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PERI-IMPLANT BONE HEALING AFTER SIMPLIFIED OSTEOTOMY WITH SINGLE DRILLING VERSUS SEQUENTIAL DRILLING PROTOCOL IN A RABBIT TIBIA MODEL; HISTOLOGICAL AND HISTOMORPHOMETRIC STUDY

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ABSTRACT

Background: In order to increase patient satisfaction with implant placement, innovative methods that preserve more tissue and are faster to prepare the osteotomy are being applied.

Objectives: The aim of the current study was to assess the impact of different drilling techniques on peri-implant bone healing using simplified fast osteotomy technique with single drill versus sequential drilling protocol in a rabbit tibia model.

Materials and Methods: Twenty rabbits were allocated into two groups: the test group received a simplified fast drilling approach, whereas the control group underwent a sequential drilling protocol. 10 implants from each group were removed 4 weeks after implantation and 10 implants were removed 6 weeks after implantation for histological and histomorphometric analysis.

Results: Histological results revealed formation of greater amount of newly formed mature bone with decreased remodeling figures and inflammatory cells in the second observation period for both groups. The histomorphometric results revealed that surface area of the bone adjacent to implant was significantly higher in the sequential drilling group than in the single drilling group at four weeks (p=.019), while there was no significant difference between the two studied groups at six weeks (p=.589). On the other hand thickness of bone trabeculae was significantly higher in the single drilling group than in the sequential drilling group at four weeks (p=.009),while there was no statistically significant difference between the two studied groups at six weeks (p=.865).

Conclusion: Regarding the proposed parameters for evaluating the peri-implant behavior; single drilling osteotomy did not differ from sequential drilling.

KEYWORDS: Osseointegration, peri-implant bone healing, single drilling,sequential drilling, fast osteotomy

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INTRODUCTION

Dental implants continue to increase worldwide and are swiftly taking over as the preferred technique for replacing teeth.[1] Today's predictable functional and cosmetic outcomes demonstrate the success of dental implants.[2]

The quantity and quality of the hard and soft tissue surrounding the planned implant site, as well as other aspects of the implant or specific surgical techniques, all play a role in whether or not the implant will be properly integrated and looks naturally.[3]

The crucial step in the surgical protocol is implant drilling. For a predictable osseointegration and an appealing natural implant restoration, a minimally invasive approach is recommended.[4]

Long-term preservation of the surrounding tissue has been shown to depend on the temperature rising during drilling. It has been documented that a temperature of 47°C for 1 minute can result in bone necrosis at the drilling site.[5] with the consequence of compromised implant stability and osseointegration .[6] However, additional researches have shown that, as long as fundamental guidelines are followed in diverse circumstances, procedures generating heat to levels that would result in implant failure are uncommon.[7]

Furthermore, the success rate of dental implants is known to be influenced by the drilling sequence and speed. Sequential drilling Using a series of increasing diameter drills has long been recognized as an implant site preparation strategy.[1] The use of several drills at various stages, however, takes time and has a number of drawbacks, including increased patient discomfort and infection risk as well as operator boredom.[8]

Therefore, both professionals and patients can be in favor of any simplification of site preparation approaches. Certain modifications to the drill design and drilling approach have been recommended in order to reduce the risk of overheating the implant site and simplify the surgical procedure.[8, 9]

If lowering the number of steps in the drilling procedure does not have a negative influence on success, it might be worthwhile. According to studies on the subject, simplifying the drilling technique produced satisfactory results.[10, 11]

Recently, there has been an upward trend in the investigation of fast osteotomy, which allows for the preparation of implant sites using a shortened technique in various types of bone.[12]

The number of drills utilised during implant osteotomy was decreased and compared to standard sequences that included multiple drills.[8, 12, 13]. In this context, Gehrke et al.[14] presented a research that proved measures of the implant stability quotient (ISQ), insertion torque value (ITV), and precision of osteotomy using standard and simplified single drilling techniques.They disclosed that fast osteotomy revealed considerably greater ISQ and ITV than the systems evaluated employing a multiple-drill procedure for the osteotomy.

The purpose of the present study was to compare through histological and histomorphmetric analysis the impact of drilling protocol on peri-implant bone healing using simplified fast osteotomy technique with single drill versus conventional sequential drilling protocol in a rabbit tibia model.

The null hypothesis was that utilizing a simplified osteotomy approach would have no effect on peri-implant bone healing when compared to implants inserted using standard osteotomy with sequential drilling.

MATERIALS AND METHODS

Study sample and setting

Sample size

The required sample size was established based on a prior study that attempted to test the hypothesis that lowering the number of drills for site preparation

compared to the normal drilling sequence would result in no differences in osseointegration.[13] According to Giro et al. (2013)[13], both approaches resulted in implant integration. There were no variations in bone-to-implant contact (BIC) or bone-area-fraction occupancy (BAFO) between drilling procedures as time passed in vivo. They came to the conclusion that the simplified drilling methodology produced comparable osseointegration results to the standard protocol, proving the initial hypothesis. The sample size was calculated to demonstrate the consistency of simplified osteotomy with single drilling and sequential drilling protocol. Based on Giro et al. (2013)[13] results, adopting a power of 80%, and level of significance 95% (α=0.05), If there is truly no difference between the single drilling and sequential drilling protocols, then 8 implant sites per group (number of groups 2, number of time points =2) are required to be 80% provided that the lower limit of a one-sided 95% confidence interval (or equivalently a 90% two-sided confidence interval) will be above the non-inferiority limit of -5. Total sample size=**10 implant sites x 2 time points x 2 groups = 40 implant site**.[15,16] Any withdrawal for any processing error was compensated for by replacement to control for attrition (withdrawal) bias.[17]

Software:

The sample size was calculated using online Power (sample size) calculators https://www.sealedenvelope.com/power/binary-noninferior/[16]

Animals

This study used 20 mature New Zealand white rabbits weighing 2.5 - 3.5 kg. They were collected from the animal house of the City of Scientific Research and Technological Application in Burj AL Arab, Egypt. Animals were housed in the experimental animal house at Alexandria University's Faculty of Medicine under the same nutritional and environmental circumstances. All animal procedures adhered to the National Research Council Guidelines for the Care and Use of Laboratory Animals [18]. The research protocol was approved by the institutional experimentation and Animal Ethical Committee of Alexandria University., (IRBNO:00010556-IORG0008839). Histological evaluation and histomorphometric analysis were done in the Department of Oral Biology, Faculty of Dentistry, Alexandria University.

Implant material:

Forty 8×4.2 mm: length × diameter Sand-blasted, acid-etched dental implants were used in this study (Dentium, Soul, Korea).

Inclusion criteria:

The selected animals were matched regarding; sex, age, weight, type of diet, and environmental housing conditions.

Exclusion criteria:

- Rabbits involved in any prior experimental research.
- Rabbits with any noticeable wounds or illnesses.

Surgical procedure

The animals were anesthetized by sodium thiopental intravenous injection (13 mg/kg) (Sandoz GmbH Biochemiestraße, Österreich, Austria). Before the surgical interventions, antibiotics (Ampicillin (Eipico, 10th of Ramadan City, Egypt) were administered intramuscularly.

