STABILITY AND THREE-DIMENSIONAL ANALYSIS OF BONE FORMATION IN LONGITUDINALLY FLUTED MINISCREW IMPLANTS

A Thesis

by

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ABSTRACT

The purpose of the present study is to evaluate the effects of longitudinal flutes on mini-screw implant (MSI) bone healing and stability. Using 11 skeletally mature New Zealand White rabbits, 33 longitudinally fluted and 33 non-fluted MSIs were placed and immediately loaded with 100g using NiTi coil springs. Insertion torque values were obtained for each MSI that was placed; removal torque values were obtained for 28 MSIs that had been in place for 6 weeks and 20 MSIs that had been in place for 2 weeks. The bone volume fraction surrounding the implant 6-to-24 μm, 24-to-42 μm, and 42-60 μm from the MSI surface using micro-computed tomography with an isotropic resolution of 6 μm. The success rate was 97%, with both the fluted and non-fluted MSIs each having one failure. Mean insertion torque was slightly higher for fluted MSIs (3.98 N cm ±0.24) compared to non-fluted MSIs (3.95 N cm ±0.24), but the difference was not statistically significant ($p=.930$). After 6 weeks, removal torque values were significantly ($p=.008$) higher for the fluted (3.42 N cm ±0.26) than non-fluted (2.49 N cm ±0.20) MSIs. After 2 weeks, removal torque values were higher for fluted (2.87 N cm ±0.22) than non-fluted MSIs (2.75 N cm ±0.22), but the difference was not statistically significant ($p=.702$). After six weeks, bone volumes of the 6-24 μm, 24-to-42 μm, and 42-to-60 μm layers were significantly ($p<.05$) greater for the non-fluted than fluted MSIs. After two weeks, bone volume of three layers were also significantly ($p<.05$) greater for the non-fluted than fluted MSIs. Fluted and non-fluted 3 mm long MSIs can have very high success rates, even with all maximum insertion torque values

being less than 6.2 N cm. Adding longitudinal flutes to 3 mm MSIs increases their removal torque by 37% after 6 weeks, despite the fact that there was less bone surrounding the fluted than non-fluted MSIs.

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CHAPTER I

INTRODUCTION AND LITERATURE REVIEW

The advent of mini-screw implants (MSIs) has provided orthodontists with useful tools for achieving increased or maximum anchorage. Anchorage allows desired tooth movement without the side effects of unwanted movement of other teeth. Traditional orthodontic mechanics have relied on control of the reactive unit tooth/teeth acting as anchorage against active unit) through the use of extra-oral reinforcement, intra-arch or inter-arch mechanics.

Newton's third law states that, for every action there is an equal and opposite reaction. With this law in mind, none of the traditional methods to control the reactive unit in orthodontics can provide absolute anchorage. Extra-oral, intra-arch and inter-arch mechanics have been used for years to reinforce anchorage units, but these mechanics can be cumbersome and subject to compliancy issues.

Miniscrew implants have gained rapid popularity and acceptance due to their versatility, minimal invasiveness, low costs, and patient acceptance.[1-4] They are particularly advantageous when extraoral appliances are impractical and patient compliance is unreliable.[1, 4] Although MSIs provide clinicians with an important mechanical tool, the concern for implant failure remains a main deterrent for many orthodontists.[5] Several studies have been performed in an effort to determine ways of increasing MSI success rates including modifications to MSI design, shape and surface treatments. One study in particular showed that adding longitudinal flutes was able to increase pullout strength by approximately 60%, however this study was performed on cadaver bone and no live model has yet to be tested.[6] Additional research needs to be performed to evaluate the intimate bone growth areas around an MSI and the resulting effect of fluting as part of MSI design.

The purpose of the present study is to evaluate the effects of longitudinal flutes on MSI bone healing and stability through analysis using micro-computed tomography (μCT), insertion, and removal torque values.

The following review will discuss the history of MSI use and development, MSI design, and bone healing. The review will first cover the history, clinical applications and success rates of MSIs in order to better appreciate the importance and value of MSIs in orthodontics and the need to continue studying and improving their clinical outcomes. Certain design aspects will be reviewed to better understand the benefits of a reduced length but equal stability. In addition, important aspects of bone healing will be discussed, including principles of primary and secondary stability, as well as methods of measuring implant stability and micro-computed tomography analysis of MSIs.

MSI History and Clinical Applications

The initial reported attempt to place an implant for stable orthodontic anchorage was in 1945 by Gainsforth and Higley, who used vitallium screws.[7] The study used six dogs with modified vitallium screws 13 mm in length placed in the ramus for retraction of the canines. Gainsforth and Higley were successful in producing tooth movement with basal bone anchorage, but effective orthodontic forces could not be sustained for more than 31 days due to implant failure. They attributed implant failure to the close association with the oral cavity and resulting contamination with oral pathogens. They also speculated that failure was due to the rejection of the vitallium metal as a foreign body, mitigating a localized immune reaction and unfavorable supporting bony changes.

In the 1960's, Linkow used blade implants to retract teeth with rubber bands.[8] Small endosseous implants were also utilized as orthodontic anchorage when they were placed in the retromolar pad area and palate to aid in the closure of extraction spaces.[9, 10] Other studies also report the use of palatal onplants and miniplates in attempts to create absolute skeletal anchorage but these devices gained minimal popularity with orthodontists due to their large and complicated nature as well as the necessity for a more invasive surgery.[11, 12]

One of the first clinical reports of screw usage for orthodontic anchorage was by Creekmore and Eklund in 1983.[13] In the study, Creekmore and Eklund placed a 13 mm surgical vitallium bone screw immediately below the anterior nasal spine and used it for anchorage in correcting a severe deepbite. Following this initial report, many clinical studies were performed using endosseous implants for orthodontic purposes, with various degrees of success.[14-16] The use of endosseous dental implants for absolute anchorage was promising but several disadvantages were evident. The larger size, surgical procedures for placement and removal, cost, and delayed loading times prevented the mainstream use of endosseous implants as orthodontic anchorage. These limitations led Kanomi et al to pioneer smaller implants specifically for orthodontic usage.[17] Originally designed for fixation in bone plates in craniofacial reconstructive surgeries, the miniscrews were 1.2 mm wide in diameter and 6 mm long. These miniscrews were designed to be small enough to be placed in between the roots of the mandibular central incisors for successful intrusion of the anterior dentition over four months. They suggested the surgical procedure should be easy enough for an orthodontist or general dentist to perform and minor enough for rapid healing and easy removal once treatment was finished.[17] The progression to a smaller size, minimal surgical invasiveness and relative affordability allowed for greater versatility and practicality in orthodontic applications.

It wasn't until 1998, when Costa and his associates introduced a simplified MSI placement protocol, that MSIs became widely accepted in orthodontics.[18] MSIs have gained significant popularity and numerous orthodontic manufacturing companies are now producing MSIs specifically for orthodontic purposes. In 2008, a survey of AAO members revealed that 54.4% of orthodontists had placed MSIs.[5] A more recent survey in 2010 of orthodontists in the northwest United States revealed that approximately 91% of orthodontists had experience placing MSIs.[19]

A number of published reports highlight successful treatment outcomes with the use of orthodontic MSIs.[20, 21] In a randomized controlled trial of forty patients with bialveolar dental protrusion that underwent extraction of all four first premolars, Upadhyay et al compared the retraction of anterior teeth using conventional anchorage compared to en-massed retraction using pre-drilled orthodontic MSIs. These MSIs measured 1.3 mm in diameter and 8 mm in length and were placed between the first molars and second premolars in all 4 quadrants. The orthodontic MSIs prevented anchorage loss and significantly reduced facial vertical dimension.[21] MSIs have steadily become more practical and are now clearly a useful tool in orthodontics. It is important to understand and explore the various ways we can continue to improve their success and clinical applications.

MSI Success Rates

Endosseous implants have an approximate success rate of 97%, while the success rates of mini-implants have varied depending on the study. Schatzle et al in 2009 produced a systematic review of MSIs analyzing 390 articles and 71 abstracts with full-text analysis of 34 articles, and found a total of 27 studies that met the inclusion criteria. The study found a success rate of 83.6% with a 95% confidence interval of 86.6%-79.9%.[22] A meta-analysis by Papageorgiou et al in 2012 evaluated 52 studies for a total of 4987 MSIs in 2281 patients. The overall success rate was 86.5% with a 95% confidence interval of 88.5%-84.2%.[23] Another systematic review of the literature by Reynders et al in 2009 searched and discovered 19 reports that met their inclusion criteria. Rates of success rate outcomes from these studies ranged from 0% to 100% but most articles' success rates were cited as greater than 80% if displaced and mobile MSIs were included as successful.[24] In the study by Cheng et al. (2004), potential risk factors associated with mini-screw failure were studied. Cheng looked at 140 mini-screws in 44 patients, including 48 miniplates and 92 freestanding miniscrews with various orthodontic loading forces. The study identified soft tissue character and anatomic location as significant factors associated with peri-implant infection and failure. Other investigators have also suggested that using excessive forces on mini screws, potential for peri-implantitis when inserted in unattached mucosa, insufficient primary stability, and bone damage on insertion (frictional necrosis) can all contribute to orthodontic mini screw failures.[25] The failure to achieve the same success as observed with endosseous implants has prompted many studies to evaluate different physical designs of MSIs and their effects on stability. As MSIs gain general acceptance and are continually utilized in orthodontic treatment plans, it is important to understand and improve the designs to reduce failures and subsequently increase clinical predictability.

Osseointegration

Since the development of endosseous implants for the use of restoring missing teeth, there has been a gradual progression toward the improvement and understanding of implant design and the subsequent surrounding bone response. The process of osteointegration, which was defined by Branemark as a direct – on the light microscopic level – contact between living bone and implant, is essential for the prognosis of implant success or failure.[26] It wasn't until the accidental discovery that titanium could form an intimate biocompatible relationship with surrounding living bone that implants were considered viable for absolute anchorage.[26] Further long-term evaluation of functional osseointegrated implants in man was investigated by Albrektsson et al., who removed 38 stable, integrated, titanium implants to evaluate the interface between bone and implant through X-ray, SEM, TEM and histology. [27] In an early 1981 study by Albrektsson et al, osseointegration was determined to be dependent on the following parameters: 1. Implant material; 2. Implant design; 3. Implant finish; 4. Status of the bone; 5. Surgical technique; 6. Implant loading conditions.[27] The study proved preliminary evidence of direct contact between titanium and bone microscopically.

