of orthodontic implant. Methods: twenty five white mature male rabbits, classified into 5 groups one control and four experimental, with five rabbits each group, were used in this study, fifteen orthodontic implants were inserted in right tibia of each rabbit, with two implants in each tibia. Four different natural products extract were used in this study include; Curcumin 15mg/kg, Nigella Sativa oil 0.5 ml/kg, Cissus Quadrangularis 1000mg/kg and Virgin coconut oil 2 ml/kg. Each product given to each experimental group start from the day of implant insertion for four weeks healing period, the implant stability tested at the time of implant inserted (primary stability) and again after four weeks (secondary stability) using periostet machine. Results: the results of this study showed a significant differences between primary and secondary stability for all groups (control and experimental) except for Nigella Sativa. A significant difference detected on comparing secondary stability between Curcumin and control group. Conclusion: Curcumin, Cissus Quadrangularis, Virgin Coconut oil and Nigella Sativa could be used for possibly increasing the stability of orthodontic implant.

Index Term-Natural products, Orthodontic implant, Stability.

1 INTRODUCTION
Anchorage control is necessary during orthodontic treatment to control the reactive forces and prevent negative side effect, therefore a stable anchorage unit is necessary [1]. In orthodontic treatment, the role of anchorage was appreciated early on, Angle as prominent orthodontists realized the limitations of anchorage was appreciated early on, Angle as possible and spread wide with mini-implants which in the last few years, anchorage reinforcement become possible and spread wide with mini-implants which proved in various clinical situation to provide reliable anchorage [3], [4]. In 1984 the successful use of loaded titanium screws placed in rabbit femurs by Roberts et al [5]. Successful orthodontic treatment is possible by achieving implant stability and its maintenance throughout the treatment. The most significant factor for survival rate and reliability of implant is the primary stability Javed et al [6]. The primary stability explained by mechanical interlock between bone and implant [7]. The ability of bone remodeling and healing as time progress is more significant to secondary stability [8]. Park et al [9] stated that establishing the primary stability and maintaining it, through healing (secondary stability), is very important factor for orthodontic implant success. Great primary stability is necessary to resist even micro-movement which if present could lead to micro fracture, necrosis, bone resorption which end with implant loss [10]. In addition with implant insertion there will be circumferential stress on adjacent bone to establish intimate interlocking with adjacent bone as possible. But if this stress is much, as insertion of self drilling screw in dense bone, could lead to excessive micro-crack and necrosis which possibly could lead to failure of implant [10], [11]. Despite all the advantages of orthodontic implant, failure could happened which may be a result of number of factors like: host factors which include bone quality and bone quantity [12], implant design and surgical factors [9], [13]. Motoyoshi et al [14] postulated that in patients with poor bone quality, immediate loading of implant is prone to failure. A number of natural product were used before, for controlling bone metabolism in an attempt to promote anabolic effect or limiting/suppressing the catabolism of bone, these are Vitamine E add to natural herbal products such as Eurycoma longifolia, Labisia pumila, virgin coconut oil, Cissus Quadrangularis, Cosmos caudatus, curcumin, Nigella sativa, Sambucus nigra, and Trifolium sp. These were mainly extracts from the whole plant or specific parts of the plants, such as the fruit, leaves, or roots. However, further studies are needed to confirm these effects [15]. Therefore the purpose of this study is to test the effect of four natural product extracts include "Curcumin, Nigella Sativa, Cissus Quadrangularis and Virgin coconut oil” on implant stability through biomechanical test.

PROCEDURES
Fifty sterile Orthodontic implants, diameter 1.3mm and length 5mm (Dentos, AbsoAnchor, SH1312-5/ tapered type, Dentos Inc. 1-5, Galsan-Dong, Dalseo-Gu, Daegu, Korea 704-900) were made with Ti-6Al-4V alloy. Twenty five healthy mature male rabbits weight 2-2.25kilogram, 7-10months-old, were used. These rabbits were divided into 5 groups, each group 5 rabbits: first group was control (Co) that subject to implant insertion but not given natural products, the remaining four groups were experimental that were
subjected to implantation and given natural products (Figure 1).