Following anaesthesia induction, the region around the proximal tibia's medial sides was extensively swabbed with an iodine and 70% ethanol solution. After a 30 mm incision down the medial side of the proximal tibia, the periosteum was reached. The dissection was then carried laterally to the entire extent of the flat medial bone surface and all the way up to the inferior attachment of the knee joint capsule. Afterward, a tissue incision was made to provide access to the bone, the flap was reflected to disclose the bone tissue, and the perforations

were performed while the region was immersed in profuse irrigation using the drilling sequence set for each implant type. One implant was inserted into each tibia of each group using a drilling sequence that followed a simplified technique with a pilot lindeman drill and a final diameter drill (test group), as opposed to a drilling sequence that followed consecutive drills as directed by the manufacturer (control group). The implants were bicortically anchored; however, their cervical parts were all positioned at the cortical bone's level. 5-0 nylon was used for the sutures, which were applied using individual simple points. (Fig.1A-D)

Post-surgical procedure

The animals were fed a soft food for two weeks after surgery. The Ampicillin antibiotic (Eipico, tenth Ramadan City, Egypt) was administered intramuscularly on the first day, then combined with their diet for seven days. Intramuscular (IM) injections of (0.09 mg/lb) body weight with nonsteroidal antiinflammatory (Meloxicam DELTA PHARMA Factory Industrial Zone B4, tenth of Ramadan City, Egypt) were also administered on the first day and daily for two days.

***Animal sacrifice*:**

For each group, 10 rabbits were euthanized at 4 weeks, and the other 10 at 6 weeks, via an intramuscular injection of sodium phenobarbitone at a dosage of 60 mg/ml/kg body weight (Phenobarbitone, Fawns & McAllan Pty Ltd, Melbourne, Victoria). This was done to evaluate bone attachment to the implant surfaces.

***Animal disposal*:**

After obtaining the operated tibias, the rest of the rabbits' bodies were safely buried in specific location under soil.

Histological procedure

From each animal, the 2 tibias were removed and immersed for one week in 10% neutral buffered formalin for fixation. The specimens were decalcified in 8 % trichloroacetic acid, followed by washing in distilled water and dehydration in ascending grades of ethyl alcohol.

Removal of the osseointegrated implants from the surrounding bone was performed by making two opposite longitudinal incisions around the center of the implant and were connected by a horizontal incision at the most external border of the peri-implant bone. This allowed the separation of the implant (using very slight and cautious movement) from the surrounding two halves of the bone cylinder accommodating it. The latter were infiltrated and embedded in paraffin wax (each two halves of the bone cylinder of the same implant in one wax block with a total number of 40 wax blocks). Then longitudinal 5 microns thick serial sections of the bone border facing the spaces of the removed implants were stained with Hematoxylin and Eosin (H&E) for general evaluation of the peri-implant bone healing, bone implant outline and bone cell activity and finally to perform the morphometric analysis.

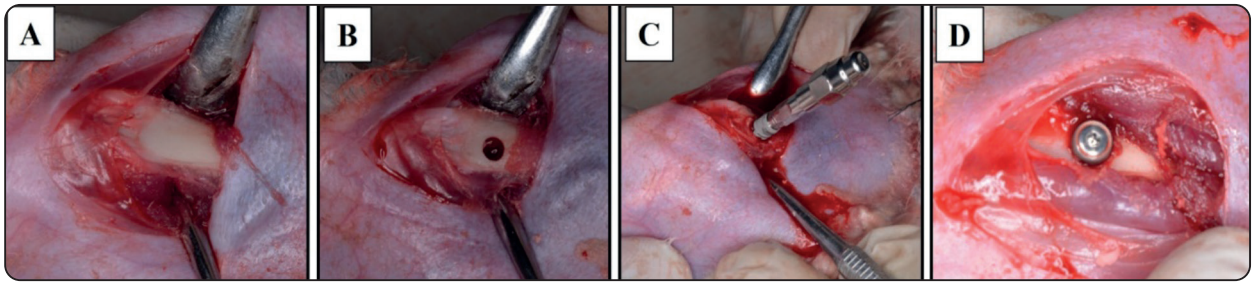


Fig. (1) (A-D) The surgical procedure for the test site prepared with single drilling technique

Histomorphometric analysis

Image analysis system (Image J software) was used to quantify the following two parameters:

a- Surface area of the newly formed bone in the spaces between the implant serrations and adjacent to the native bone of tibias.

b- Thickness of the trabeculae of this bone.

Three sections obtained from specific depths of each of the prepared wax blocks were used for calculations. The means of the values obtained from images taken from these three sections were calculated to finally get 20 measurements for each of the two variants for each group with subsequently 10 values for each observation period of each group.

Surface area of the formed bone:

Images containing the whole bone segments formed in the spaces of the implant serrations and that adjacent to the border of the native bone of tibia were captured at X100 using Optica microscope (OptikamB5, C-B5) to represent the total surface area of the field in (mm^2).

Calculation of the surface area of the spaces around the bone and those of the bone marrow and any surrounding soft tissues was performed then subtracted from the total surface area of the field to obtain the surface area of the formed bone only and this was followed by calculating the percentage of the formed bone in relation to the total surface area of the field.

Thickness of the trabeculae of the formed bone:

This was done on the same images used for calculating the bone surface area. A straight line parallel to the boundary of the bone formed in the spaces between the implant serrations facing the implant outline was drawn connecting the two closest marrow spaces in one plane. A similar line was drawn connecting the two farthest marrow spaces in the same plane. The lengths of these two lines were measured by choosing the symbol (measure) from analyze of the image J tools then recorded. The average of the two values was calculated to record a value of mean thickness of bone trabeculae in each image.

Statistical methodology

The data was collected and analyzed using the Statistical Package for Social Science (SPSS) program (version 25).[19] Parametric statistics were utilized since the Kolmogorov-Smirnov test of normality indicated no significance in the variable distribution.[20] The data was described using minimum, maximum, mean, standard deviation, standard error of the mean, and 95% CI of the mean. [21] The independent sample t test was employed to compare the two normally distributed variables that were evaluated independently. To compare two dependent, normally distributed variables, the paired t-test was utilised.[22]

An alpha level was set to 5% with a significance level of 95%. Statistical significance was tested at p value <.05[23]

Percentage change was calculated as follows:

Percentage of change (%) = measurements (after) subtracted by measurements (before) divided by measurements (before) and all are multiplied by100

Percentage change (%)

$$= \frac{\text{Measurement (after) - Meaurement (before)}}{\text{Meaurement (before)}} \times 100$$

RESULTS

Clinical observation

All implant sites experienced ordinary healing after implant insertion. Throughout the experiment, for both groups, there were no signs of infection or inflammation. After sacrifice, all implants were osseointegrated.

Histological Results

In the obtained decalcified sections, the implant space appeared facing the bone formed in contact with it. Accordingly, the outline of the bone corresponds to the implant outline before its removal from the surrounding bone and subsequently in the description of images it was simply mentioned as implant outline or bone implant interface. Really it is the presumptive or hypothetical implant outline.

First observation period (4 weeks)

Single drilling group (figs.2 A-F)

A prevailing observation of regular and smooth bone implant interface was noted along the whole implant outline. Depressions corresponding to the spaces of implant serrations appeared intervening with the formed bone projections establishing osseointegration (fig.2A).

Higher magnification of the different segments of the integrated bone revealed the formation of cancellous bone trabeculae of moderate thickness and maturity in close contact with the implant outline. The boundary of the formed bone exhibited regular, smooth, and continuous contact with the implant outline (figs.2 B&C). Extension of the blood supply from the native bone towards the newly formed bone was observed (fig.2 B).

At the most external regions of the tissues facing the implant outline, condensation of highly cellular fibrous tissues was observed between the implant integrated bone and native bone of tabia. Formation of immature bone was traced in this region that is thought to reveal continuation of the regenerative process. In focal areas devoid of bone formation adjacent to the implant outline usually a cementing line was traced completing a biological seal between fibrous tissue and the implant outline (fig.2D).

Higher magnification of the different segments of the integrated bone provided more insight of the biology of the regenerative events adjacent to the implant. The fibrous tissue enclosed between the different forming trabeculae contained many blood vessels of different calibers. Also, active osteoblasts were seen bordering the trabeculae and revealing active occurrence of the machinery needed for bone formation and remodeling (figs.2 E&F). In most of the sections examined obvious lines of continuation were traced between the integrated bone and that of tibia (figs. 2 B&C and fig. 2E).

