Effect of Platelet-Rich Fibrin on Implant Stability

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ABSTRACT

Background: Preparation of platelet-rich fibrin (PRF) is a simple, low cost and minimally invasive method to obtain a natural concentration of autologous growth factors that is widely used to accelerate soft and hard tissue healing, thus, PRF is used in different fields of medicine. The aim of this study was to evaluate the effect of local application PRF on stability of dental implants.

Materials and methods: nineteen healthy patients with adequate alveolar bone with two or more adjacent missing teeth and/or bilaterally symmetric to the midline (split-mouth design) missing teeth participated in this study. Each patient received at least two dental implants (Dentium Co., Korea). After surgical preparation of the implant sockets, the PRF was applied randomly into one of the implant socket before the placement of implant fixture (study group), while the second implant was inserted without PRF (control group). The implant stability was measured by resonance frequency analysis (RFA) using Osstell™ ISQ, at the time of surgery (primary stability), and at 4, 8 and 12 weeks postoperatively (secondary stability).

Results: Although in the three records of secondary stability, the mean implant stability quotient (ISQ) in the study group was higher compared to the control group, this elevation was statistically not significant (P value > 0.05). On the other hand, PRF showed a significant effect on implants stability by 2.367 folds for implants that achieved primary stability ≥ 70 and maintained this stability after 12 weeks.

Conclusions: Within the limitations of this study, local application of PRF exhibited that there was no statistical beneficial effect on implant stability. No significant correlation was found between local bone density and implant stability in both groups.

Key words: Platelet-rich fibrin, stability, dental implant, resonance frequency analysis.**(J Bagh Coll Dentistry 2017; 29(4): 58-64)**

INTRODUCTION

The worthy of modern dentistry is to reestablish the patient's facial contour, masticatory, speech, esthetic and function after tooth or teeth extraction. Many methods were used for the replacement of missing tooth/teeth with natural or synthetic substitutes since centuries, however all these restorations take the support from the adjacent teeth and many problems occurred due to these replacement methods. The recent modality for the replacement of missing teeth is dental implants. Endosseous dental implant is like the natural tooth root that restores the missing teeth without the need for the adjacent teeth for support and the basic advantage of implants is to preserve the alveolar bone like the healthy tooth[(1)].

Extensive work by Brånemark who discovered that commercially pure titanium when placed in a suitably prepared site in the bone could become fixed in place due to close bond between the two. A phenomenon that later described as osseointegration, from that time many researches were done to influence the osseointegration process by studying the implant design, host site, surgical technique and loading time. In addition, many materials inserted in the prepared sites immediately before the insertion of the dental implants in order to enhance and reduce the time of osseointegration[(1)].

Recently applications of platelet-rich products; platelet rich in growth factor (PRGF), platelet rich plasma (PRP) and the platelet rich fibrin (PRF) have been proposed as an aid to enhance regeneration of osseous and epithelial tissues in oral surgery[(2)]. Studies showed that the application of these materials on titanium implants enhance the bone-implant contact (BIC) and hastening the osseointegration[(2,3)]. Platelet rich fibrin is a second-generation platelet concentrate, developed in France by *Choukroun et al. in 2000* and defined as an autologous leukocyte and platelet-rich fibrin biomaterial. It represents a new step in the platelet gel therapeutic concept, which attempts to accumulate leukocytes, platelets and released cytokines in a fibrin clot, which is widely used to accelerate soft and hard tissue healing[(4)]. Unlike other platelet concentrate, the PRF preparation is simple protocol made by centrifugation of natural blood without additives[(5)].

Formation of a fibrin scaffold is the first step in peri-implant bone healing. Platelets stick to this fibrin and are activated over the implant surface. Activated platelets release many growth factors locally as; bone morphogenetic proteins (BMP), platelet-derived growth factor (PDGF), insulin like growth factor (IGF), vascular endothelial growth factor (VEGF), transforming growth factor-β1and β2 (TGF-β1,and TGF-β2), that accelerate the healing process by attracting undifferentiated mesenchymal cells to the injured site[(6)].

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In an animal study, it was observed that more rapid healing process and bone formation observed in implants placed with PRF than in the control implant, as proved by histological examination, in addition, immunohistochemical findings revealed a high positive expression for IGF and PDGF in implants placed with PRF in comparison to control one[7].