Bone Healing

The events of bone healing following implant placement appear to be the same as osseous wound healing in normal bony injury. Initial stages of endosseous wound healing can be divided into stages of hematoma, clot resolution, and osteogenic cell migration, which subsequently leads to new bone formation at the wound site.[28] In describing endosseous peri-implant bone healing, Davies in 1998 described three distinct phases: osteoconduction, de novo bone formation, and bone remodeling.[29] Occasionally, authors divide the osteoconduction phase into two related but distinct processes: osteoinduction, the phenotypic conversion of mesenchymal cells into bone forming cells, and osteoconduction, the appositional bone growth permitting bone formation on a surface.[30] Osteoinduction is part of normal bone healing and is responsible for the majority of newly formed bone. De novo bone formation is the formation of a mineralized interfacial matrix equivalent to that found in the cement line in natural bone tissue. Through osteoconduction and de novo bone formation with an appropriate implant surface, adequate bone formation may be formed around an implant surface through contact osteogenesis.[28] The third healing phase, bone remodeling, generally relies on slower processes. In a study by Berglundh et al, 20 Labrador dogs received 160 experimental devices and histologic cross-sections were evaluation between 2 hours and 12 weeks to observe healing. The study showed that as early as 1 week, a provisional matrix with newly formed woven bone was observed around most vascular units. At the 2-week interval, new bone formation was found in all compartments around the device and osteoblasts lined the implant walls. By 4 weeks, continued bone formation occurs and bone remodeling is observed around the screw threads.[31]

Primary Stability and Secondary Stability

It is important to understand that although primary and secondary stability are often described as two unique processes, they are co-dependent phenomena. It is only possible for osseointegration of an MSI to occur if it is stable at placement, also known as primary stability or the initial mechanical stability. According to Miyawaki et al 2003, one of the most critical risk factors in MSI failure is relative movement between the implant body and the adjacent surrounding bone during early wound healing.[32] Any micromotion early after initial MSI placement leads to fibrous repair of peri-implant bone, which eventually impairs the proper osseointegration process.[33]

Immediately after insertion of a miniscrew implant, retention is purely mechanical and depends on how the threads cut into the bone. Bone contact at insertion, or mechanical macroretention, is responsible for the initial primary stability and largely important in miniscrew endosseous implant success since there is not the same requirement for long-term stability and full osseointegration as in dental implants.[34] There are multiple factors that influence the primary implant fixation stability including: implant geometry, preparation technique, and quality and quantity of regional bone.[35] Clinical and basic research of endosseous implants indicates that stability at the time of surgery is important for therapeutic success.[36-39] Cortical density, in particular, is thought to be an important factor for primary stability in miniscrew implants.[34, 40] During initial healing, osteoclastic activity reduces the initial mechanical stability of an implant while formation of de novo bone will lead to secondary stability.[41] This secondary stability stage includes the deposition of bone along and intimately between implant threads, and thus is responsible for bone-to-implant contact and osseointegration. Adequate primary stability is the main factor that determines secondary stability. Insufficient stability facilitates the formation of fibrous tissue rather than proper bone formation at the implant surface.[42, 43]

Host Factors

The regenerative and healing potential for the body to remodel and deposit bone are critical to develop long-term implant stability. Some known factors related to decreasing MSI stability include: lack of primary stability at placement, peri-implant inflammation, absence of keratinized tissue, thin cortical bone, bone density, and bone quality.[32, 40, 44] Smoking, osteoporosis, uncontrolled diabetes, parafunctional habits and poor oral hygiene have also been linked to decreased implant stability and implant failure.[44, 45] Implant failure is often caused by inflammation around the peri-implant surfaces leading to implant mobility and inhibition of proper osseointegration. Oral hygiene has been shown to play a critical role in minimizing inflammation and reducing implant failures.[44, 45] In addition, the lack of proper hygiene has been suggested as an important factor relating to MSI success rate.[32]

Methods of Testing Osseointegration

Several methods for the evaluation of osseointegration have been used to assess bony-implant adaptation and to test various implant designs and materials. The three main areas of testing are histomorphometric evaluation of peri-implant tissue at the cellular level, biomechanic evaluation of actual physical forces required to dislodge a MSI from surrounding bone, and bone volume analysis using micro-computed tomography. One of the big limitations of biomechanic evaluation measuring physical forces and histomorphometric evaluation are that they require destruction of the bone-implant specimen.

Insertion Torque

Insertion torque has often been associated to be an indirect measure of primary stability of a screw. In a study by Motoyoshi et al (2006), 124 orthodontic MSIs were examined for implant placement torque. The authors found that an ideal implant placement torque in the range of 5 N cm to 10 N cm was ideal for this specific miniscrew, yielding an overall success rate of 96.2%. They also observed a decrease in success rates with insertion placement torque values below 5 N cm and even greater significant decreases when values increased beyond 10 N cm. Motoyoshi et al concluded that a lower insertion torque is suggestive of poor primary stability and potential failure of an orthodontic miniscrew. Conversely, a very high insertion placement torque places significant stresses on the surrounding bone leading to bone degradation or frictional necrosis.[46] Another study places the validity of insertion placement torque as an indirect measure for primary stability into question. A study by Degidi et al, (2009) examined the insertion torque of seventeen endosseous dental implants of various manufacturers removed from patients for a variety of reasons. Although the study design was flawed and was of limited quality, the authors concluded that there was no statistically significant correlation between insertion torque and bone-implant contact (p=0.892). The authors concluded two possible reasons for the lack of correlation: primary stability is not only influenced by bone volume, but also by density and thickness of the cortical layer; or that there is no true relationship between bone structure and insertion torque values.[47]

Pull-Out Testing

Pull-out testing of implants are used to test the shear failure load of bone during the application of a tensile force on the implant. Application of a force is placed along the long axis of the implant, typically with an Instron machine, while the peak force is recorded prior to failure.[40] Pull-out studies commonly utilize cylindrical or press-fit implants to eliminate any variability associated with the threads.[40] These tests require very accurate orientation of the implant with the direction of pull to avoid any unwanted directions of force.[40] Also, like any biomechanical method of implant testing, pull-out testing requires destruction of the bone-implant specimen and can require exact application of force vectors to achieve accurate results. Various small changes, such as hand position causing tipping or change in distance from the jig, can influence reading measurements.

Removal Torque

Torque removal testing systems are usually performed with a torque gauge instrument connected to an implant bone-specimen. Removal torque is generally the indirect measure of the shear force required to fracture the bone-implant surface,[48] and may be good indicator of implant stability.[49] However due to the flawed nature of hand-held devices used for torque measurements, variable forces such as tipping are not singularly isolated along the long axis of the implant and can influence removal force registered.[50] The implant thread design and apical self-tapping features can also influence torque removal test results.[50] Generally, biomechanical tests evaluating osseointegration are based upon the amount of force needed to achieve sheer separation of peri-implant bone from the implant surface. The stability and bone integration are indirectly inferred from these measurements without regards to the quantity, location and type of adherence at the bone-implant interface unless complementary analysis are performed. Quality of bone integration is then inferred from the amount of force necessary for separation; no actual evaluation of osseointegration or bone quality takes place with these biomechanical evaluations.

Histomorphometric Analysis

Histomorphometric analysis is a method used to accurately quantify the cellular activity and the amount of existing bone mass in the surrounding environments. Light microscopy of thin histological sections has made qualitative and quantitative analysis of bone-implant interface possible.[51] In a study by Wigianto et al in 1997, a three-dimensional image of bone structure around an implant was constructed through serial slices of two-dimensional images histological slides.[52] The construction of a three-dimensional image using serial two-dimensional sections provided limited information and the models remain largely incomplete.[53] In addition to the lack of full three-dimensional rendering, this histologic preparation prevents the specimen from being further utilized with other analysis.[53] Preparation procedures for histomorphometric analysis can potentially cause artifacts through excess grinding or detachment or disruption of structures. Although histomorphometric analysis is considered as the gold standard for peri-implant tissue evaluation, drawbacks such as time consuming preparation of ground sections and visualizing a three-dimensional structure in only two-dimensions provide some limitations in its use.[54, 55] Another limitation of histomorphometric analysis is the destruction of the bone implant specimen, however despite these drawbacks histomorphometry remains a valuable method of measurement and is a commonly used tool to evaluate bone-to-implant contact.[31, 56]

Micro-Computed Tomography

The gold standard for the evaluation of bone-implant contact has been histomorphometric analysis since the earliest reports of osseointegration and titanium implants.[26] Micro-computed tomography (μCT) allows for high resolution three-dimensional images with the ability to take quantitative and qualitative measurements of surrounding bone.[57] Several studies have compared the accuracy of μCT in bone specimens with that of the same evaluation using histomorphometric analysis and have shown high accuracy and high correlations between the two.[58, 59] In a study by Muller et al, bone excised from the left radius of a 46-year-old healthy man was selected for the measurement and analysis using three-dimensional CT. These scans were then compared and validated with two-dimensional morphometric analysis and were found to differ by only a factor of 0.9-1.1 when comparing the three-dimensional CT and histologic sections. Another study by Kuhn et al also evaluated micro-CT as a tool for nondestructive analysis of bone by comparing 6-mm trabecular bone cubes assessed by micro-CT and histologic sections to determine accuracy of representation. The results suggested that the micro-CT measures of bone volume fraction were not significantly different from those obtained histologic sections measurements and therefore were very accurate. Measures of trabecular plate density differed by approximately 14%, which was the equivalent to discrepancies of about 19 microns in trabecular plate thicknesses. One reservation about µCT in assessing intimate bone and implant space is because of the ionizing radiation similar to conventional computed tomography, it is possible to have missing or distorted data due to metallic artifacts.[60] Zhao describes the three main causes of artifacts due to metallic objects reported are: 1) beam hardening by x-ray spectrum dispersion 2) photon starvation and poor signal to noise ratio 3) enhanced motion interference between the metal and adjacent structure caused by high contrast.[60] Rebaudi et al in 2004 scanned a cylindrical biopsy of trabecular bone containing a 2 mm wide and 5 mm long titanium implant retrieved 12 months after implantation in a human maxilla.[54] The sample was analyzed using a 15 µm resolution µCT scanner and subsequently was evaluated using histomorphometric analyses. The comparison between µCT and the histomorphometric analyses produced similar results in terms of bone implant contact. However, bone implant contact measure through histomorphemtric analysis were slightly higher than those obtained through µCT. Rebaudi designated an arbitrary boundary of 45 µm to account for the titanium artifacts, and used this border in determining BIC. For this study, bone implant contact was defined as "bone in a 45 µm neighborhood of the titanium surface". This 45 µm boundary could have obscured some of the bone apposition to the implant surface during measurements with µCT. However, since the images were scanned at a high resolution of 15 μm, Rebaudi et al postulated that sophisticated image processing techniques could be applied to further alleviate the titanium metal artifacts. In another study by Stoppie et al, 3.5 mm wide and 13 mm long titanium implant screws were placed in the femoral condyles of female Saanen goats.[61] The implant and surrounding tissue samples were retrieved after 6 weeks and scanned with a resolution of 24 μm. Histomorphometric analysis was also done on the samples and was subsequently digitized and overlayed against identical μCT sections. Histomorphometric analysis compared to μCT of these titanium implant specimen showed an average correlation of 89%. However according to the Stoppie et al, due to a thin layer of noise around the implant (60 μm) present along the entire implant surface, bone implant contact was not accurately measured.

Improvements to scanning equipment and software have further refined resolution limits. In a study by Butz et al, unthreaded dual acid-etched pure titanium cylindrical implants 1.0 mm in diameter and 2.0 mm in length were placed in the femur of rats for 2 weeks.[55] Scanning resolution was 8 μm and observed correlations between μCT and histomorphometric analysis were significant in cortical bone (r=0.65) and trabecular bone (r=0.92) at distances of 24 to 240 μm from the implants surface. An observation from this study was that there was not a significant correlation from 0 to 24 μm from the implant and the mean bone volume reported from the μCT was 2-3 times higher than that shown with histology. The bone observed at 0 to 24 μm was considered to be visualized poorly due to noise attributed to metallic halation artifacts.

During Micro CT scanning, metallic objects absorb and scatter x-ray energy at various rates, which can often cause halation artifacts; a phenomenon also known as "partial volume effect." It is found that by producing narrower slices through a higher resolution, the artifacts are substantially reduced. The presence the artifacts can be attributed to axial partial volume effects. Axial partial volume effects occur when the object has axial variations at any point in the slice. In these situations, the determined algorithm of integrated intensity measured it not a linear function of the integrated attenuation. Subsequently, this nonlinearity causes irregularities in the data set, which can result in streaks produced in the image acquired.[21] As seen in the study by Butz et al, halation artifacts have the potential to produce an overestimate of bone volume fraction when directly compared to histomorphometric measurements.