Sequentional drilling group (figs. 3 A-F)

The general appearance of the histological picture in this group was comparable to that of the single-drilling group with slightly thinner and more spaced trabeculae of the formed bone adjacent to the implant outline and the thin walled blood vessels. Also, fibrous tissue appeared more abundant around trabeculae of uniform thickness and outstanding communication. Also, the bone implant interface appeared sharply regular with continuous bone segments adjacent to the implants, (figs.3 A-C).

The formed trabeculae were outlined by almost similar density of voluminous osteoblasts to those observed in the single drilling group and enclosed highly cellular fibrous tissue, (figs.3D-F).

Small focal areas of bone discontinuously at the bone implant interface were seen associated with cementing lines that could be followed on the bone surface facing the implant outline. Blood vessels were seen on both sides of the cementing lines. (figs.3 E&F).

Evident lines of continuation between the newly formed bone and native bone of tibia were an important observation in this group, (figs. 3 B&C).

Second observation period (6 weeks)

Generally, the main histological difference from the 4 weeks observation period was the greater amount of formed bone with decrease of the fibrous tissue in between. Also, greater figures of mature bone prevailed. Inflammatory cells could hardly be traced among the interstices of the fibrous tissue.

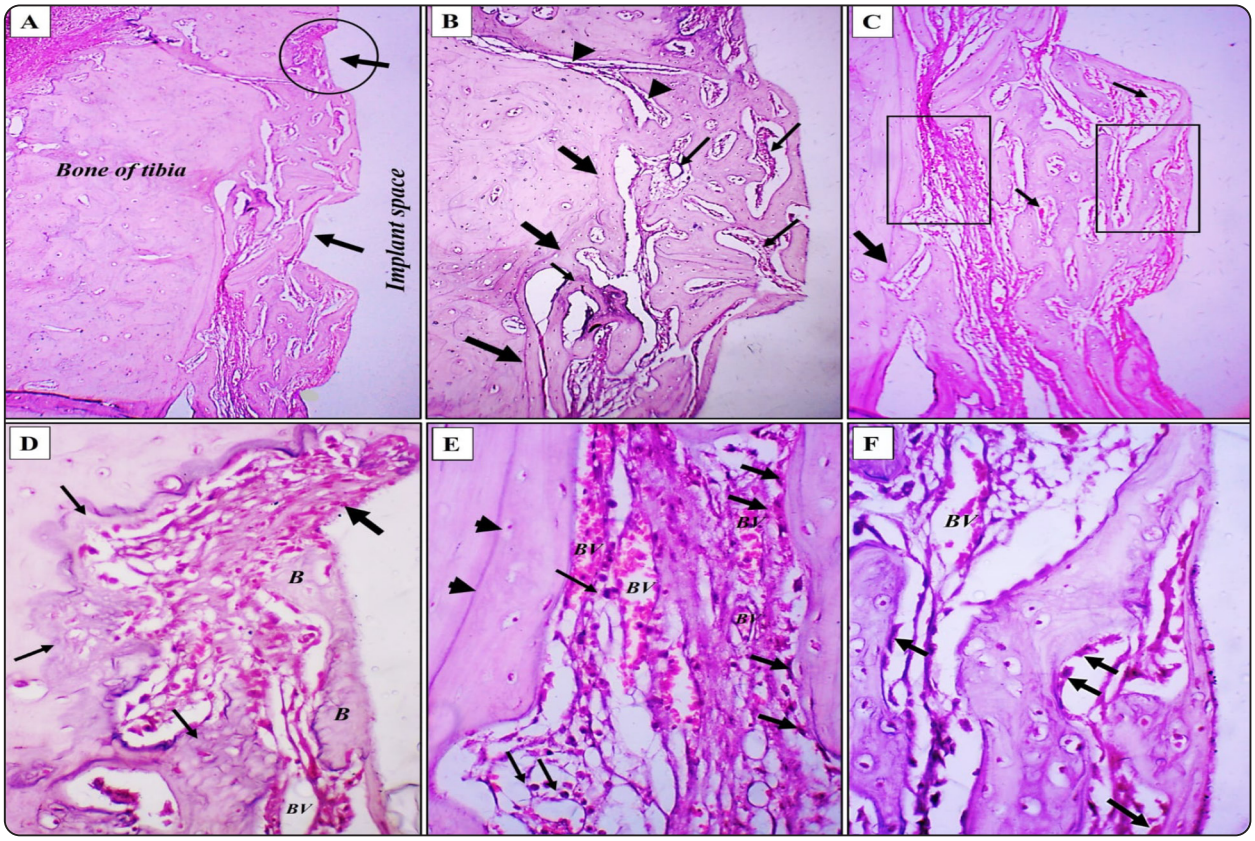


Fig. (2) [A-F, Longitudinal decalcified sections (LDSs), H&E stain (4 weeks, single drilling)]. (A): showing the projections of the newly formed bone into the spaces between the implant serrations and the intimate contact of the integration bone with the implant boundary, X:40. (B&C): Higher magnification of two successive segments of the implant outline seen in A, illustrating the thickness of the formed bone trabeculae and its intimate contact with the implant outline. Note the rich blood supply among the formed trabeculae (thin arrows) with extension from the native bone of tibia (arrowheads). In the two images, noticeable lines of continuation between the native bone and the implant integration bone are seen (thick arrows), X:100. (D): Higher magnification of the most coronal encircled segment in A revealing the formed bone adjacent to the implant outline. Note the highly cellular dense fibrous tissue, the immature bone adjacent to it (thin arrows) and the cementing line in the region devoid of bone (thick arrow), X:400. (E&F): Higher magnification of the two boxed area in D, illustrating the thickness of the trabeculae, the intervening fibrous tissue and density of the blood vessels (BV) among the formed trabeculae. Active osteoblast cells are seen on the border of the trabeculae (thick arrows). In (E) note the few inflammatory cells among the fibrous tissue (thin arrows) and the line of continuation between the native bone and the formed bone (arrow heads), X:400.

Single drilling group (figs. 4 A-F)

Like the previous observations, a regular and smooth bone-implant interface was seen. However, greater density of the formed bone was a noticeable feature (figs.4A-C)

In two successive sections, the integrated bone trabeculae appeared thick and enclosed considerable blood supply and few fibrous tissues. Few figures of immature bone could be traced and were confined to the central segments of the large trabeculae, (figs.4 B, C, E)

On the bone surface facing the implant space, cementing lines were seen, (fig. 4D). Among the formed trabeculae, an outstanding regenerative activity of the intervening osteogenic cells could be traced, they appeared voluminous, arranged in groups mapping new trabeculae or bordering the surface of the already formed ones, (fig.4F)