Recently in a clinical study done by *Öncü & Alaaddinoglu in 2015* they observed that PRF application appeared to increase implant stability during the early healing period, as evidenced by higher implant stability quotient (ISQ) values and they stated that simple application of this material seems to provide faster osseointegration[3]**.** The hypothesis of this study based on the ability of local application of PRF to improve implant stability.

MATERIALS AND METHODS

Nineteen patients; 12 females and 7 males with age range of 28-66 years initially participated in this prospective clinical study, at the Dental Implant Clinic at the Department of Oral and Maxillofacial Surgery/College of dentistry/ Baghdad University, during the period from December 2015 to July 2016.

They received 58 implants divided into; 29 implants for each group (control and study group), each patient received at least two dental implant fixtures at the same session in the same edentulous region or bilaterally symmetric to the midline (split-mouth design), with the same or nearly the same diameter and length of implant fixtures. The PRF was applied to one of the implant socket immediately before the placement of implant fixture to serve as a "study group" and the second fixture placed in the implant socket without PRF, to serve as a "control group". The patients who were enrolled to this study fulfilled the inclusion and exclusion criteria and screened for fitness to participate in the existing clinical trial. The inclusion criteria were as follow: patients ag $\geq$ 18 and $\leq$ 70years, healed edentulous area for at least 6 months after extraction, the edentulous alveolar ridge span should receive at least two or more adjacent implants on one side or bilaterally symmetric to the midline of the same jaw and the implant sites should have a suitable height and width to avoid dehiscence and fenestration. Exclusion criteria were; insufficient bone volume, parafunctional habits, smoking more than 10 cigarettes per day, excessive consumption of alcohol, localized radiotherapy, tumor and metastatic disease, chemotherapy, current corticosteroid or bisphosphonate use, pregnancy, and poor oral hygiene. The patients were informed about the study timetable in detail, and a written informed consent was obtained.

PRF Preparation:

Platelet rich-fibrin preparation started with minimally invasive venipuncture technique using 21-gauge needle. The preferred vein chosen for venipuncture is the larger and fuller median cubital vein in the antecubital fossa.

Blood samples were collected in 10-ml plain blood collecting tube **(**AFCO, Jordan) and immediately centrifuged at 3000 rpm for 12 minutes. Approximately five-milliliter of the collected blood was used as a standardized amount for each study osteotomy site to get PRF clots each have the same size and features in case of multiple study fixtures ***Fig (1).***

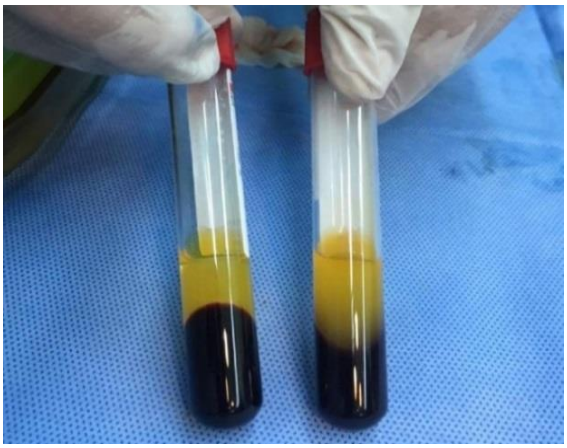

Figure (1): Identical PRF clot obtained from 10ml of blood (5 ml for each tube).

The fibrin clot that was formed in the middle part of the tube was picked up and the remnants of red blood cells milked off with tweezer. The fibrin clot was placed on clean gauze that have been wetted with normal saline and compressed gently to be ready for installing in the prepared implant site ***Fig (2).***

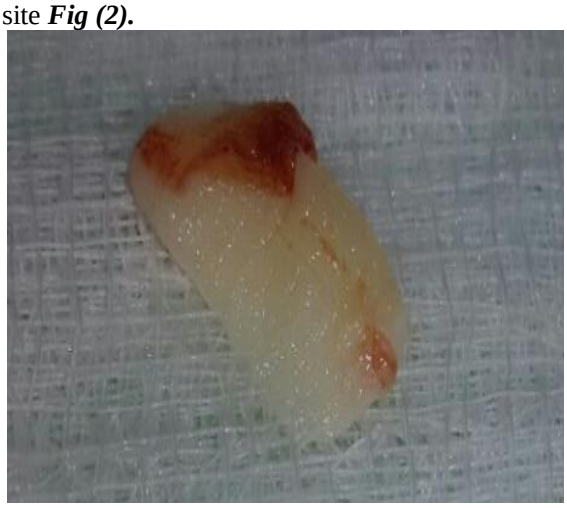

Figure (2): PRF clot after compression.