In a study by Ikeda et al in 2011, SLA surface miniscrews and machine-surfaced miniscrews were compared using micro-computed tomography at 6 µm resolution to assess bone volume to total volume in the peri-implant space.[62] After correcting for potential metallic artifacts, Ikeda et al were able to accurately visualize and quantify bone in cortical and trabecular areas by analyzing the bone volume to total volume from regions 6 to 24 µm and 24 to 42 µm; the region from 0 to 6 µm was disregarded because of potential metallic effects. Another study in 2012 by Massey et al used micro-CT to assess the effect of force on alveolar bone surrounding MSIs. This study also analyzed bone volume fractions surrounding MSIs and calculated this for layer regions of 6 to 24 µm, 24 to 42 µm, and 42 to 60 µm; the region from 0 to 6 µm was also not assessed due to potential metallic effects.[63]

MSI Design

Miyamoto et al determined that initial stability was influenced more by cortical bone thickness than by implant length. [64] Studies have shown the diameter of the screw was significantly associated with its stability.[32, 65] A study evaluated the external diameter of MSIs compared 6 mm – 1.75 mm diameter MSIs , 3 mm – 1.75 mm diameter, and 3 mm – 2 mm diameter MSIs placed in synthetic bone. Using insertion torque and pullout strengths, the 0.25 mm wider diameter MSI showed increased insertion and pullout strengths of 12%-14% and 13%-21% respectively when compared to the 3 mm – 1.75 mm diameter screws.[65] In a study evaluating the shape and taper of MSIs, twenty-eight MSIs were placed in the leg of New Zealand White rabbits differing in diameter and shapes. Stability was measured with maximum insertion torque for the 4 classifications of screws; 1.5C, 2C, 1.5T, and 2T, denoting the diameter of the screw and whether it was cylindrical or tapered in shape. Lee et al found statistically significant increase in maximum insertion torque for increased diameter and the tapered shape designs.[66] Miyawaki et al. in 2003 studied 134 MSIs placed in 51 patients and evaluated 1 year success rates. The MSIs with a 1.0 mm external diameter were significantly less stable than the MSIs with 1.5 mm or 2.3 mm diameters. With this in mind, the recent use of shorter but wider diameter MSIs has become increasingly popular and have shown increased removal torque and lateral displacement forces.[67] A mere increase of 0.25mm in the outer diameter of MSI significantly increased primary stability.[65] Ultimately, shorter MSIs that can provide equal or increased stability can subject patients to less risks and discomfort during placement.[68]

Force and Osseointegration

The accidental discovery of titanium as a viable implant material to form a direct connection to living bone suggests the existence of a unique environment at the interface between bone and metal.[26] The dynamics between implant and living bone have been well investigated.

Frost described his "mechanostat" principle in the 1960's as the model describing the thresholds of stress and strain that help or hinder to bone modeling process.[69] He defined the successful biomechanical interaction between bone and implant to be contingent on 2 major factors. First, the peri-implant tissues should be below the microscopic fatigue damage threshold of bone. Any strains above this threshold were thought to cause mechanical overload, which would lead to pathologic fracture of the bone. The second factor states that the strain should fall between the disuse-mode threshold strain range where bone strength can be reduced, and the "modeling threshold strain range" where modeling is induced to strengthen with load bearing bone. (Frost 1987) The mechanostat principle has proven to be useful in setting theoretical limits for defining thresholds of stress and strain during bony maintenance. Several studies have reported the applications of force loads on endosseous implants and the resulting increases in osseointegration values.[70]

Although some research has attempted to explain the bone-implant osteodynamic in endosseous implants, research on the influence of force load on osseointegration of MSIs has been limited and inconclusive. In a study by Melsen and Costa, 16 titanium vanadium screws were loaded with 25- and 50-g Sentalloy springs and observed at 1, 2, 4 and 6 months. Correlations were made between the amount of osseointegration with time, however there was no relationship with the amount of force.[71] In 2007, Freire et al placed 78 MSIs in six beagle dogs. They were loaded with 250 g immediately, after 1 week, after 3 weeks, and after 12 weeks of healing.[72] Histologically, no significant differences were observed between the unloaded controls and loaded groups, but some differences could be observed at the bone-implant interface. In another study by Ohmae in 2001, 36 MSIs were placed in three adult beagle dogs for intrusion of mandibular posterior teeth.[73] After a six week healing period, 150g of intrusive force was applied and animals were observed and sacrificed at 12 or 18 weeks of activation.

Histomorphometric analysis showed a slight difference between overall osseointegration with loaded (25.0%) and the unloaded (18.9%) MSIs. A study in 2009 by Woods et al described the effect of force, timing, and location on bone-to-implant contact of MSIs. Seven skeletally mature male beagle dogs and followed over a 110 day period. Different forces of 50 versus 25 g loading were used in both the maxilla and mandible. Mobility was evaluated using a 0-3 point scale before the MSIs were assessed by histologic analysis. The bone-to-implant contact percentage for 25 g versus 50 g were 43.4% and 37.9% respectively, however these were not shown to be statistically significant.[74] Massey et al utilized three-dimensional microcomputed tomography to analyze the effect of force on alveolar bone surrounding MSIs. The study compared MSIs loaded with 200 or 600 g and compared that to unloaded control screws. Using micro-CT, they assessed bone at the cortical and trabecular levels for both the compression side and noncompression zones. They observed that larger loads of 600 g produced less bone in the noncortical regions than smaller loads of 200 g. In addition, loaded miniscrew implants displayed less bone than unloaded miniscrew implants in the cortical regions, but more bone than unloaded miniscrew implants in the noncortical regions.[63] Vannet et al in 2007 placed 20 MSIs in the lower jaws of five male beagle dogs.[75] Force loads of 100 cN were applied immediately to eight MSIs and to another eight after 12 weeks of healing. After 25 weeks of total observation, 11 of the 20 were lost primarily due to the lack of primary stability. Of the remaining nine screws, eight were evaluated using histomorphometric analysis. Vannet et al reported osseointegration ratios of 74.48 ± 15.63% with no significant different between loaded and unloaded MSIs.

Fluting

A design aspect of MSIs that has yet to be thoroughly assessed in either endosseous or orthodontic literature is the effect of longitudinal fluting on stability and bone formation. It is important to differentiate between traditional cutting flutes, which are longitudinal channels in an implant tip designed to carry away bone debris from the cutting edge as the screw rotates and facilitate its self-tapping nature. These flutes can extend from 1.7 mm to 5.1 mm up the shaft from the tip of what are typically endosseous implants.[76] Traditional cutting flutes need to be distinguished from these that run vertically along the entire shaft to the screw.

Decreased insertion torque and cortical damage occur with the increase in the number and length of cutting flutes.[76, 77] The rationale is that increased clearance of bone debris accumulating around the threads would result in reduced resistance.[76, 78]

However, bone debris clearance is also dependent on having adequate flute dimensions and can increase insertion torque when bone chips or debris accumulate around threads of a fluted MSI, producing greater insertion resistance.[6, 76, 78] Bone debris, created during implant placement and adherence to moderately rough surfaces, significantly contributes to the initiation of bone deposition and mediates the connection between the old bone and the new bone on the implant surface.[79] The presence of cutting flutes has been reported to both decrease [80, 81] and increase[76, 82, 83] pullout strength. A study by Brinley et al 2009 compared 6 mm longitudinally fluted miniscrews with 6mm control non fluted miniscrews in synthetic bone. Miniscrews with fluting showed significant increases in insertion torque and pullout strength.[6] With only an approximate increase of 15% in insertion torque, pullout strength was increased by 420% in the synthetic bone model. Similarly, the placement torque was increased by about 119% but pullout strength was increased by 63% in cadaver bone. While increased insertion torque is generally not a desired trait for MSIs, Okazaki et al 2008 and Ikeda et al 2011 have shown good stability and clinical success despite high placement torque values.[62, 84] According to Wu et al 2011, endosseous implants have shown that fluting (depending on the shape) plays a significant role in altering resistance to insertion as well as holding strength.[85]

Minimal research has been done on the healing process and stability of MSIs with fluting in mature bone and more importantly, a live animal model. Currently there is a lack of information and understanding of the effects of longitudinal fluting on MSIs. Some research has been done to assess stability characteristics, with the majority of the studies focusing on insertion torque and pullout strength. However, no studies have yet shown what the effect MSIs with longitudinal fluting has on peri-implant bone formation as well as stability in a living bone model.

CHAPTER II

BACKGROUND

The introduction and use of mini-screw implants (MSIs) has provided orthodontists with useful tools for achieving increased or maximum anchorage. Anchorage allows desired tooth movements without the unwanted side effects of movements of other teeth. Traditional orthodontic mechanics have relied on control of the reactive unit (i.e. tooth or teeth) acting as anchorage against the active unit using extra-oral reinforcement, intra-arch or inter-arch mechanics. Such mechanics have been used for years to reinforce anchorage units, but they can be cumbersome and subject to compliancy issues. None of them provides the absolute anchorage. Miniscrew implants have rapidly gained popularity and acceptance due to their versatility, minimal invasiveness, low costs, and patient acceptance.[1-4] They are particularly advantageous when extraoral appliances are impractical and patient compliance is unreliable.[1, 4] Although MSIs provide clinicians with an important mechanical tool, the concern for implant failure remains a main deterrent for many orthodontists.[5] MSI size is another limitation. If size could be decreased, the risk of root injuries during and after insertion could be minimized.[67] One study that potentially addressed both stability and size showed that longitudinal flutes placed in 3 mm long MSIs increased pullout strength by approximately 60%.[6] However, the MSIs placed were in cadaver bone; further research is needed to evaluate the stability in living bone and to understand bone growth around fluted MSIs.

A number of methods of testing are considered useful in determining MSI stability including insertion torque, removal torque, and micro-computed tomography. Insertion torque has often been used as an indirect measure of primary stability.[18] Removal torque is generally considered as an indirect measure of the shear force required to fracture the bone-implant surface,[48] and may be good indicator of implant stability.[49] Lastly, micro-computed tomography (μCT) allows for high resolution three-dimensional images with the ability to take quantitative and qualitative measurements of surrounding bone.[57] When μCT and histomorphometric analyses are performed on the same bone specimens, the two have been shown to be highly correlated.[58, 59]

The purpose of the present study was to evaluate the effects of longitudinal flutes on bone healing and stability by evaluating MSI failure rates, insertion torque, removal torque, and the quality of bone surrounding the miniscrews.

CHAPTER III

MATERIALS AND METHODS

Animals

Eleven skeletally mature female New Zealand white rabbits 7-8 months of age, were used in this study. The animals were purchased from a licensed grade A breeder and the Institutional Animal Care and Use Committee at Texas A&M University Baylor College of Dentistry (Dallas, Texas, USA) approved the care of the animals and the experimental protocol. A total of 66 miniscrews were placed with various expansive forces delivered. The MSIs were subjected to a constant force of 100 grams.

Materials and Appliance

Two miniscrews were specifically fabricated for this study. The MSI's were self-drilling and made of titanium alloy; 3 mm long, with a 1.6 mm outer diameter, and the same thread count and pitch (0.5mm). The experimental MSIs had 3 flutes spaced approximately 120 degrees apart that were of 0.225 mm deep and extended the entire length of the threads. The control MSIs had standard continuous threads (Figure 1). Each rabbit received a total of six MSI's, inserted adjacent to the midsaggital cranial suture. The two anterior MSIs were always placed and loaded at the initial timepoint. Four additional posterior MSIs were either placed at the initial timepoint or 2 weeks prior to sacrifice (Figure 2). These posterior MSIs were loaded in an anterior-posterior direction with 100 grams of force. MSIs were randomly assorted to have a fluted and non-fluted pair at each position.