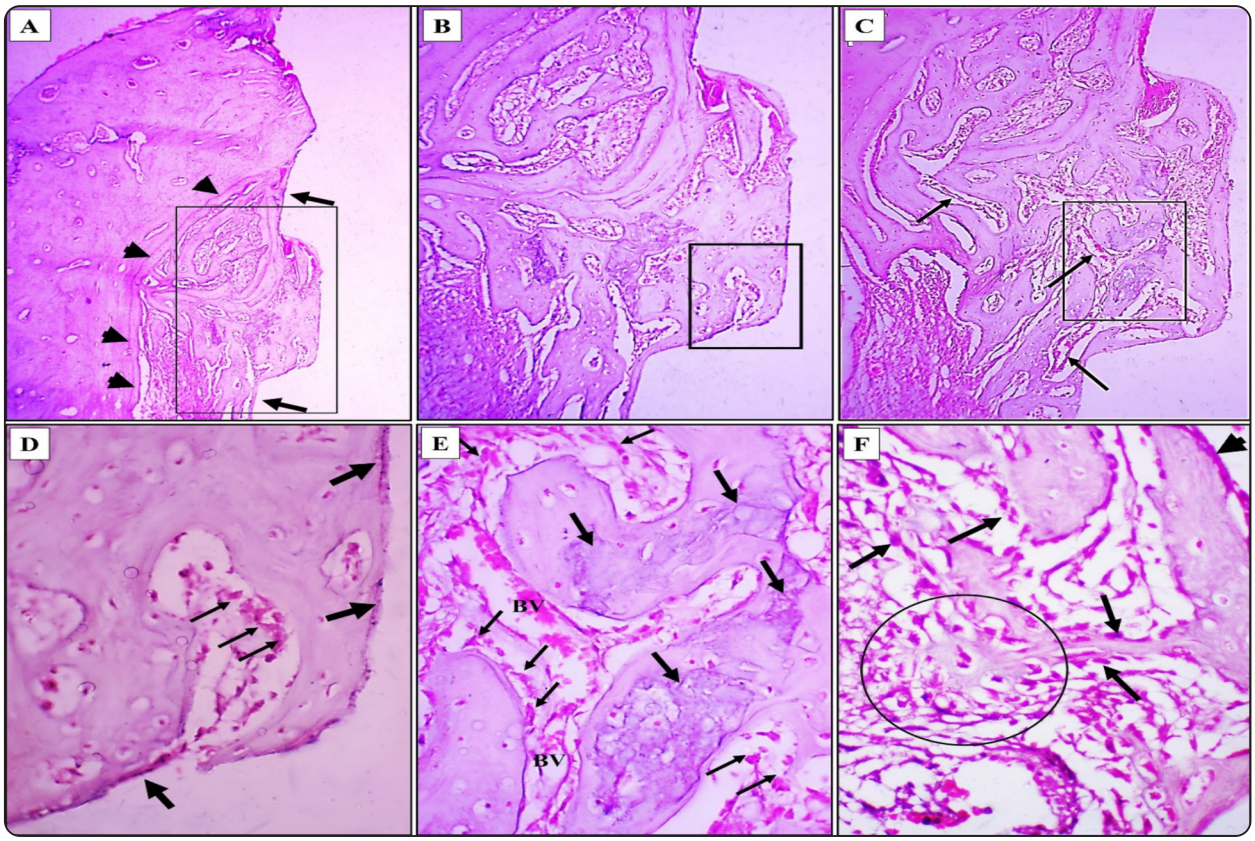


FIGURE 3: [A-F (LDSs), H&E stain (4 weeks, sequential drilling)]. (A): Showing the implant serration sites (arrows) and the integration bone formed in the spaces between the serrations and adjacent to the native bone of tibia. Note the regular and continuous outline of the bone implant interface, X:40. (B&C): Higher magnification of two successive segments of the implant integrated bone seen in A revealing slightly thinner bone trabeculae than in the single drilling group, and more intervening fibrous tissue (arrows). Cementing lines (arrowheads) are seen. Note the line of continuation between the native bone and the formed bone (chevrons), X:100. (D-F): Higher magnification of the three boxed areas (1,2,3) in C. (D): Illustrating the cellularity of the fibrous tissue and activity of osteoblasts (thick arrows). (E): Cementing lines (thin arrows. (F): High density of thin-walled blood vessels (BV) on both sides of the cementing lines with extravasated RBCs, X:400

Sequentional drilling group (figs. 5 A-F)

The density of the integrated bone appeared comparable to that of the single drilling group with an outstanding trabecular thickness and communication, (figs. A&B). The surface of the forming bone trabeculae accommodated continuous lines of active osteoblast cells providing cellular machinery for bone formation, remodeling and renewal at the deep level of the implant surface, (figs. C&D). Supporting this view was the appearance of osteocytes trapped in lacunae filled with cellular products, and osteoclast cells traced on some edges of the trabeculae completing the circle of bone cells' biology and function in these areas, (figs .5 C&D).

The formed bone exhibited a continuous interface with the implant outline, only interrupted by short threads of cementing lines, (figs, B, E and F)

Examination of the cementing lines at high magnification revealed its continuous associations with cellular elements which in some areas were organized in more than one raw. This is thought to reveal the involvement of these biological lines in a continuous process of maintenance of implant bone contact and attachment (figs.5 E&F).

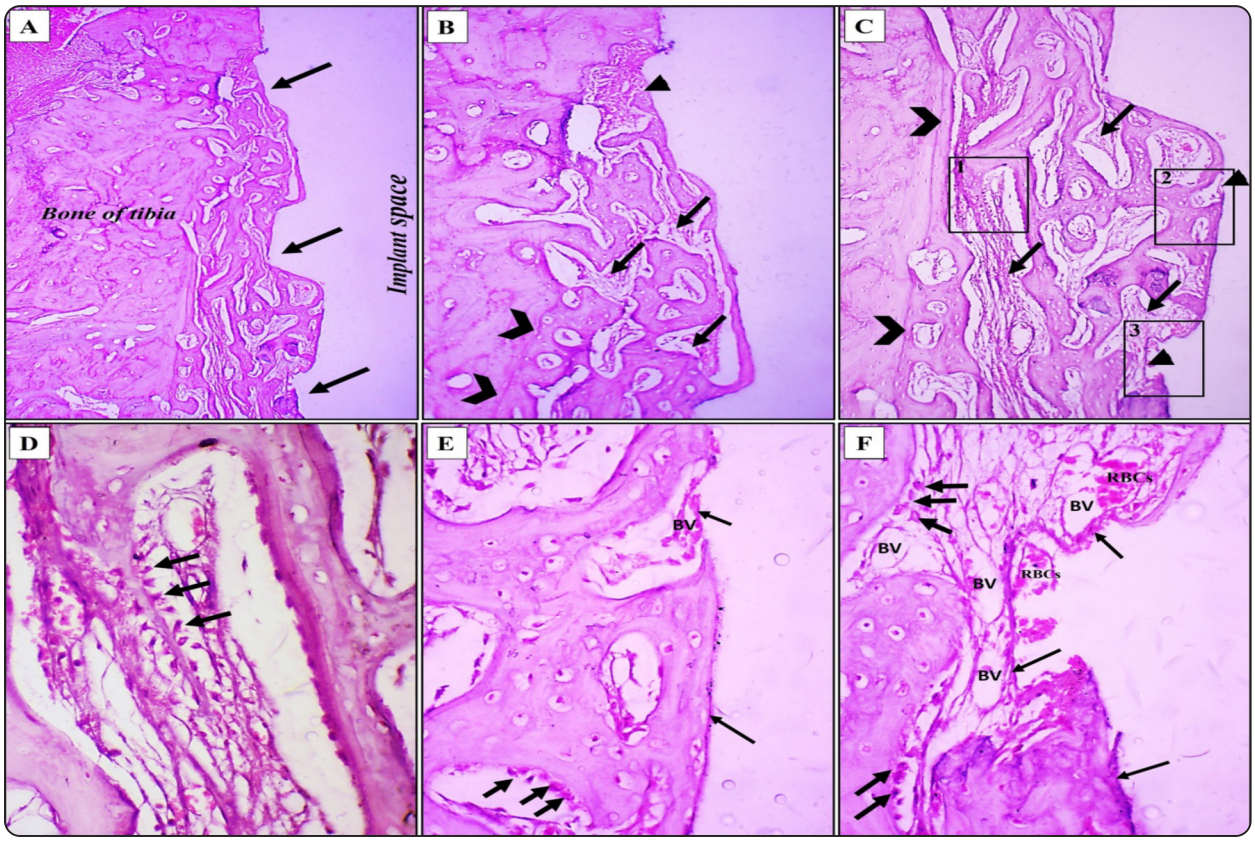


Fig. (4): [A-F, LDSs, H&E stain (6 weeks, single drilling)], (A): showing smooth and regular bone implant interface with bone formation in the spaces between the implant serrations (arrows). Note the greater density of the formed bone in comparison to that formed during the 4 weeks observation period of the same group and the continuous line of continuation between the formed bone and that of tibia (arrow heads), original magnification X:40. (B&C): High power view of the boxed area in A and (an equivalent area from another section of the same specimen taken at different tissue depth) revealing the density of the formed bone, its thick trabeculae, figures of blood supply (arrows) and fewer connective tissue than in the 4 weeks observation period. Original magnification X:100. (D): Higher magnification of the inset in B revealing the cementing line (thick arrows) on bone surface and the cells it encloses. Note the osteoblasts adjacent to the formed bone (thin arrows), X:400. (E): Higher magnification of the boxed area in C revealing the large blood vessels (BV), the immature bone in the center of the trabeculae (thick arrows) and voluminous osteoblasts (thin arrows), X:400. (F): Many voluminous osteoblasts are seen bordering the trabeculae (arrows) and surrounding a spot of active bone formation (circle). A small segment of a cementing line is observed (arrowhead), X:400