Figure (1) The four natural products used.

The implantation procedures were common to all animals and consisted of placement of 2 implants 10mm between each other in medial surface of the right tibia of each animal. All operations were performed under sterile conditions in a certain operating room, prepared for this purpose. Immediately before the surgery, the animals were anesthetized with intramuscular injection of 0.2 ml/kg b.w. ketamine 10% (KEPRO B.V.-Maagdenburgstraat 38-7421 ZE Deventer- Holland) and 0.15 ml/kg b.w. Xylazine 2% (alfasan. Woerden-Holland).

The surgical area anaesthetized with 1ml local anesthetic solution 2% xylocain. The hair on the medial surface of the right leg clipped and the skin wiped thoroughly with Betadine solution (10% povidone iodine topical solution, purdue products L.P., Stamford) with a sterile surgical gauze. An incision approximately 20 mm in length down parallel to longitudinal axis of the tibia, in the medial aspect (figure 2).

Figure (2) Soft tissue incision and bone exposure.

The skin, fascia and muscles were dissected then periosteum was stripped and elevated denuding the bone in medial aspect of tibia. Implants holes drilled with a 1 mm rounded drill under profuse sterile saline-solution irrigation and at low rotary speed (figure 3).

Figure (3) Implant hole preparation in tibia.

The implants threaded in the tibia with a manual driver (figure 4) keeping their longitudinal axes parallel to each other and perpendicular to the external cortical tibia with 10mm distance in between (figure 5).

Figure (4) implant insertion in tibia.

The implant were then tested for their stability (figure 6) using the periotest machine (Medizintechnik Gulden e.k. Eschenweg 3, 64397 Modautal, Germany). The Periotest machine was designed by Schulte et al [16] and d’Hoedt et al [17].

Figure (5) two implants inserted with 10mm distance in between

Figure (6) Stability testing with periotest
The hand piece of the machine was oriented parallel to the floor before being activated. Each Periotest was calibrated before each use using the calibration sleeve provided with the unit [18]. The periotest scale extends from -8 to +50, the lower the value, the greater is the stability with the periotest value range from -8 to 0 this mean a good osseointegration and implant can be loaded, +1 to +9 values mean clinical examination is required and loading is not yet possible, more than +9 mean that the implant must not be loaded. The peristium and deep fascia repositioned in their original place and the skin sutured (figure 7) with black silk suture (three, 000).

Figure (7) soft tissue closure and suturing.

The rabbit was then placed in postoperative recovery. Once fully recovered, the rabbits were placed in their respective holding areas. Then the natural products loaded as follow:
Curcumin (Cu) for first group involve one capsule of curcumin "CurcuVET-SA150/ Curcumin complexed with phosphatidylcholine for superior bioavailability/ pure ingredient" (Throne Research, Inc. p.o. Box 25 Dover, ID 83825 USA) which contain 30mg of curcumin for each rabbit By dissolution of capsule content in distilled water and given directly through oral gavages”.
Nigella Sativa oil (NSO) for the second one "Pure cold pressed 100ml Black Seed oil from Nigella sativa seeds 0.50 ml/ Kilogram (kg) body weight (b.w.)" (The Blessed Seed Oil/ Black seed oil specialist/ Beverley, United Kingdom), given freshly through oral gavages. Cissus Quadrangularis (CQ) for the third group 1000mg/ kg b.w. *100% pure organic Cissus Quadrangularis extract by Keter Wellness/ under strict of good manufacturing practice (GMP) guidelines/ United State of America* (capsule weight 1gm) By dissolution of capsule content in distilled water.
Virgin Coconut oil (VCO) for the fourth group 2 ml/ kg b.w. Organic Pure Raw (Live superfoods, 20739, High Desert Court Bend, Oregon 97701).

All of these natural products given as a loading dose for 4 weeks, started from the day of implant insertion. After 4 weeks healing periods all of the rabbits were sacrificed in the same place specially prepared under same circumstances. The stability again tested with periotest machine with the end result are two reading of stability, immediate after implant insertion and after 4 weeks healing period. All of the procedures were performed by 1 operator to reduce potential errors.