Surgical Procedure

At the day of MSI placement, the animals were weighed and then sedated with ketamine (75mg/kg IM) with xylozine (1mg/kg). The surgical sites were shaved and disinfected and anesthetized with 2% Lidocaine containing 1:100,000 epinephrine. A 3.0 mm tissue punch was used to prepare the sites. The anterior two MSIs were placed approximately 5 mm apart on either side of the midsagittal suture, approximately in the center of the anterior-posterior length of the orbits. Screws were placed so that a straight 20 mm long stainless steel wire could pass passively through the eyelets of both MSIs. All the screws were placed initially with a manual driver leaving approximately 2-3 threads visible, and then tightened with a continuous turn with the Mecmesin (Mecmesin Ltd, West Sussex, UK) static torque screwdriver with custom adaptor until they were fully inserted and the eyelet holes were aligned. The highest torque value registered during insertion was recorded. The posterior 4 MSIs were placed similarly, approximately 7 mm posterior to the first pair and 10 mm apart from each other (Figure 2).

The expansion apparatus consisted of a 20 mm long, .020 inch diameter, stainless steel guide wire engaged into islets in the heads of the MSI's. The wires were placed through 15 mm long nickel-titanium open coil springs, which were compressed between the MSIs to deliver 100 grams of continuous forces. The ends of the stainless steel wire were bent and composite was bonded to the ends to avoid irritation. Gut suture (5-0) was used to close up surgical tissue punch sites.

The animals were then euthanized with 2cc of Beuthanasia D given Intracardiac and perfused with 1-2 liters of normal saline followed by 1 liter of 70% ethanol. Six weeks following the initial placement, removal torque was measured on pairs of MSIs using the same Mecmesin (Mecmesin Ltd, West Sussex, UK) static torque screwdriver. Removal torque values were recorded immediately after sacrifice while still intact in the rabbit cranium. The highest removal torque value for each MSI was recorded.

The remaining MSIs were grossly sectioned from the cranium bloc using a fine circular saw. Sections were later refined with a marathon (Saeyang, Korea) handpiece with diamond disc under copious irrigation to ensure the passive fit of the bone-implant specimen in the 9.8 mm inner diameter of the cylindrical specimen carriers for Micro-CT scanning (Figure 3a).

Additional removal torque values were recorded after the MSIs had been scanned for Micro-CT analysis for remaining 6 week MSIs and all 2 week MSIs. Each bone section with MSI still intact was fixed firmly into die-stone plaster. A small piece of wax was placed on the inner portion of bone covering the apex of the MSI to prevent any added rigidity if the MSI had pierced the cranial bone. Die-stone plaster surrounded the bone section on all sides but did not cover the top or interfere with the implant bone interface.

Analysis

A maximum of five bone-implant specimens were placed in each holder, along with 4% PFA. The samples were then radiographically evaluated using micro-computed tomography (μCT 35; Scanco Medical, Basserdorf, Switzerland) with an isotropic resolution of 6 μm. X-ray energy levels were set to 70 kVp, current to 114 μA, and integration time to 800 ms. A 0.5 mm aluminum filter and a high resolution setting of 1,000 projections per 180 degrees were utilized to ensure highest quality scans with minimal metal implant artifact. A scout view was used to visually confirm the specimens in the tube prior to full scan (Figure 3b).

A volume of interest (VOI) for each specimen was defined as an approximate circle encompassing the implant at the center. The coronal limit of the VOI was visually defined as the slice at which bone was observed to completely surround and contact the MSI. The apical limit of the VOI was set to the tip of MSI. To account for variations in how much each screw was embedded, the analyses used the ratio of slice per total number of slices for every screw to determine bone volume total volume. Following scanning, a threshold was determined based on multiple specimens to distinguish the MSI from mineralized bone and background space. The threshold number, one in which one grayscale "brightness" number, above which all voxels are considered bone, and below which all voxels are considered non-bone (Figure 4). The threshold for segment 1 or the titanium selection had a gauss value of 3, sharp value of 4 and segmentation of 650. For segment 2 or the bone selection, the same gauss and sharp value of 3 and 4 were used respectively. The segmentation number used was 280. Three-dimensional images of the bone-implant specimens were reconstructed from the defined threshold parameters. Proprietary Scanco Medical programming scripts were used to produce and separate the different 3-dimensional voxel zones used in bone volume to total volume analysis.

BV/TV Analysis

Based on methodology previously established by Massey et al and Ikeda et al, the ratio of bone volume to total volume (BV/TV) was calculated from the layers around the MSIs.[62, 63] Each layer was 3 voxels, equivalent to 18 μm thick. The layers were defined as: 6-to-24 μm, 24-to-42 μm, and 42-to-60 μm from the MSI surface. (Figure 5 and 6) BV/TV ratios from 0-to-6 μm were excluded in this study due to a possible metallic halacion effect.

Statistical Analysis

Insertion and removal torque means were analyzed by independent t-tests. To quantify the BV/TV for the three-dimensional segmental analysis, each of the three defined layers were assessed separately for the screws that were in bone for 2 weeks and 6 weeks. The number of slices per screw varied on the amount of bone and the depth of placement. In order to standardize the relative number of slices, the slice number was divided by the total number of slices which produced relative section thickness ranging from 0-100%. Multilevel statistical models were used to determine any differences between the fluted and control MSIs at each layer: 6-to-24 μm, 24-to-42 μm, and 42-to-60 μm. Multilevel modeling was chosen to describe this data for two reasons. First, the multilevel models used made no assumptions as to the equality of the intervals used for the independent variable.[86] Secondly, multilevel statistics make it possible to easily determine the appropriate curves to use and to evaluate group differences within the changes that took place.[86, 87]

CHAPTER IV

RESULTS

Success Rates of MSIs

The overall success rate for the study was 97.0% for the fluted (1 failure out of 33 screws) and 97.0% for the non-fluted (1 failure out of 33 placed) MSIs. Both failures were complete failures; the MSIs were completely displaced out of bone and the expansion springs were no longer active. The opposing MSIs that remained stable were not included in the analyses because their loads differed from the other screws assessed.

Insertion Torque Values

Insertion torque of the fluted MSI's ranged from 1.5 to 7.0 N cm, with a mean of 3.98 N cm ±0.24. The insertion torque for the non-fluted MSI's ranged from 1.7 to 6.1 N cm, with a mean of 3.95 N cm ±0.24 (Figure 7). There was no statistically significant ($p=.930$) difference between the fluted and non-fluted MSIs. (Figure 8)

Removal Torque (2 weeks)

The removal torque of the fluted MSI's at 2 weeks ranged from 1.9 to 3.9 N cm, with a mean of 2.87 N cm ±0.70. The removal torque values for the paired non-fluted MSI's taken from sections ranged from 1.7 to 3.7 N cm, with a mean of 2.75 N cm ±0.69 (Figures 9 & 10). The difference between the two groups of MSIs was not statistically significant ($p=0.702$).

Removal Torque (6 weeks)

The removal torques of the fluted and non-fluted MSI's taken in situ, with a mean of 2.98 N cm ±1.14 (range from 2.2 to 5.0 N cm) and 1.98 N cm ±0.55 (range from 1.2 to 2.7 N cm), respectively (Table 1). The difference between fluted and non-fluted MSIs was not statistically significant ($p = 0.116$). The removal torque values for the fluted MSI's taken from sectioned skull fragments ranged from 2.6 to 5.1 N cm, with a mean of 3.67 N cm ±0.80. The removal torque for the paired non-fluted MSI's taken from the sectioned skull fragments ranged from 1.5 to 3.8 N cm, with a mean of 2.77 N cm ±0.73 (Table 1). The removal torques of the fluted and non-fluted MSI's combined were 3.42 N cm ±0.26 (range from 2.2 to 5.1 N cm) and 2.49 N cm ±0.20 (range from 1.2 to 3.8 N cm), respectively (Figure 11). The difference was statistically significant ($p = .024$) (Table 1). Removal torque of the fluted MSIs was significantly ($p = .008$) higher than the removal torque of the non-fluted MSIs when the MSIs were combined (Figure 12 & Table 1).

Bone Volume Fraction [6-to-24 μm, 24-to-42 μm, and 42-to-60 μm] Fluted and Non-Fluted MSIs – 2 weeks

After two weeks, bone volumes of the three layers were significantly greater ($p < .05$) for the non-fluted compared to fluted MSIs (Figure 13, 14, 15 & Table 3).

Bone Volume Fraction [6-to-24 μm, 24-to-42 μm, and 42-to-60 μm] Fluted and Non-Fluted MSIs – 6 weeks

After six weeks, bone volume of the 6-24 μm, 24-to-42 μm, and 42-to-60 μm layers were significantly greater ($p<.05$) for the non-fluted than fluted MSIs compared to fluted MSIs (Figure 16, 17, 18 & Table 3).

CHAPTER V

DISCUSSION

High success rates can be obtained when using loaded 3 mm long MSIs. Both the fluted and non-fluted MSIs had a 97% success rate. The success rates were equal or higher than previous studies using animal models, and substantially higher than success rates reported for human studies. Liu et al reported success rates of 98%, 86%, 88%, and 90% for similar 3 mm non-fluted MSIs inserted into immature rabbit frontal bones.[88-91] Using a canine model, Mortensen et al produced net success rates of 100% and 85.7% for the 3 mm MSIs loaded with forces of 600 or 900 g, respectively.[68] The two most recent systematic reviews reported clinical success rates to be 83.6% and 86.5% for MSIs 6 mm in length or longer.[22, 23] The high success rates observed in the present study suggests that the clinical success of MSIs is dependent on a number of other confounding factors (i.e. it is not just MSI length). For example, the rabbit studies that placed MSIs in the cranium did not have to be concerned about root proximity, which has been indicated as a major factor for screw failure.[44, 87] In addition, bacteria play a role in the failure of orthodontic miniscrews; a high rate of implant failure has been reported with peri-implant infection.[44] The ability to control variables in animal studies certainly enhances the success of MSIs, and it provides a better understanding of the potential risk factors for MSI instability clinically.

There were no differences in insertion torques between fluted and non-fluted MSIs. The present in-vivo study showed virtually no difference (0.76%) of insertion torque between the fluted and non-fluted MSIs. The only comparable study involving fluted MSIs reported a significant 14.8% increase in insertion torque, but they were placed in denser, thicker, synthetic bone and the MSIs were 6 mm long.[6] Simply by decreasing cortical density by 12.5%, insertion torque decreases by 12%–19%; decreasing cortical thickness by 50% decreases insertion torque values by 27%–32%; shorter 3 mm MSIs require 26%–33% less insertion torque than 6 mm MSIs.[92] The shorter 3-mm MSIs placed in rabbit skulls, which are composed of thinner, less dense bone, could explain the lower insertion torque values and the smaller differences in insertion torque between fluted and non-fluted MSIs.

Miniscrew implants can maintain stability and have high success rates even with low maximum insertion torque values. Insertion torques of successful fluted and non-fluted MSIs were as low as 1.5-1.7 N cm. Based on 124 MSIs placed into 41 orthodontic patients, Motoyoshi et al reported significantly greater stability for screws with maximum insertion torques ranging between 5-10 N cm, than for MSIs whose insertion torques were less than 5 N cm and more than 10 N cm.[18] It is thought that the initial primary stability cannot be maintained if insertion torque is too small, and that later secondary stability (proper healing and osseointegration) may not be achieved if insertion torque is too great. Insertion torque has been closely associated with bone density.[92, 93] If an implant is not sufficiently stable at the time of placement, micromotion may occur and the normal healing process may then be disrupted from fibrous tissue formation.[93, 94] In the present study, the average maximum insertion torques were well below the ideal range suggested. Due to the reduced density and thickness of rabbit cranial bone, the proposed optimal insertion torque values are potentially different and should be considered unique for different bone density and quality.