Stage one Surgery (Implant placement):

The surgery started by anesthetizing the area with local infiltration technique (Septodent, France), then crestal incision preformed slightly lingualy or palately to the crest of the ridge and an extensive three-sided mucoperiosteal flap performed and reflected. After exposing the implant site, the implant osteotomy was prepared according to recommendations of (Dentium, Korea) implant system. In addition, the resistance of bone to drilling through the preparation of the implants holes was categorized and documented as a bone type for each drilling site by the same surgeon according to *lekholm and zarb classification, (1985)*[8]**.**

The osteotomy sites were thoroughly irrigated with normal saline before the placement dental implants, PRF clot gently introduced into one site (study implant site) and the other site left without PRF (control implant site) and the implants placed at a level with or just below the crestal bone level ***Fig (3).***

After the implants placement, primary implant stability for both implants (study and control implants) was measured by Osstell^{TM} ISQ and two readings of the ISQ values were recorded; in a bucco-lingual and in mesio-distal directions. The mean value of the two ISQ measurements was used.

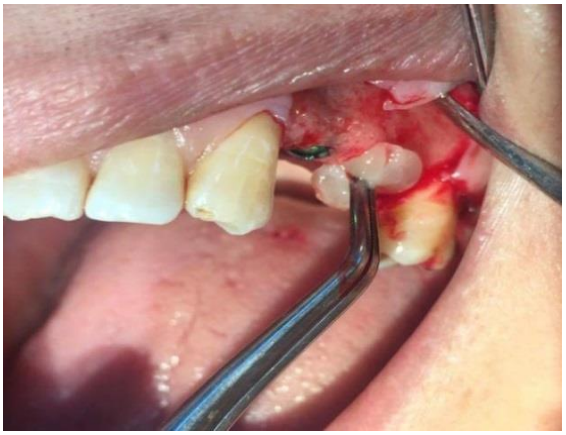

Figure (3): Application of PRF clots at site No.13.

Postoperatively, patients were advised to apply cold pack extraorally for 8 hours. The patients medicated with; antibiotic (Amoxicillin capsule 500mg three times daily or azithromycin tab 500mg one time daily for allergic patient and metronidazole tablet 500mg three times daily), analgesic (Paracetamol tablet 500 mg as required) and antiseptic oral rinse (0.12% chlorhexidine gluconate) three times per day for a week, and the suture removed after 10 days.

Second stage Surgery and Follow up Visits:

At the fourth week, the second stage surgery was done using XTS 4.0 soft tissue punch (Dentium., Korea) to excise the gingiva covering the top of the implant fixture under local anesthesia. The cover screws were exposed and removed and secondary implant stability measurements recorded in a similar technique of primary stability measurement. At that time a proper healing abutments (gingival former) were placed according to the site of implants, gingival thickness and inter-ridge distance.

Then, measurement of secondary implant stability was done at 8, and 12 weeks after implants placement time, after removal of the gingival former and fix the SmartPeg as in the measurement of the primary stability ***Fig (4&5).***

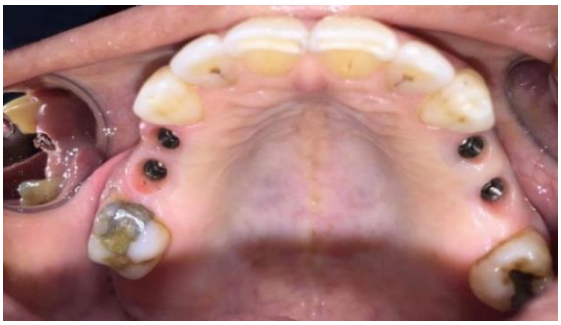

Figure (4): Detaching of healing abutments 12 weeks postoperatively.