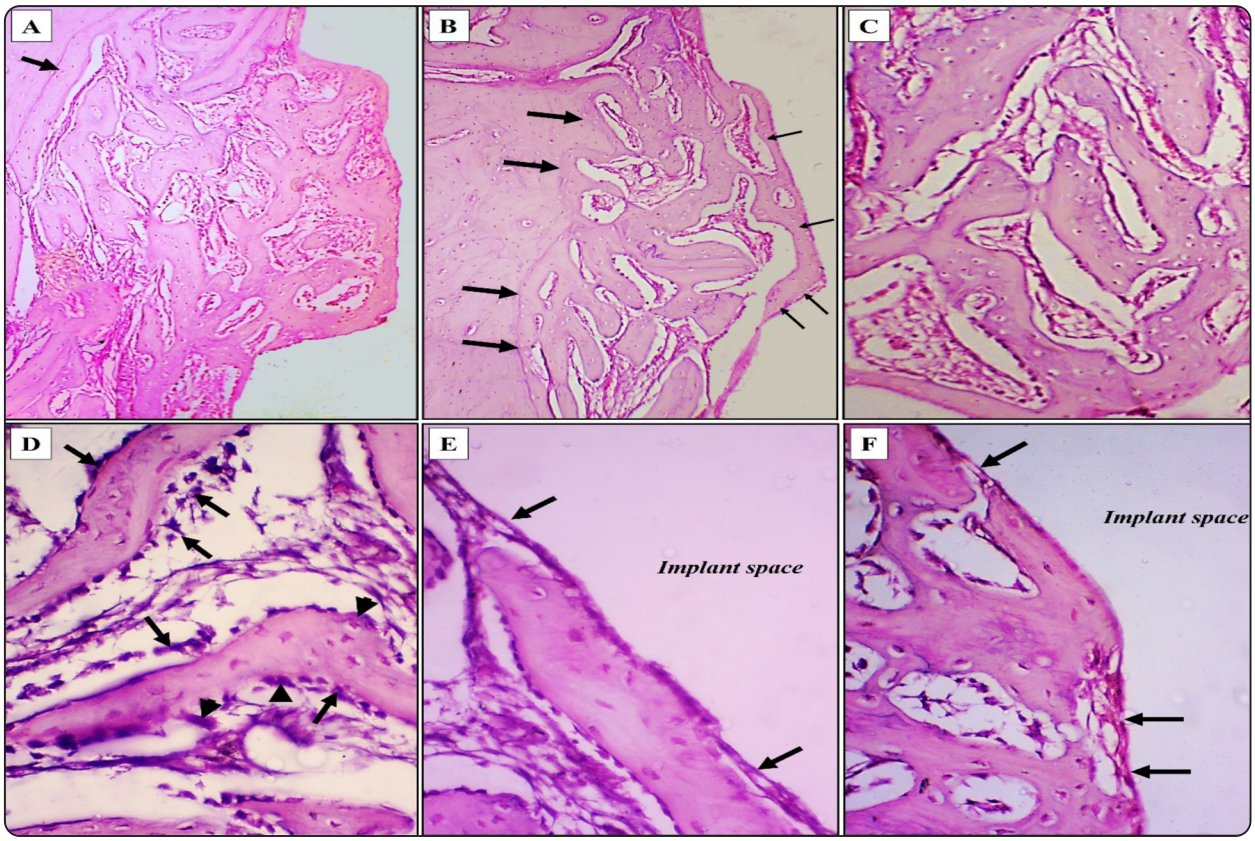


Fig. (5): [A-F, LDSs, H&E stain (6 weeks, sequential drilling)], (A&B): Showing two different segments of the implant integrated bone from sections obtained from different specimens revealing high density of the formed bone with continuous bone implant interface formed mainly of thick ribbons of bone, Obvious lines of continuation between the newly formed bone and that of tibia are seen (arrows), X: 100. (C&D): Active osteoblasts on the surface of the forming bone trabeculae (arrows) which contain viable osteocytes in their lacunae. Some osteoclasts are also seen (arrow heads), X:400. (E&F): High power views of the cementing lines on the surface of the forming bone adjacent to the implant outline and spanning the distances between the neighboring unconnected trabeculae (arrows). High cellular density is seen in association with all these cementing lines, X:400.

Histomorphometric Results

Bone surface area (mm^2):

At four weeks

In the single drilling group (n=10), the Bone surface area ranged from 35.227 to 46.591 mm^2, with a mean±SD of 41.335±3.007 mm^2, SEM of 0.951 mm^2, 95% CI of the mean 39.184-43.486 mm^2. In sequential drilling group (n=10), the bone surface area ranged from 38.017 to 51.791 mm^2, with a mean±SD of 45.455±4.084 mm^2, SEM of 1.291 mm^2, 95% CI of the mean 42.533-48.376 mm^2. The Bone surface area was statistically significantly higher in the sequential drilling group than in the Single drilling group at four weeks (p=.019).

At six weeks

In the Singel drilling group (n=10), the Bone surface area ranged from 68.504 to 75.328 mm^2, with a mean±SD of 72.99±2.200 mm^2, SEM of 0.696 mm^2, 95% CI of the mean 70.525-73.673 mm^2. The Sequential drilling group's Bone surface area ranged from 67.979 to 76.115 mm^2, with a mean±SD of 71.517±2.522 mm^2, SEM of 0.797 mm^2, 95% CI of the mean 69.713-73.321 mm^2. The Bone surface area showed no statistically significant

difference between the two studied groups at six weeks (p=.589).

The bone surface area in the single drilling group and the sequential drilling group was statistically significantly increased at six weeks compared to four weeks (p<.001 and p<.001, respectively)

Percentage change (%)

In the single drilling group (n=10), the percentage change of bone surface area ranged from 47.033 to 109.365%, with a mean±SD of 75.365±15.254%, SEM of 4.824%, 95% CI of the mean 64.453-86.277%. In the sequential drilling group (n=10), the percentage change of Bone surface area ranged from 43.419 to 100.2135%, a mean±SD of 58.685±18.062%, SEM of 5.712%, 95% CI of the mean 45.764-71.606%. The percentage change in bone surface area was statistically significantly higher in the sequential drilling compared with the single drilling group (p=.039).

The thickness of bone trabeculae (mm):

At four weeks

In the single drilling group (n=10), the thickness of bone trabeculae ranged from 0.21 to 0.29 mm, with a mean±SD. 0.25±0.03 mm, Standard Error of the Mean (SEM) of 0.01 mm, 95% Confidence Interval (CI) of the mean 0.01-0.23 mm.

In sequential drilling group (n=10), the thickness of bone trabeculae ranged from 0.18 to 0.25 mm, with a mean±SD. 0.22±0.03, SEM of 0.01 mm, 95% CI of the mean 0.01-0.20 mm.