At 2 weeks, longitudinally fluted MSIs increased average removal torque compared to control MSIs but were not significantly different. Removal torques were only 4.4% higher for fluted than non-fluted MSIs. While removal torque of fluted MSIs has not been previously evaluated, Brinley et al showed that pullout strength of 6 mm fluted MSIs was 62% greater than their non-fluted counterparts[6], but pullout strength is a different stability variable and their study was performed on synthetic bone. Sohn et al evaluated spontaneous healing capacity of surgically produced cranial defects in rabbits and showed that the earliest phase of healing started after 2-4 weeks, and that 8 weeks were required before new bone, bone remodeling, and bone regeneration could be observed.[95] With only a minor increase in removal torque at 2 weeks and a more substantial increase at 6 weeks, it appears that increases in removal torque are higher if given time for adequate bone formation.

At 6 weeks, longitudinally fluted MSIs significantly increased average removal torque compared to control MSIs. Removal torques were 37.3% higher for fluted than non-fluted MSIs. While removal torque of fluted MSIs has not been previously evaluated, Brinley et al showed that pullout strength of 6 mm fluted MSIs was 62% greater than their non-fluted counterparts.[6] Interestingly, Kim et al showed that adding dual-threads to MSIs increases removal torque.[49] They theorized that the dual thread MSIs might have had a reduced backward removal velocity due to the smaller pitch. The addition of flutes on MSIs may have a similar mechanical effect on removal torque.

More importantly, the new woven bone that formed in the flutes may serve as a physical barrier that resists MSI removal and thereby, increases removal torque.

Adding flutes does not result in increased amounts of bone surrounding the MSI. After six weeks, the control MSIs had 0.2-10.4% more bone surrounding them than the fluted MSIs, with the 6-24 µm layer showing the greatest and the 42-60 µm layer showing the smallest differences. The pattern was the same after two weeks, although the differences were smaller. More microdamage could have been caused by the fluted MSIs, requiring more time for new bone to be laid down. It has been suggested that the ultimate fracture strength of bone occurs around 25,000 microstrains, leading to microfractures.[69] If remodeling after implant insertion cannot repair the microdamage, local ischemia, bone necrosis, and bone resorption can occur.[8, 20, 61] Even when pilot holes are drilled, microdamage has been demonstrated as far as 200 µm from the MSI.[8] Chen et al showed that heavily fractured cortical bone is produced after punching holes with mini-microfracture awls, with substantial osteocyte necrosis and sealed off the marrow blood supply.[25] As such, the reduced bone volumes found around fluted MSIs after two and six weeks may also be due to immature bone that has not calcified sufficiently to be detectable. Due to their poor blood supply and limited bone marrow, rabbit craniums resemble human mandibular bone.[46] Sohn et al, who evaluated spontaneous healing capacity of surgically produced cranial defects in rabbits, showed that the earliest phase of healing started after 2-4 weeks, and that 8 weeks were required before new bone, bone remodeling, and bone regeneration could be observed.[95] However, they used immature rabbits, whose healing capacity and bone formation might be expected to be greater than those of more skeletally mature rabbits. Endosseous implants placed in the hind legs of rabbits showed postoperative remodeling of woven bone to well-organized and mature secondary osteons was routinely observed from weeks 6 to 16.[96] Bodde et al, also evaluated cranial size defects in rabbits, recommended an observation of at least 12 weeks for bone formation.[47] The extended duration required for mature bone formation in a skeletally mature rabbit and potentially increased micro damage of fluted MSIs may suggest that bone volume fraction assessed in our study did not have adequate time for new bone to mature and be measureable. In addition, the bone that was formed in flutes was immature and less calcified than the bone surrounded in non-fluted screws and the amount and density of immature bone has previously been shown to be less than mature bone.[41] Given enough healing time, bone volume to total volume might be expected to be equal to that of control MSIs.

The layer closest to the MSI exhibited less bone than the outer two layers, which showed no differences. This difference was evident for both the two and six week specimens, suggesting that bone remodeling is initiated away from the implant surface. Less bone in the intimate layer 6-to-24μm than in the outer 24-60 μm layers has been previously been reported for dogs.[63, 97] Based on 7 μm μCT slices to evaluate bone volume fraction around MSIs placed in 40 rats, it has been shown that bone formation starts 30-50 μm away from the bone-implant interface after approximately five days and progresses toward the implant over subsequent 23 days.[98] If bone formation starts at 30-50 μm from the MSI, then the 24-to-42 and 42-to-60 μm layers might be expected to be similar and more ossified than the 6-to-24 μm layer.

The bone volumes observed at the most intimate 6-to-24 µm layer do not reflect scatter or metallic halation artifacts produced by the implant. Studies that that have evaluated regions of bone immediately adjacent to larger endosseous implant surfaces have consistently reported greater amounts of bone due to halation effect.[62, 99, 100] This has led to the exclusion of the region directly adjacent to an implant due to overestimations of bone. In contrast, the present study showed less bone in the most intimate layer at both time points and minimal differences further from the MSI at layers 24-to-42 µm and 42-to-60 µm. Furthermore, the previous µCT studies relied on hardware had more limited resolution (24-30 µm), and much larger cylindrical implants (10-13mm long, 3.5-4.2 mm diameter), which might be expected to produce more scatter than the 3 mm MSI used in the present study.[99, 100] Also, inaccuracies in the algorithmic software reconstruction and the overall orientation of MSI during the MicroCT scan could cause variations in measured bone values.

The equal success rate, increased removal torque, and potential for increased bone formation over longer healing times could make 3 mm fluted MSIs a viable clinical option. Adding flutes to miniscrews can potentially increase mechanical parameters generally associated with MSI stability, such as removal torque. However measureable increases in bone formation may require a longer healing periods. The present study clearly shows the miniscrew implants can be placed in animal models and remain stable with insertion torque values less than once thought to be optimal, making it necessary to consider confounding variables in clinical situations including technique, hygiene, and inflammation.

CHAPTER VI

CONCLUSIONS

- Loaded 3 mm fluted and 3 mm non-fluted MSIs can have very high success rates.
- Miniscrews can be stable with maximum insertion torque less than 6.1 N cm.
- Adding flutes to 3 mm MSIs increased the removal torque or holding power by 37% after 6 weeks of healing.
- There was less bone in the segment closest to the MSIs (6-to-24 μm) than those further away (24-to-42 and 42-to-60 μm) from the implant surface.
- There was greater bone surrounding control non-fluted than fluted MSIs after 2 and 6 weeks of healing.

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APPENDIX A

FIGURES

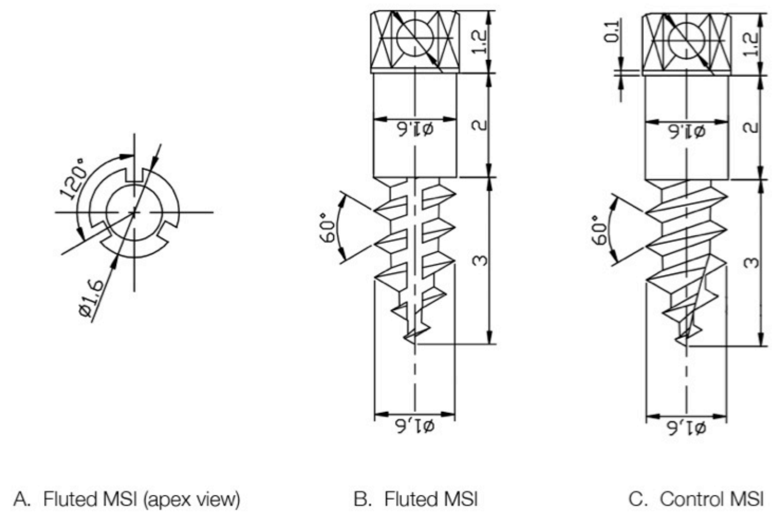

Figure 1. A) Fluted MSI as seen from the apex view, 3 longitudinal rows of flutes spaced 120 degrees apart. B) Fluted MSI with dimensions. C) Control MSI with dimensions.

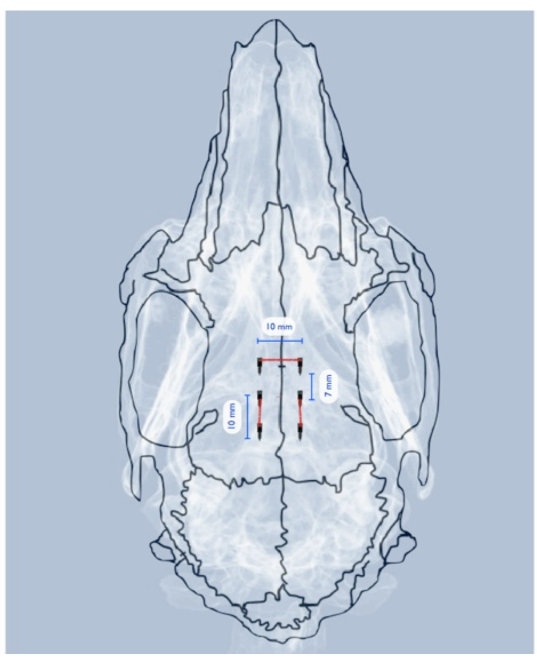

Figure 2. Coronal view diagram of rabbit skull with MSI placement and approximate measured distances.

A. B.

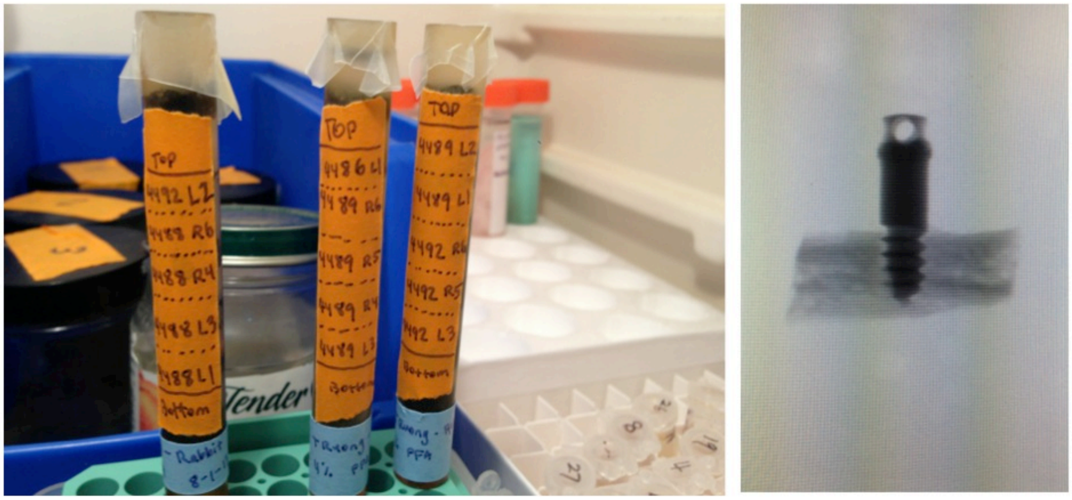

Figure 3. A) MicroCT tubes filled, labeled and ready to be scanned B) Scout view of specimen prior to MicroCT scan