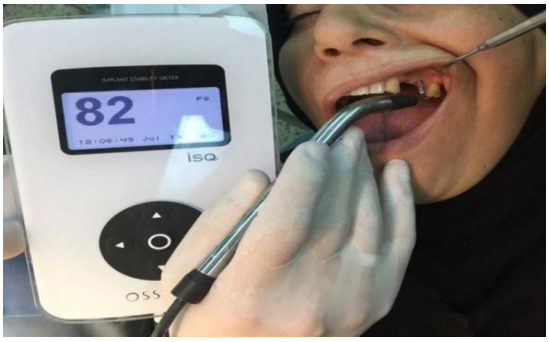

Figure (5): Secondary implants stability measurement by osstell ISQ.

Statistical Analysis:

Data were translated into a computerized database structure. All data analysis was done using SPSS version 23 software, and Minitab version 17 software packages. Paired t-Test was used to analyze continuous variables that followed prospectively in time, and to compare two groups (study and control) at two different periods, repeated measure of ANOVA test (Two ways ANOVA) used to see which of them is significantly better. Mantel-Haenszel-Cochran (MHC) Test was used to see whether control or

study groups better in achieving ISQ ≥ 70 from baseline until the end of treatment at 12 weeks.

RESULTS

From the nineteen patients who initially participated in this study, two patients had a flap dehiscence before removing the suture, both were in the control group; These two patients were managed by irrigation, refreshing the flap edges and sutured again. One case completely healed without further complication, while for the other case the control implant fixture was lost during the 4th weeks after surgery, so the case was excluded from the study, also another control implant in another patient was lost during the 4th week in this study. Therefore, two patients were excluded, so the actual number of patients who complete the study after exclusion was 17 patients with 56 implant fixtures. Making the survival rate for all patients who initially participated in this study before exclusion ***96.56% (***93.1% in control group and 100% in study group), within the limit of the study time.

Bone quality around implants was judged according to lekholm and zarb classification as follows; four implants were placed in type 1 bone density, two implants in type 2 bone, 46 implants in type 3 bone, and two implants in type 4 bone. No statistical significant correlation existed between bone density and implants stability in both groups.

Comparing the effect of healing periods on ISQ between study and control group.

Immediate postsurgical (primary stability) the mean and standard deviation values of ISQ were 73.15 ± 8.41 for the study group and 75.52 ± 4.93 for the control group. At the end of the 4th week, the mean and standard deviation values of ISQ were 68.1 ± 7.52 for the study group and 68.52 ± 8.84 for the control group. Therefore, there was a significant reduction in stability compared to the primary stability for both groups with (p=0.023) for the study group and (p=0.001) for the control group. Then from the 4th week till 8th week there was a significant increase in mean ISQ, the mean and standard deviation values of ISQ were 71.75 ± 8.08for the study group and72.48 ± 6.07 for the control group and this increase in stability was significant for both groups (p=0.009) for the study group and (p=0.005) for the control group. Finally, in the third record at the 12th week, the mean and standard deviation values of ISQ were 74.46 ± 8.06 for the study group and 75.04 ± 6.16 for the control group, displaying a significant increase in mean ISQ from the 8th week toward 12th week, (p=<0.001) for both groups ***Fig. (6).***

However, when comparing the differences in stability between the study and the control group at each time point record, these differences were statistically not significant (p=0.507).