The thickness of bone trabeculae was statistically significantly higher in the single drilling group than in the sequential drilling group at four weeks (p=.009).

At six weeks

In the single drilling group (n=10), the thickness of bone trabeculae ranged from 0.25 to 0.38 mm, with a mean±SD. 0.31±0.04 mm, SEM of 0.01 mm, 95% CI of the mean 0.01-0.28 mm. In the sequential drilling group (n=10), the thickness of bone trabeculae ranged from 0.25 to 0.37 mm, with a mean±SD. 0.31±0.04 mm, SEM of 0.01 mm, 95% CI of the mean 0.01-0.28 mm. The thickness of bone trabeculae has no statistically significant difference between the two studied groups at six weeks (p=.865).

The thickness of bone trabeculae in the single drilling and sequential drilling groups statistically significantly increased at six weeks compared with four weeks (p=.015 and p<.001, respectively).

PERCENTAGE CHANGE (%)

In the single drilling group (n=10), the percentage change of thickness of bone trabeculae ranged from 0.00 to 71.43%, with a mean±SD of 24.26±26.83%, SEM of 8.49%, 95% CI of the mean 8.49-5.07 %. In the sequential drilling group (n=10), the percentage change of thickness of bone trabeculae ranged from 4.17 to 78.95%, with a mean±SD of 42.41±26.41%, SEM of 8.35%, 95% CI of the mean 8.35-23.52%. The percentage change in thickness of bone trabeculae at six weeks has no statistically significant difference between the two studied groups (p=.145).

TABLE (1) Bone surface area (mm2) in the two studied groups

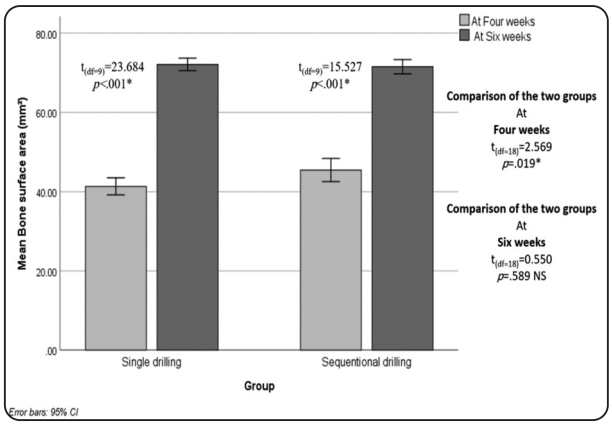
Bone surface area (mm^2)	**Group**		**Test of significance**
	Single drilling (n=10)	**Sequential drilling** (n=10)	***p* value**
At four weeks			
- Min-Max	35.227-46.591	38.017-51.791	
- Mean ± Std. Deviation	41.335±3.007	45.455±4.084	$t_{(df=18)}$=2.569
- SEM	0.951	1.291	*p*=.019*
- 95% CI for mean	39.184-43.486	42.533-48.376	
At six weeks			
- Min-Max	68.504-75.328	67.979-76.115	
- Mean ± Std. Deviation	72.099±2.200	71.517±2.522	$t_{(df=18)}$=0.550
- SEM	0.696	0.797	*p*=.589 NS
- 95% CI for mean	70.525-73.673	69.713-73.321	
Paired t-test of significance	$t_{(df=9)}$=23.684	$t_{(df=9)}$=15.527	
***p* value**	*p*<.001*	*p*<.001*	
- Percentage change (%) Min-Max	47.033-109.365	43.419-100.213	
- Mean ± Std. Deviation	75.365±15.254	58.685±18.062	$t_{(df=18)}$=2.231
- SEM	4.824	5.712	*p*=.039*
- 95% CI for mean	64.453-86.277	45.764-71.606	

n: Number of implants placed in each group for each observation period ***Min-Max: Minimum – Maximum***
CI: Confidence interval ***SEM: standard error of the mean***
**** : Statistically significant (p<0.05)*** ***NS: Statistically not significant (p≥0.05)***

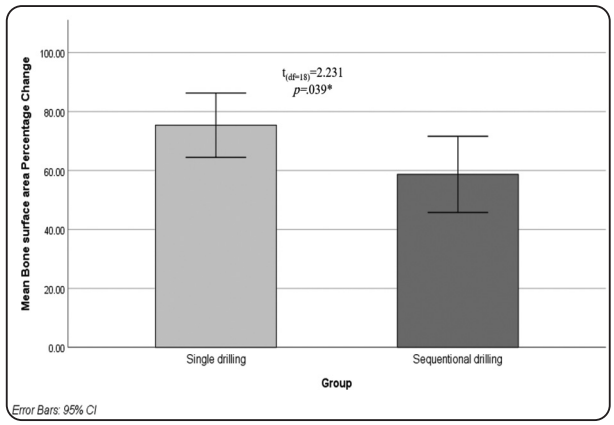
TABLE (2) The thickness of bone trabeculae (mm) in the two studied groups

The thickness of bone trabeculae (mm)	**Group**		**Test of significance**
	Single drilling (n=10)	**Sequential drilling** (n=10)	***p* value**
At four weeks			
- Min-Max	0.21-0.29	0.18-0.25	
- Mean ± Std. Deviation	0.25±0.03	0.22±0.03	$t_{(df=18)}$=2.943
- SEM	0.01	0.01	*p*=.009*
- 95% CI for mean	0.01-0.23	0.01-0.20	
At six weeks			
- Min-Max	0.25-0.38	0.25-0.37	
- Mean ± Std. Deviation	0.31±0.04	0.31±0.04	$t_{(df=18)}$=0.173
- SEM	0.01	0.01	*p*=.865 NS
- 95% CI for mean	0.01-0.28	0.01-0.28	
Paired t-test of significance	$t_{(df=9)}$=3.015	$t_{(df=9)}$=5.510	
***p* value**	*p*=.015*	*p*=.000*	
Percentage change (%)			
- Min-Max	0.00-71.43	4.17-78.95	
- Mean ± Std. Deviation	24.26±26.83	42.41±26.41	$t_{(df=18)}$=1.524
- SEM	8.49	8.35	*p*=.145 NS
- 95% CI for mean	8.49-5.07	8.35-23.52	

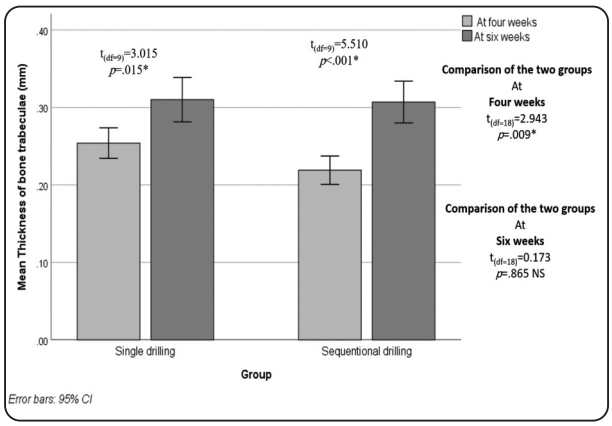
n: Number of implants placed in each group for each observation period ***Min-Max: Minimum – Maximum***
CI: Confidence interval ***SEM: standard error of the mean***
**** : Statistically significant (p<0.05)*** ***NS: Statistically not significant (p≥0.05)***