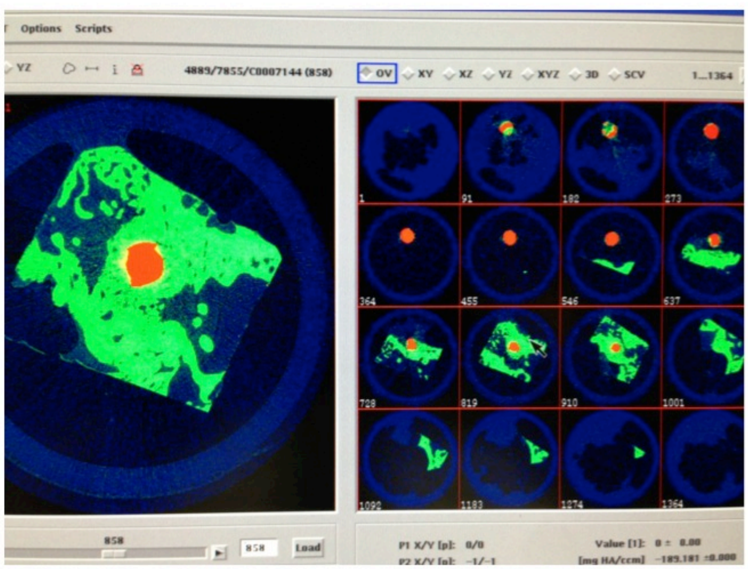

Figure 4. Selection of volume of interest (VOI) and threshold

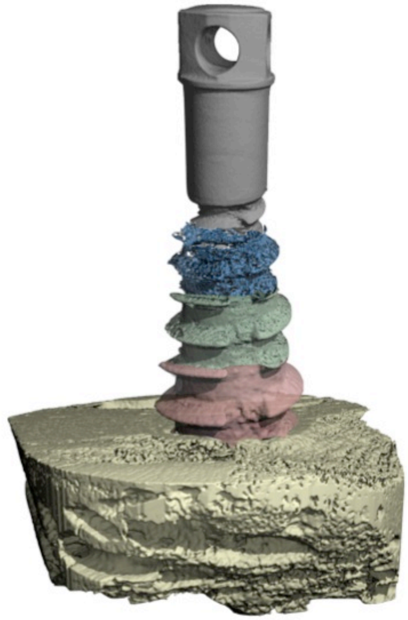

Figure 5. Generated three-dimensional rendering of MSI with bony layers of interest from Micro-CT. Blue represents the most intimate layer from 6-24µm, green represents the middle layer from 24-42µm, and pink represents the most outer layer from 42-60µm.

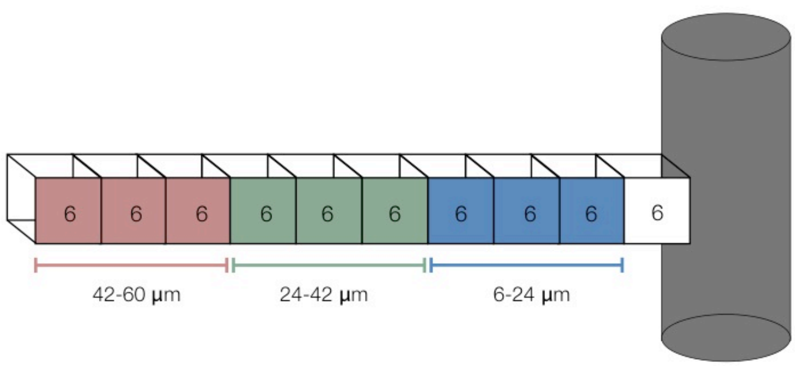

Figure 6. Schematic representation of MSI and bone regions of interest. Blue represents the most intimate layer from 6-24µm, green represents the middle layer from 24-42µm, and pink represents the most outer layer from 42-60µm.

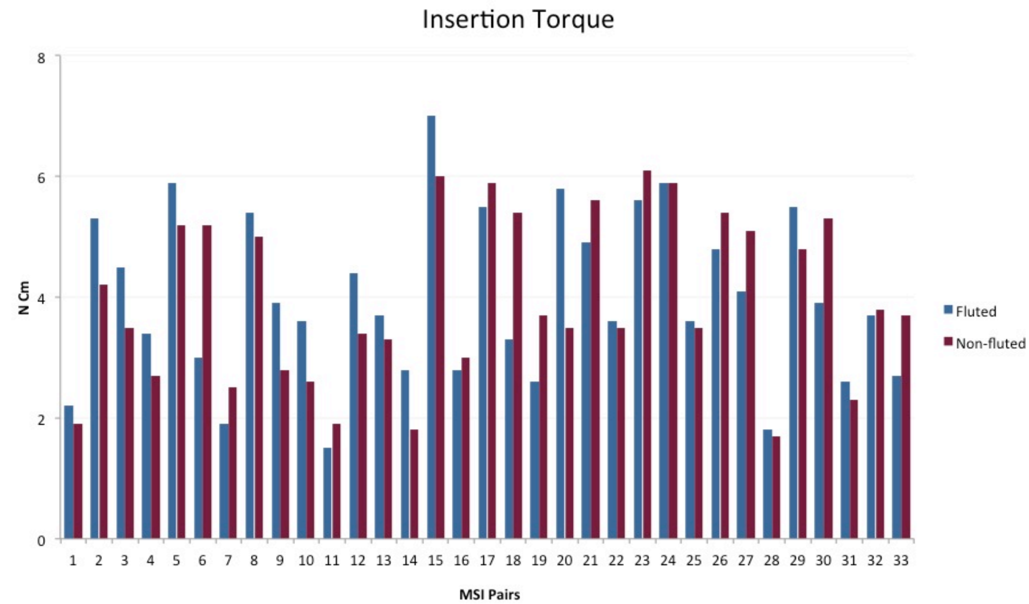

Figure 7. Insertion torque values for 33 MSI pairs in N cm.

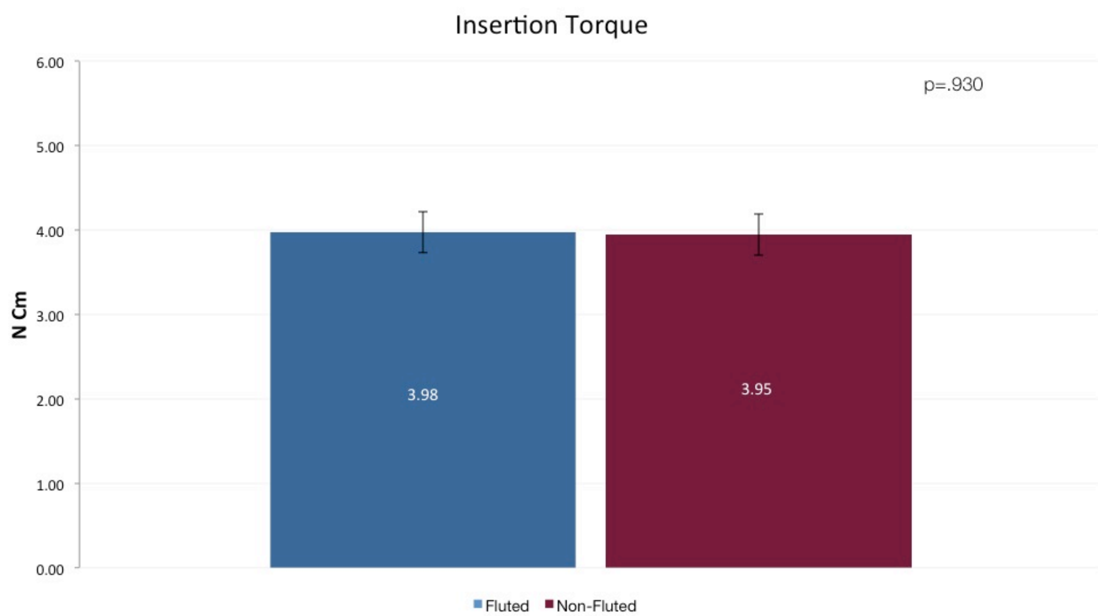

Figure 8. Insertion torque averages for fluted and control MSIs in N cm.

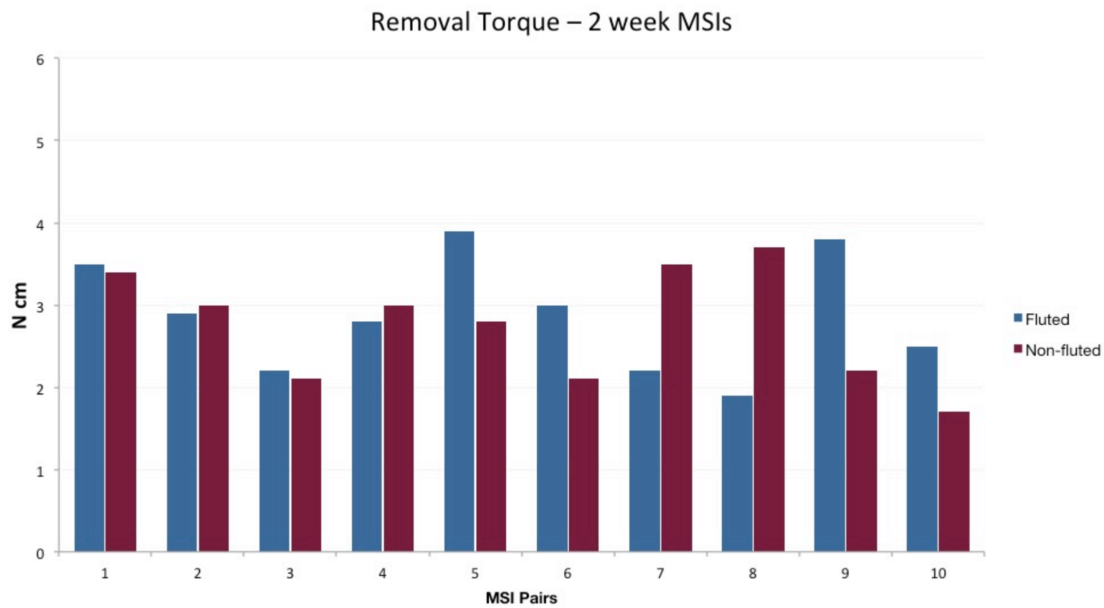

Figure 9. Removal Torque values for 10 MSI pairs in N cm taken on MSIs placed and left to heal for 2 weeks

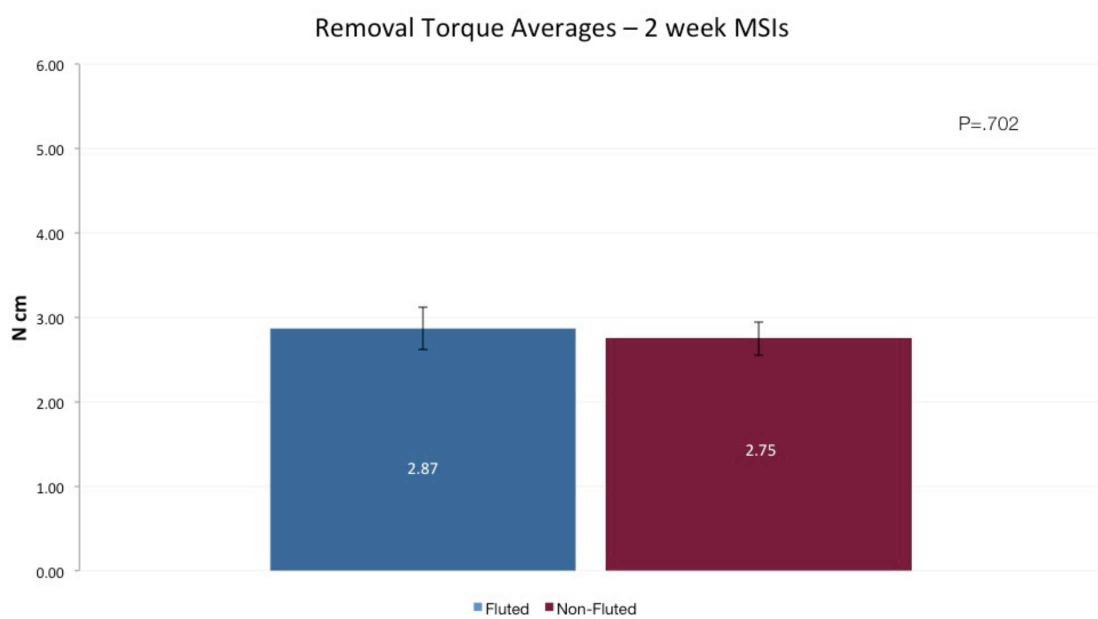

Figure 10. Removal torque averages for fluted and control MSIs in N cm taken on MSIs placed and left to heal for 2 weeks.