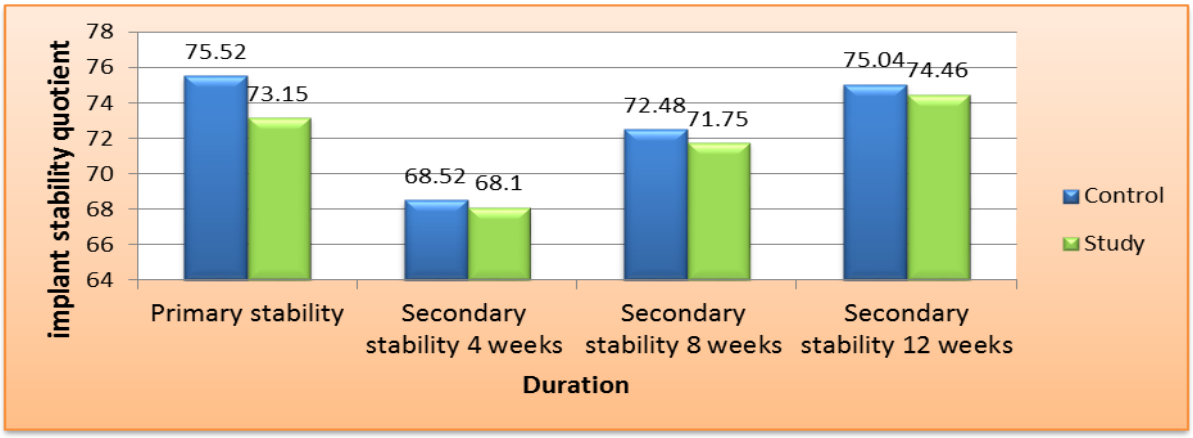

Figure (6): Comparison of the changes in mean of ISQ in both control and study group throughout the healing periods after surgery.

DISCUSSION

Nowadays, dentists are facing a new challenge in modern implant dentistry to meet the esthetic and functional expectations of the patients within a short time. Researchers have attempted to accelerate and enhance the process of osseointegration to achieve these expectations. In this study, the hypothesis was based on this concept, by using of PRF to improve implant stability by accelerate bone formation and osseointegration.

Although, the three records of secondary stability in both groups follow nearly the same pattern, but the mean of ISQ in the study group exhibited less reduction in stability than that of the control group when compared to the primary stability, especially in the early healing periods at

the 4th week record. On the other hand, when comparing the differences in stability between the study and control group at each time point record, these differences were statistically not significant (P value > 0.05).

Understanding these results may require more definitive examination methods at the histological level with a precise radiographical examination for assessment of osseointegration along with the resonance frequency analysis.

Since, in this study, the average ISQ at surgery for the control group was ***75.52*** and for the study group was ***73.15,*** indicating a high primary stability that was higher than the results obtained by different studies as *Al-Gailani & Abdul- Lateef 2015*[9] *& Öncü & Alaaddinoglu, 2015*[3]. Thus, the secondary stability will be high in most occasions. Therefore, no big statistical difference between primary and secondary stability will be measured.

Sennerby and Meredith in 2002[10] considered that ISQ ≥ 70 is a high level of stability, and predict that if the initial ISQ value is high, a small drop in stability normally levels out throughout the healing periods.

This result was in agreement with *Monov et al. (2005)*[11] who placed 34 dental implants in the mandibular arch and applied PRP to the implant osteotomies on one side, then followed the variations in the stability of the implants with RFA every 4 days until the 44th day after implants placement. It is found that there were no statistically significant differences between PRP+ and PRP– implants in terms of stability and stated that the healing process has only a little influence on future implant stability if the primary stability is high, and if the primary stability is high it can predict that secondary stability will be high in most instances. From statistical point when the study depends on RFA only, especially in cases of high primary stability, it may not reflect the real effect of the studied material[9].

Moreover, *Al-Gailani & Abdul-Lateef in 2015* [9] also found that at all time points the PRP implants have a consistently **greater** ISQ values than that of the control implants, but also it was statistically not significant.

The explanation for this result may be revealed**:**

1. The degree and effect of compression, which may damage the platelets and exude significant quantities of valuable growth factors cannot be measured. In addition**,** considerable quantities of growth factors, which are believed to be involved in tissue regeneration, are really removed by pressing. Therefore, the squeezing process could influence the quality and clinical effectiveness of the PRF preparations[12,13]**.**

2. Since the early times of development of PRF, researchers detected that the (weight and size) of the Leukocytes-PRF clot are affected by the protocol of centrifugation and the selection of centrifuge. Many studies did not use the same centrifuge and tubes and did not get the same product, even if the protocols appeared identical same gravitational force (g force) and centrifugation time[14].

Other studies in contrast with this study results, showed that PRF application significantly increase implants stability during the early healing period (1st month after implantation), and stated that simple application of this material seems to provide faster osseointegration[3].

Furthermore, another study showed that more rapid healing process and bone formation in implants with PRF than in the control implant, as proved by histological examination. Beside Immunohistochemical findings revealed high positive expression for insulin like growth factor (IGF) and platelet derived growth factor (PDGF) in experimental implant in comparison to control one[7]**.**

In this study most of the implants (43 implant fixtures) were placed in D3 bone type, and no significant correlation existed between bone density and (primary and secondary stability) in both study and control groups.