RESULT
The result of this study showed that on comparing primary and secondary stability for all groups (Table 2), a statistically significant differences, higher for secondary stability, were achieved for all groups except the NS group. NS group showed a difference between primary and secondary stability, on referring to the mean values (Table 1), although statistically not significant. For control group a statistically significant differences had been achieved but in opposite direction that higher for primary stability.

<table>
<thead>
<tr>
<th>Group</th>
<th>Primary Stability (PS)</th>
<th>Secondary Stability (SS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Co PS</td>
<td>3.7778</td>
<td>.66667</td>
</tr>
<tr>
<td>Co SS</td>
<td>2.5556</td>
<td>1.13039</td>
</tr>
<tr>
<td>Cu PS</td>
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<tr>
<td>Cu SS</td>
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<tr>
<td>CQ PS</td>
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<td>.70711</td>
</tr>
<tr>
<td>CQ SS</td>
<td>3.5000</td>
<td>1.08012</td>
</tr>
<tr>
<td>NSO PS</td>
<td>3.0000</td>
<td>.86603</td>
</tr>
<tr>
<td>NSO SS</td>
<td>3.2222</td>
<td>1.09291</td>
</tr>
<tr>
<td>VCO PS</td>
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<td>VCO SS</td>
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<td>1.39841</td>
</tr>
<tr>
<td>VAR00001</td>
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<td>1.42857</td>
</tr>
</tbody>
</table>

On comparing the secondary stability between the experimental groups and the control one, a statistically significant differences detected only for Cu group with control one (Table 3).
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STABILITY FOR ALL GROUPS

tried to use these natural products to test their possible structure, and strength [15]. Here, in this study, we bone remodeling, effects on bone metabolism, products previously evaluated for their involvement in clinical situation. In assessing stability, periotest could Although such studies do not exactly replicate the different from previous [15], due to the traumatic injury, inflammation and bone response "pathologic remodeling" [23], [24] that formed around the implant. Inaba in 2009 [28] stated that "the bone-implant interface". Olivé and Aparicio in 1990 [29] reported that PTV, for dental implant, above than +10 consider failure of implant. All of our study measurement were under this value (Table 1).

Our study results showed a significant differences between primary and secondary stability, with improvement in stability for implants in Curcumin, Cissus Quadrangularis and VCO as represented by higher PTV for secondary stability (Table 2). This probably be due to bone anabolic or anti catabolic activity of these products. An in vitro studies [30], [31], [32] showed that Curcumin could stop osteoclastogenesis through suppression of RANKL activity completely, another in vivo studies [33], [34], [35], [36], [37] showed that using Curcumin could limit bone loss, improve remodeling and improve bone strength in osteoporosis animal models. For Cissus Quadrangularis an in vitro studies [38], [39], [40] showed its ability in promoting osteoblast proliferation and differentiation and as a potent antiosteoporotic substance in osteoporotic animal model [41], [42], [43]. Add to promote healing of bone fracture [44]. These added to anti-oxidative property of VCO in prevention of bone loss and preserving bone structure [45], [46].

For control group higher PTV for primary stability (Table 2). Cho in 2006 [47] explained that once the OMIs is inserted the bone will subjected to stress and fatigue, this micro-strain is a result of difference in modulus of elasticity between the titanium screw and the bone material, which result in micro-fracture. This surgical trauma initiate healing response which includes the formation of a blood clot, inflammation, migration and differentiation of cells, formation of granulation tissue and finally remodeling process "pathologic remodeling" this possible be the probable cause of the lowering the secondary stability compared to primary stability in control group, while for experimental groups same event also happened but the different results possibly be the cause of biological activities, at osseous level, of natural products mentioned above.

On comparing secondary stability between each experimental and control group (Table 3), higher PTV detected for all experimental groups, with a significant result for Curcumin this could come in accordance with other studies [33], [34], [35], [36], [37].

**CONCLUSION**

On conclusion, according to the results achieved, the natural products mentioned above could be used possibly for increasing the bone implant integration expressed by implant stability. Further the best of these used natural products is Curcumin.

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