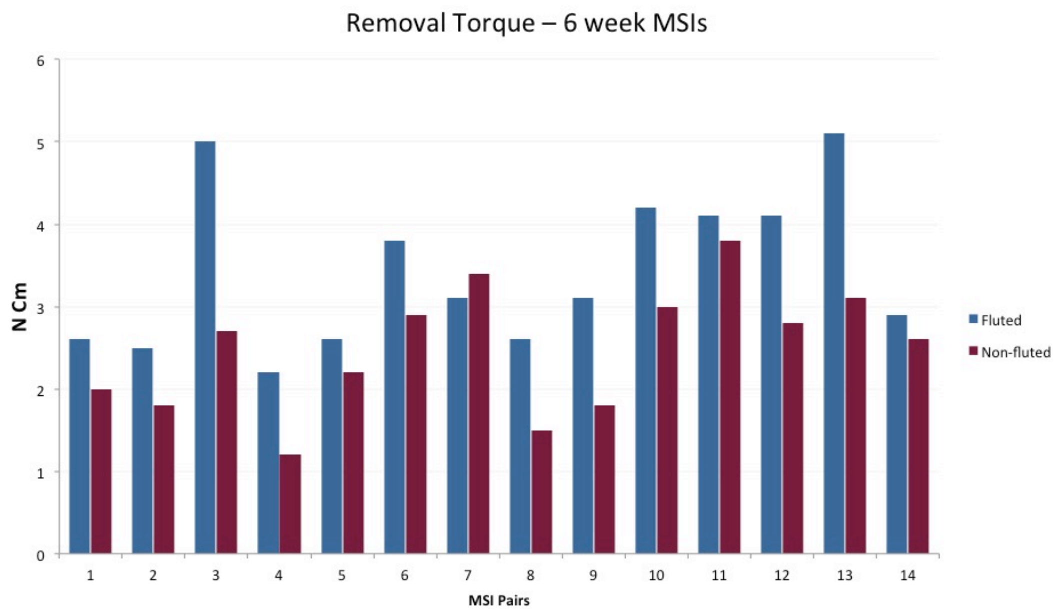

Figure 11. Removal Torque values for 14 MSI pairs in N cm taken on MSIs placed and left to heal for 6 weeks.

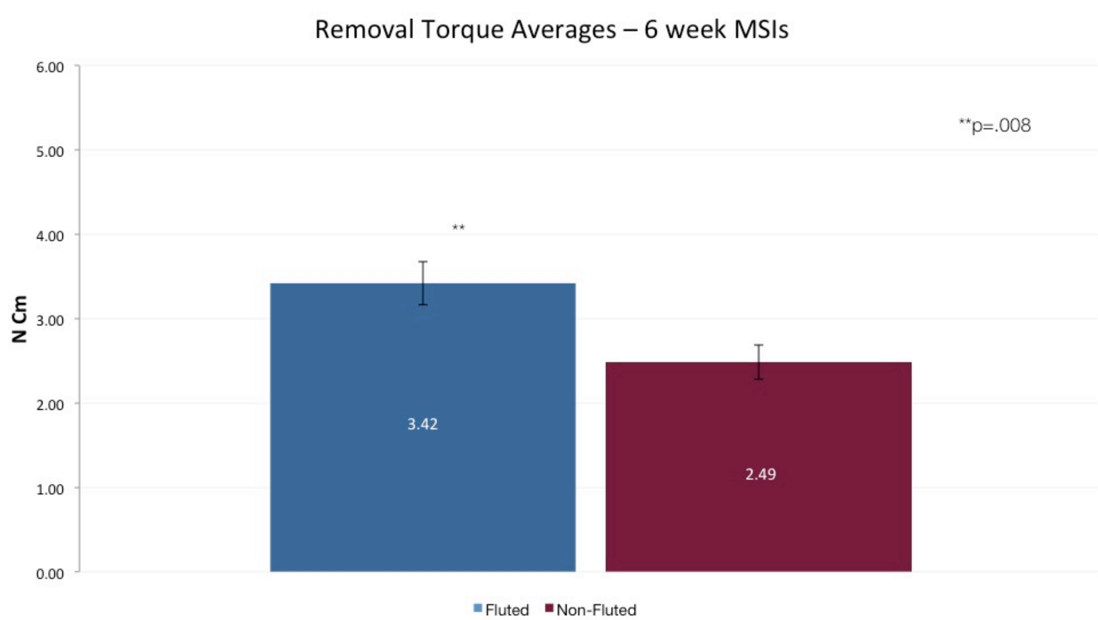

Figure 12. Removal torque averages for fluted and control MSIs in N cm taken on MSIs placed and left to heal for 6 weeks.

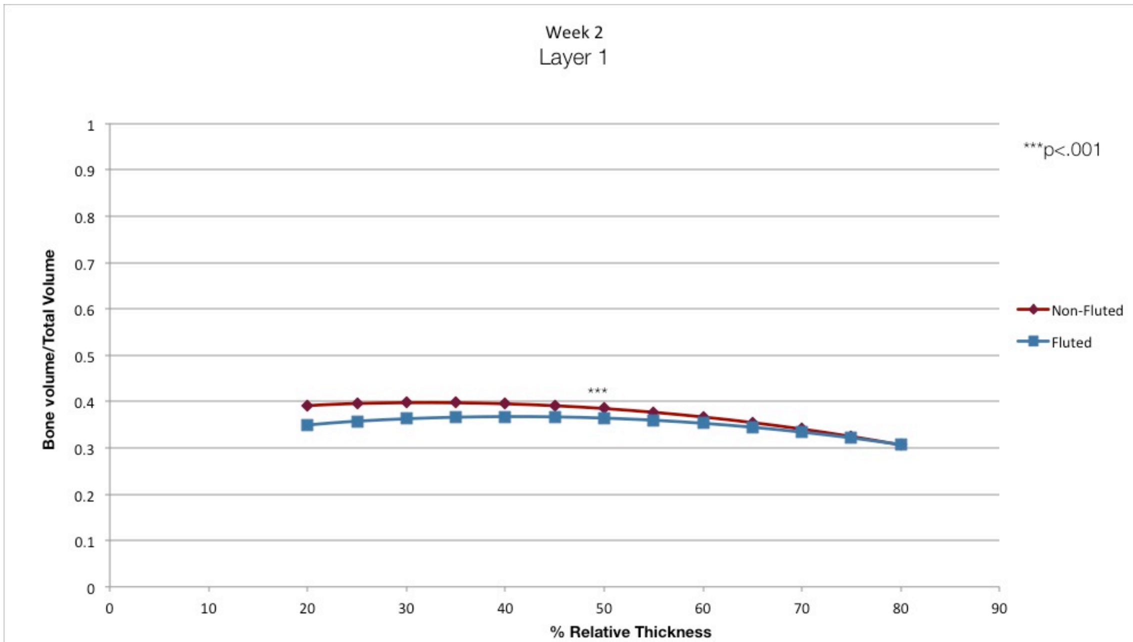

Figure 13. Bone volume/total volume of layer 1 from 6-24µm for fluted MSIs compared to non-fluted MSIs for MSIs placed and left to heal for 2 weeks.

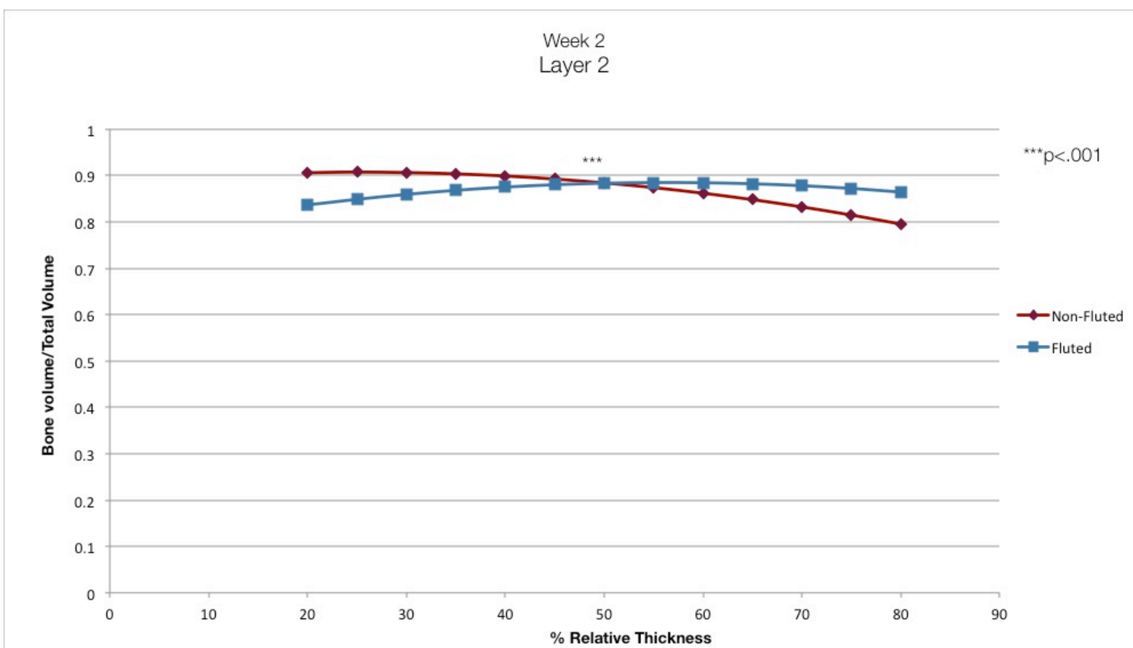

Figure 14. Bone volume/total volume of layer 2 from 24-42µm for fluted MSIs compared to non-fluted MSIs for MSIs placed and left to heal for 2 weeks.

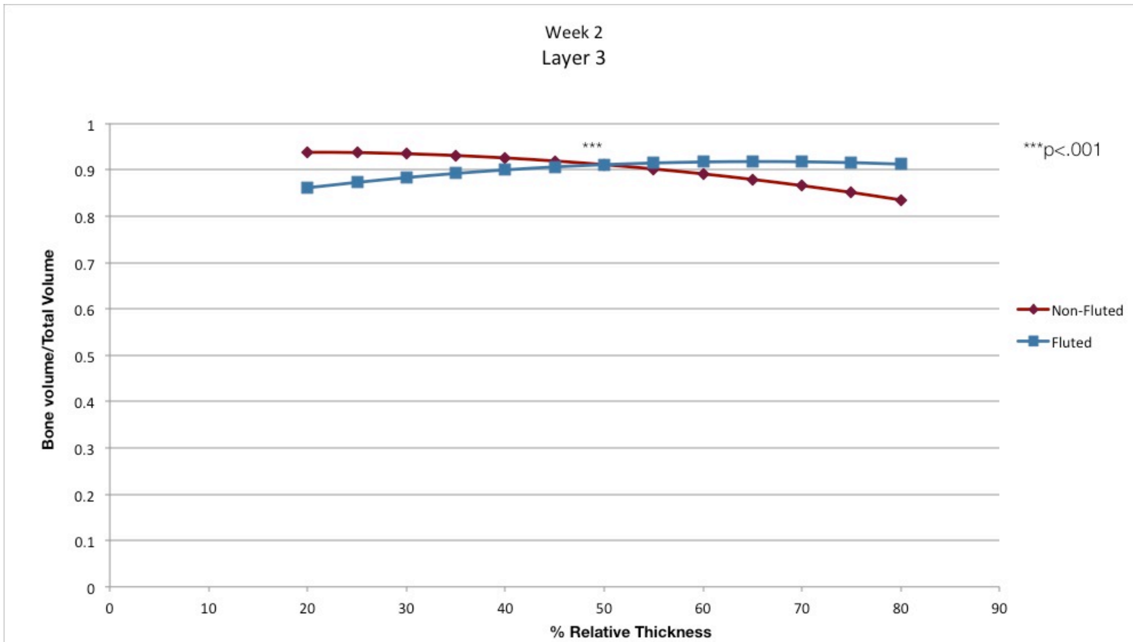

Figure 15. Bone volume/total volume of layer 3 from 42-60μm for fluted MSIs compared to non-fluted MSIs for MSIs placed and left to heal for 2 weeks.