Similar results were also published by *Bischof et al. in 2004*[15] who observed that bone type did not influence implant primary stability and implant stability after 12 weeks.

While, in contrast many studies revealed that the local bone density has a dominant influence on primary implant stability, which is an important determinant for implant success[16, 17].

In Conclusions, within the limitations of this study, when comparing the control and study group throughout the three-time points follow up periods the PRF exhibited no statistical beneficial effect on implant stability. However, PRF showed a significant effect on implants stability by 2.367 folds for implants that achieved primary stability ≥ 70 and maintained this stability after 12 weeks. No significant correlation was found between local bone density and implant stability in both groups. Further clinical and histological studies are required to get more precise results about the effect of PRF on osseointegration.

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تأثير الليفين الغني بالصفيحات الدموية على ثبات زرعات الأسنان

الخلاصة

الخلفية: الفيبرين الغني بصفائح الدم (PRF) هي طريقة بسيطة ومنخفضة التكلفة وقليلة العنف للحصول على التركيز الطبيعي من عوامل النمو ذاتي التي يتم استخدامها على نطاق واسع لتسريع شفاء الأنسجة الناعمة والصلبة، وبالتالي فإنه يستخدم في مختلف مجالات الطب. والهدف من هذه الدراسة كان لمقارنة استقرار الزرعات المقاسة بتحليل تردد الرنين (RFA) لزرعات الأسنان التي أدخلت في بروتوكول جراحي على مرحلتين مع أو بدون وضع الفيبرين الغني بصفائح الدم.

المواد وطرق البحث: تسعة عشرمريضا معافى, وله عظم سنخي كاف واثنين أو أكثر من الأسنان المتجاورة المفقودة و/ أو متماثلة ثنائيا على خط الوسط (تصميم انقسام الفم), والتي اقتلعت 6 أشهر على الأقل قبل إدراجها في هذه الدراسة. على الأقل اثنتان من زرعات الأسنان (شركة العاجية، كوريا)، وضعت في كل مريض, وبعد إعداد المغارس الجراحية للزراعة، وضع الليفين الغني بصفائح الدم عشوائيا في واحدة من مغارس الزرعة قبل وضع الزرعة (مجموعة الدراسة) ، بينما المغرس الثاني زرع بدون وضع الليفين الغني بصفائح الدم (مجموعة السيطرة). استقراريه الزرعات قيست بواسطة تحليل تردد الرنين (RFA)باستخدام Osstell™ ISQ (غوتنبرغ ,السويد, الجيل الرابع), أولا قيست في وقت وضع الزرعات (الاستقرار الأولي) ، وعند 4 و 8 و 12 أسبوع بعد العمل الجراحي (الاستقرار الثانوي).

النتائج: على الرغم من أن في التسجيلات الثلاثة من الاستقرار الثانوي، فأن معدل حاصل ثبات الزرعة (ISQ) في مجموعة ألدراسة (مجموعة الليفين الغني بالصفيحات الدموية) كان أعلى خاصة في فترات الشفاء المبكرة عند الأسبوع الرابع مقارنة مع مجموعة السيطرة (بدون إضاقة الليفين الغني بالصفيحات الدموية), ولكن هذا الارتفاع لا يعتد به إحصائيا (قيمة $P> 0.05$). ومن ناحية أخرى، الليفين الغني بالصفيحات الدموية أظهر تأثيرا كبيرا على عملية استقرار الزرعات بقوة 2.367 للزراعات التي حققت استقرار أولي ≤ 70 وحافظوا على هذا الاستقرار بعد 12 أسبوع.

الاستنتاجات والمقترحات: ضمن حدود هذه الدراسة, وعلى الرغم من أنه وجد بان معدل ألISQ في مجموعة الدراسة كان اعلى في جميع مراحل المتابعة خلال فترة الشفاء وكان هذا الارتفاع لا يعتد به إحصائيا. تبين أن الليفين الغني بالصفيحات الدموية أظهر تأثيرا مهم ب (2.367) ضعف للزرعات اللاتي حصلن على استقرار أساسي ≤ 70 وحافظوا على هذا الاستقرار ل12 أسبوع. لذلك يبدو أن التأثير الكامل لليفين الغني بالصفيحات كبيرا على الاندماج العظمي للزرعات ويحافظ على مستوى عال من الاستقرار.