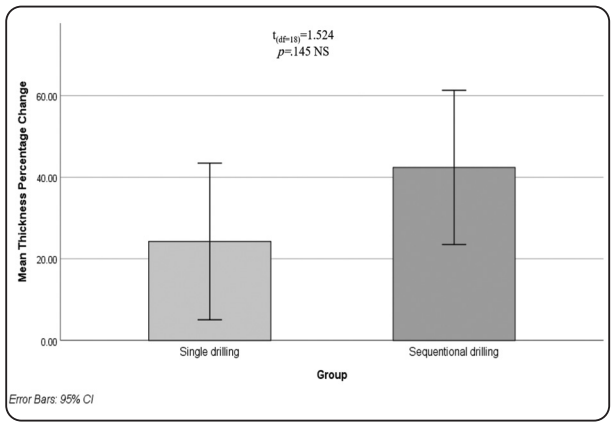
Graph (1) Clustered bar chart of Mean bone surface area (mm2) in the studied groups



Graph (2) Box and whisker graph of Mean bone surface area percentage change (%) in the studied groups



Graph (3) Clustered bar chart of the Mean Thickness of bone trabeculae (mm) in the studied groups



Graph (4) Box and whisker graph of the Mean Thickness of bone trabeculae percentage change (%) in the studied groups

DISCUSSION

Simplified, fast osteotomy preparation is the intention of the new era of implant placement in terms of patient-reported outcomes. The length of time that tissue is exposed during surgery is shortened because of this simplified approach. therefore decreasing tissue morbidity ,postoperative discomfort, and; improves patient satisfaction. [1] It was for this reason that the current study was carried out to compare the drilling sequence using a simplified protocol with a pilot lindeman drill and final diameter drill (test group) with a drilling sequence using consecutive drills as per manufacturer instructions (control group). A total of 20 drilling procedures and implant placements were performed for each group, totaling 40 titanium dental implants. The samples were further divided into two observation periods 10 implants from each group were removed 4weeks after implantation (first observation period) and 10 implants were removed 6 weeks post implantation (second observation period). Histological and histomorphometric analysis were carried out 4 and 6 weeks after the implantation.

The current method of examining the implant bone interface in demineralized sections by

removing the implant from the surrounding bone prior to specimen embedding has proved to be a convenient method for examining this interface without the need for using the complicated procedure and equipment for cutting undemineralized sections and which allow only examination of the interface at one level of implant circumference. Accordingly, this method allowed examination of serial sections of the interface almost all over its circumference. Also, it has provided an insight of the bone configuration deeper to the level of the implant bone interface horizontally in serial sections.

The current histological observations revealed an excellent biological interaction of osseointegration between the SLA implants and the bone of rabbit tibia with considerable figures of bone formation and identical pattern of trabeculation in both groups and the two observation periods.

The greater amount of formed mature with decreased remodeling figures in the second observation period clearly reflected the cumulative response of bone formation with proceeding in time and this was confirmed by the results of the morphometric analysis. Also, the limited figures of inflammatory cells in histological sections of the second observation period are thought to reflect progressing stability of synthetic activity of the tissue by the associated cells and their biological interaction.

The regular and straight lines of bone implant interface clearly reflected the intimate contact between both and hence strong osseointegration which was also detected clinically.

The noted continuous and regular configuration of the cementing line in sections of both groups, especially of the second observation period and its association with increased density of active cells reveal its equal exerted role in both groups. The cementing line is [an extracellular non collagenous proteins (specifically osteopontin and bone sialoprotein) and proteoglycans from the plasma (osteonectin)]. It enhances the biological responses promoting cellular adhesion, migration, and differentiation at bone implant interface. Osteopontin and bone sialoprotein have nucleation sites for calcium phosphate mineralization Thus cement line forms a non-collagenous, calcified layer that covers and continues along the implant surface cementing it with the adjacent bone.[24]

In the current study newly formed peri-implant bone trabeculae that developed from the bone side towards the implant surface (distance osteogenesis, figs 2 (B, C, D) and figures 4 (A, B, C) and 5(A&B) and peri-implant bone healing that developed from the implant to the bone (contact osteogenesis, figs3 B, 4 B&C and 5B), were traced in association with both implants inserted by single or sequential method. This is thought to reduce the hesitation about the choice between either method.

On the other hand, the results of the histomorphometrical analysis of the present study showed that the bone surface area in contact to implant was statistically significantly higher in the sequential drilling group than in the single drilling group at four weeks (p=.019). While the results showed no statistically significant difference between the two studied groups at six weeks (p=.589). However, the bone surface area in the single drilling group and the sequential drilling groups was statistically significantly increased at six weeks compared to four weeks (p<.001 and p<.001, respectively) (table1, graph1)

The thickness of bone trabeculae was statistically significantly higher in the single drilling group than in the sequential drilling group at four weeks (p=.009), while showed no statistically significant difference between the two studied groups at six weeks (p=.865). However ,The thickness of bone trabeculae in the single drilling and sequential drilling groups statistically significantly increased at six weeks compared with four weeks (p=.015 and p<.001, respectively).(table2,graph3)

The current findings are comparable to the results of several studies which revealed that bone tissue behavior around implants placed using a simplified

approach is equivalent to theat utilizing standard protocol employing multiple sequential drills[6,8,9, 13]

Giro and coworkers evaluated the effects of pilot drill + final diameter drill versus sequential drilling on the osseointegration of dental implants installed in beagle dogs' tibia in 2013 [13]. Their findings demonstrated that, throughout various observing times—1, 3, and 5 weeks—both approaches resulted in implant integration with no differences in terms of bone-to-implant contact (BIC) and bone-area-fraction occupancy (BAFO). They came to the conclusion that the traditional protocol and the simplified drilling protocol both produced comparable osseointegration results.

In a study utilizing a rabbit tibia model, Gehrke 2018[12] compared the impact of single drill versus multiple sequence drill on peri-implant bone behavior and osseointegration. Their research revealed that employing a single drill for the osteotomy produced a comparable histology (bone-to-implant contact percentage) and biomechanical (in terms of Resonance frequency analysis (RFA) and removal torque test (RTt)) response to using a standard multiple drilling.

The majority of studies focusing on the simplification of drilling protocols have generally found no differences over time between groups.[8,11] However, a beagle experiment study revealed that the simplified drilling group showed more favorable bone reactions than the standard drilling group one week after implant insertion. These findings were interpreted as indicating less harm to the cortical bones in the area when simpler techniques were used. [25]

Furthermore, a recent study by Paolo Trisi et al. in 2020[26] demonstrated that single drill triggers bone corticalization more than those seen in conventional drilling in terms of both bone to implant contact and bone volume, and that these differences were statistically significant in favor toin the single drilling group.

In light of the finding that osteonecrosis via heat transfer occurs when temperatures more than 47˚C are created within the bone for more than 1 minute, the results of our investigation and the aforementioned histology studies can be explained.[27] It is well recognized that this heat slows the healing of bones. Simplified drilling procedures[28,29] do not produce heat levels that affect the nearby tissues.[13] In addition; speeding up and simplifying the drilling protocol decreases the osteotomy site temperature [30] provided that sharp drills with copious irrigation was maintained all over the procedure [31]

These histologically evidenced results are supposed to improve both clinical and patient reported–outcomes.

The clinical results of implants inserted at sites prepared with a single drilling process were evaluated by Bettach et al. in 2015 [1]. The key factors evaluated were implant survival, peri-implant bone level change, and patient satisfaction. No patient dropouts were detected during the follow-up period (which lasted between 12 and 27 months), and a mean implant survival rate of 98.0% was noted. There were no biological or mechanical issues. Every patient showed complete satisfaction. They came to the conclusion that using a single bur for implant site preparation allowed for a shorter surgical procedure without sacrificing clinical results.

In order to compare the stability of implant placements after simplification of the drilling process (just an initial and final drilling) to that after a typical drilling sequence, Kim et al. undertook a prospective clinical trial in 2019[31]. They came to the conclusion that simplifying the drilling routine will not impede the osseointegration process and are anticipated to help advance therapies for implants in the future.

In a randomized clinical study, Zahran et al. 2020, [32] compared the use of a recently invented single drill to sequential drills for implant placements, and they found no statistically significant difference between the two groups in terms of implant success, bone loss, or patient satisfaction.