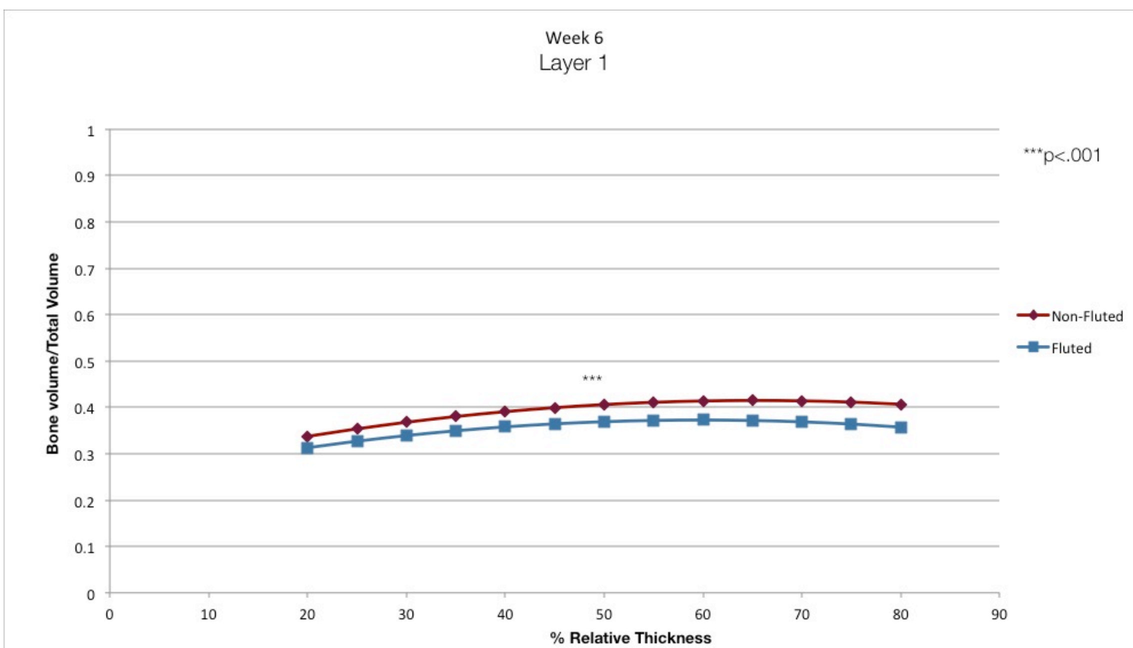

Figure 16. Bone volume/total volume of layer 1 from 6-24μm for fluted MSIs compared to non-fluted MSIs for MSIs placed and left to heal for 6 weeks.

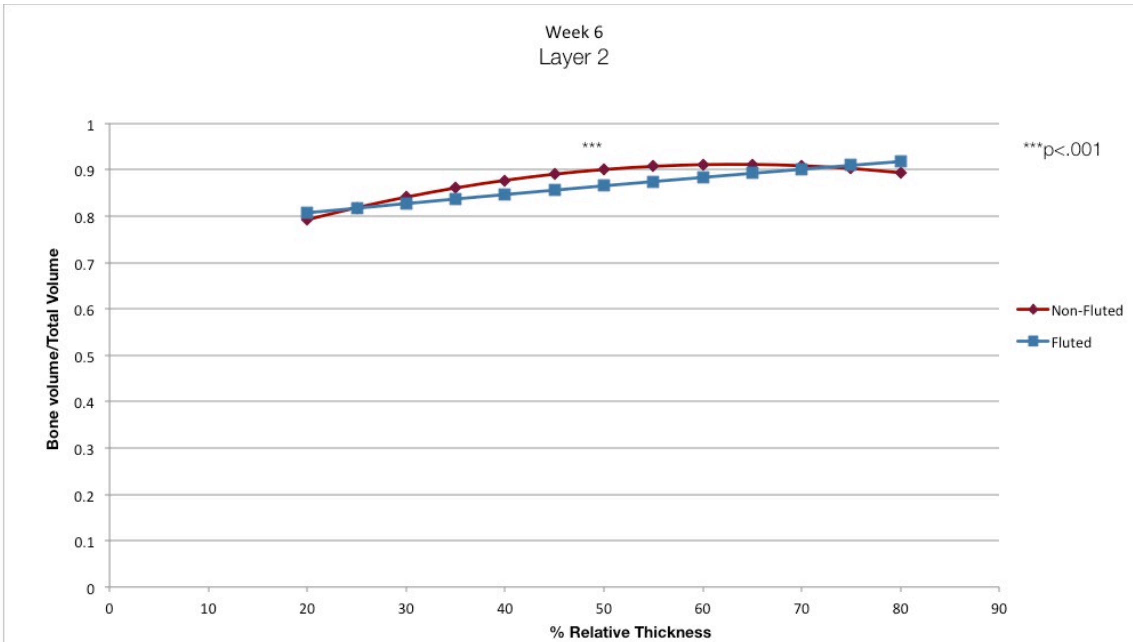

Figure 17. Bone volume/total volume of layer 2 from 24-42μm for fluted MSIs compared to non-fluted MSIs for MSIs placed and left to heal for 6 weeks.

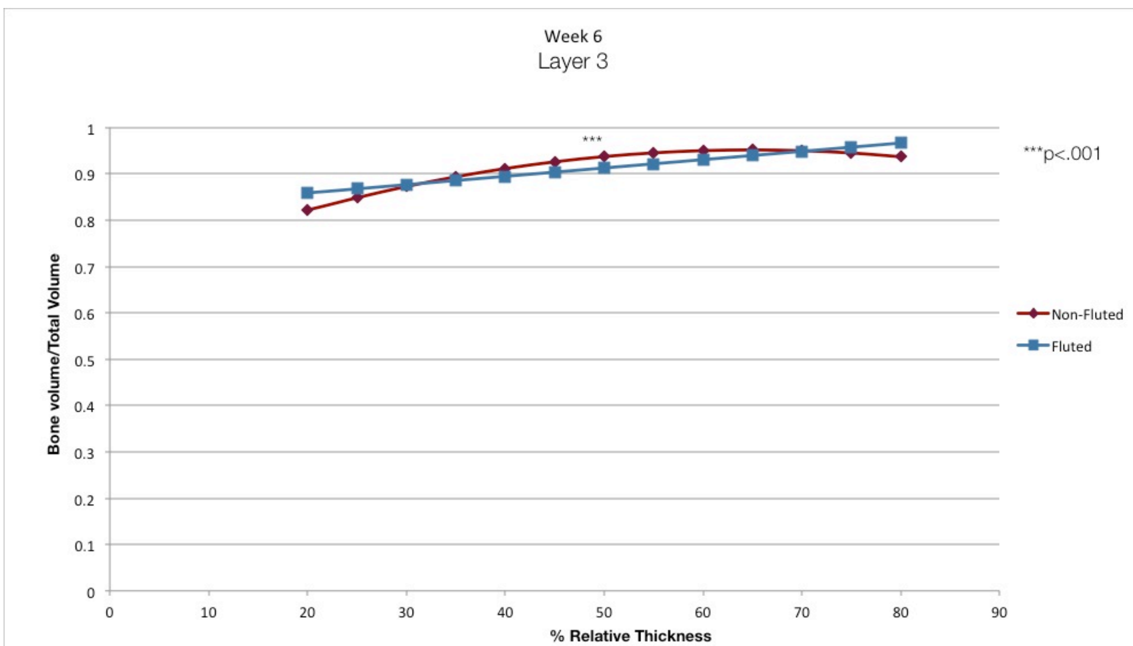

Figure 18. Bone volume/total volume of layer 3 from 42-60μm for fluted MSIs compared to non-fluted MSIs for MSIs placed and left to heal for 6 weeks.

APPENDIX B

TABLES

	Fluted		Control		
	Mean	Standard Deviation	Mean	Standard Deviation	P-Value
Intact, N cm	2.98	1.14	1.98	0.55	.116
Re-fixed Sections, N cm	3.67	0.80	2.77	0.73	.024*
Combined, N cm	3.42	0.95	2.49	0.76	.008**

Table 1. Table comparison of removal torque of 6 week MSIs between skeletally intact, sectioned and re-fixed specimen, and all combined means with standard deviations.

	Fluted		Control		
	Mean	Standard Deviation	Mean	Standard Deviation	P-Value
Insertion Torque, N cm	3.98	1.39	3.85	1.39	.930
Removal Torque (6 weeks), N cm	3.42	0.95	2.49	0.76	.008**
Removal Torque (2 weeks), N cm	2.87	0.70	2.75	0.69	.702

Table 2. Table comparison of insertion and removal torque (6 and 2 week) means including standard deviations.

	Control Non-fluted						Difference Between Control and Fluted					
	Intercept	SE	Slope	SE	Quadratic	SE	Intercept	SE	Slope	SE	Quadratic	SE
2 weeks												
6-24μm	3.848e-1	1.322e-2	-1.424e-1	1.112e-2	-3.957e-2	5.136e-2	-2.059e-2	2.813e-3	7.034e-2	1.593e-2	--	--
24-42μm	8.841e-1	1.250e-2	-1.854e-1	1.364e-2	-3.692e-1	6.271e-2	6.584e-4	3.433e-3	2.324e-1	1.945e-2	--	--
42-60μm	9.113e-1	1.125e-2	-1.721e-1	1.194e-2	2.715e-1	5.490e-2	-3.904e-5	3.006e-3	2.574e-1	1.703e-2	--	--
6 weeks												
6-24μm	4.060e-1	2.168e-2	1.155e-1	1.046e-2	-3.777e-1	4.792e-2	-3.696e-2	2.618e-3	-4.152e-2	1.488e-2	--	--
24-42μm	9.005e-1	2.553e-2	1.690e-1	1.430e-2	-6.407e-1	9.212e-2	-3.518e-2	5.327e-3	1.576e-2	2.033e-2	6.078e-1	1.310e-1
42-60μm	9.373e-1	2.161e-2	1.933e-1	1.354e-2	-6.486-1	8.725e-2	-2.457e-2	5.045e-3	-1.330e-2	1.926e-2	6.470e-1	1.240e-1

Table 3. Multilevel comparisons of control non-fluted MSIs and differences to fluted MSIs with linear and quadratic estimates. Bone volume fraction (BV/TV) for bone at 6-24 μm, 24-42 μm , and 42-60 μm.

Bone volume/Total Volume BV/TV = Intercept (at 0% relative section thickness) + (Slope *% relative section thickness)