One potential limitation is that, in comparison to the sequential drilling procedure, the simplified protocol may have increased the likelihood of uneven bone preparation for implant placements. Since misalignments cannot be fixed, a higher level of precision is needed. As a result, in order to boost stability, it would be crucial to develop a better level of experience in employing the simplified protocol or make use of a surgical guide during the installation process.

Various designs for the fast osteotomy drills were used (Zahran[31], Kim[30], Paolo[26], Emad[33]). So further histological and clinical studies are recommended to be conducted in order to compare between those disparate designs.

CONCLUSION

Single drilling osteotomy did not differ from sequential drilling in terms of the proposed and evaluated test parameters for evaluating peri-implant behavior.

The use of a single drill in a fast, simplified osteotomy procedure could yield outstanding outcomes for the patient and the surgeon. For the surgeon in terms of accelerating the surgical process and simplifying the approach for preparing the implant site. And for the patient as well, since a shorter recovery period and less postoperative pain may result in improved acceptance of the implant therapy.

REFERENCES

1. Bettach R; Taschieri S; Boukhris G; Massimo Del Fabbro M D: Single Drilling for Implant Site Preparation, Clinical Implant Dentistry and Related Research, Volume 17, Number 1, 2015
2. Karaca F, Aksakal B. Effects of various drilling parameters on bone duringimplantology: an in vitro experimental study. Acta Bioeng Biomech.2013;15(4):25-32.
3. Misch CE, Qu Z, Bidez MW. Mechanical properties of trabecular bone in the human mandible: implications for dental implant treatment planning and surgical placement.J Oral Maxillofac Surg 1999; 57:700–706.
4. SharawyM,Misch CE,Weller N, Tehemar S.Heat generation during implant drilling: the significance of motor speed.J Oral Maxillofac Surg 2002; 60:1160–1169.
5. Brägger U, Wermuth W, Török E. Heat generated during preparation of titanium implants of the ITI Dental Implant System: an in vitro study. Clin Oral Implants Res 1995;6:254–259. (Betach4)
6. Gehrke S, Bettach R, Taschieri S, et al. Temperature changes in cortical bone after implant site preparation using a single bur versusMultiple drilling steps: an in vitro investigation. Clin Implant Dent Relat Res 2015; 17:700e7.
7. Carvalho AC, Queiroz TP, Okamoto R, et al. Evaluation of bone heating, immediate bone cell viability, and wear of highresistance drills after the creation of implant osteotomies inrabbit tibias. Int J Oral Maxillofac Implants 2011;26:1193e201.
8. Patel A, Gil LF, Castellano, A, et al. Effect of simplified one-Step drilling protocol on osseointegration. Int J Periodont Rest 2016; 36:82-87.
9. Jimbo R, Giro G, Marin C, et al. Simplified drilling technique does not decrease dental implant osseointegration: a preliminary report. J Periodontol 2013;84:1599-605.
10. Gil LF, Sarendranath A, Neiva R, et al. Bone healing around dental implants: simplified vs conventional drilling protocols at speed of 400 rpm. Int J Oral Maxillofac Implants 2017;32:329e36.
11. Guazzi P, Grandi T, Grandi G. Implant site preparation using a single bur versus multiple drilling steps: 4-month post-loading results of a multicenter randomised controlled trial. Eur JOral Implantol 2015;8:283e90.
12. Gehrke S, Bettach R, Aramburú J, Prados-Frutos C , Del Fabbro M and J. A. Shibli. Peri-Implant Bone Behavior after Single Drill versus Multiple Sequence for Osteotomy Drill BioMed Research InternationalVolume 2018, Article ID 9756043, 8 pages https://doi.org/10.1155/2018/9756043
13. Giro G, Tovar N, Marin C, Bonfante EA, Jimbo R, Suzuki M, et al. The Effect of Simplifying Dental Implant Drilling Sequence on Osseointegration: An Experimental Study in Dogs. International Journal of Biomaterials 2013;2013.
14. Gehrke S. A., J. L. C. Guirado, R.Bettach, M.D. Fabbro, C. P.-A. Mart´ınez, and J. A. Shibli, "Evaluation of the insertion torque, implant stability quotient and drilled hole quality for different drill design: an in vitro investigation," Clinical Oral Implants Research, 2016.

15. Julious SA. Sample Sizes for Clinical Trials with Normal Data. Statistics in medicine 2004;23(12):1921-86.

16. Sealed Envelope Ltd. Power Calculator for Binary Outcome Non-Inferiority Trial. 2012 [cited 2021 5 August]. Available from: https://www.sealedenvelope.com/power/binary-noninferior/

17. Pannucci CJ, Wilkins EG. Identifying and Avoiding Bias in Research. Plast Reconstr Surg 2010;126(2):619-25.

18. National Research Council Committee for the Update of the Guide for the C, Use of Laboratory A. The National Academies Collection: Reports funded by National Institutes of Health. Guide for the Care and Use of Laboratory Animals. Washington (DC): National Academies Press (US) Copyright © 2011, National Academy of Sciences.; 2011.

19. IBM Corp. IBM SPSS Statistics for Windows, Version 25.0. Armonk, NY: IBM Corp.; Released 2017.

20. Field A. Discovering Statistics Using IBM SPSS Statistics. 4th ed. London, California, New Delhi: SAGE Publications Ltd; 2013.

21. Greenland S, Senn SJ, Rothman KJ, Carlin JB, Poole C, Goodman SN, et al. Statistical tests, P values, confidence intervals, and power: a guide to misinterpretations. Eur J Epidemiol 2016;31(4):337-50. doi: 10.1007/s10654-016-0149-3.

22. Box JF. Guinness, Gosset, Fisher, and small samples. Statistical science 1987:45-52. doi:

23. Curran-Everett D. Evolution in statistics: P values, statistical significance, kayaks, and walking trees. American Physiological Society Bethesda, MD; 2020. p. 221-4.

24. Smeets, R., Stadlinger, B., Schwarz, F., Beck-Broichsitter, B., Jung, O., Precht, C., & Ebker, T. (2016). Impact of dental implant surface modifications on osseointegration. BioMed Research International, 2016.

25. Jimbo R, Janal MN, Marin C, et al. The effect of implant diameter on osseointegration utilizing simplified drilling protocols. Clin Oral Impl Res 2014;25:1295e300.

26. Trisi, P Falco A and Berardini M Single-drill implant induces bone corticalization during submerged healing: an in vivo pilot study International Journal of Implant Dentistry (2020) 6:2 https://doi.org/10.1186/s40729-019-0198y

27. Eriksson AR, Albrektsson T. Temperature threshold levels for heat-induced bone tissue injury: a vital-microscopic study in the rabbit. J Prosthet Dent 1983;50:101e7.

28. Eriksson RA, Adell R. Temperatures during drilling for the placement of implants using the osseointegration technique. J Oral Maxillofac Surg 1986;44:4e7.

29. Yoshida K, Uoshima K, Oda K, et al. Influence of heat stress to matrix on bone formation. Clin Oral Implants Res 2009;20: 782e90

30. Rafel SS. Temperature changes during high-speed drilling on bone. J OralSurg Anesth Hosp Dent Serv. 1962;20:475-477.

31. Kim H.M. , Cho J.Y. Ryu y, J. Evaluation of implant stability using different implant drilling sequences Journal of Dental Sciences (2019) 14, 152e156

32. Amr Zahran, Ahmed Mortada, Basma Mostafa,Implant placement using a newly designed single drill versus conventional sequential drills june 15, 2020 by implant practice us

33. Emad A. Senada1 , Samraa A. El Sheikh, Mervat M. Evaluation of success of single drilling implant system (clinical and radiographic study) adjalexu.2020.86759 doi: 10.21608/