# EXPERIMENTAL STUDY OF BONE REBUILDING IN THE PERIIMPLANTATION AREA UNDER IMMEDIATE LOADING ON DENTAL IMPLANTS

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Anatoliy M. Potapchuk<sup>1</sup>, Yevhen L. Onipko<sup>1</sup>, Vasyl M. Almashi<sup>1</sup>, Ninel V. Dedukh<sup>2</sup>, Oleksandr Ye. Kostenko<sup>1</sup> <sup>1</sup>UZHHOROD NATIONAL UNIVERSITY, UZHHOROD, UKRAINE <sup>2</sup>STATE INSTITUTION "D. F. CHEBOTAREV INSTITUTE OF GERONTOLOGY, NATIONAL ACADEMY OF MEDICAL SCIENCES OF UKRAINE", KYIV, UKRAINE

#### ABSTRACT

**The aim:** Study of the dynamics of morphological rearrangement of bone under conditions of immediate occlusive functional load and the effect of splinting of implants with temporary orthopedic structures with the analysis of the coefficient of stability of implants during immediate implantation in the experiment.

**Materials and methods:** A series of experiments was performed on 6 male Duroc pigs at the age of 6 months and weighing 40-60 kg. In the course of recent advances, the following methods have been used: the clinical protocol of immediate – implantation of time-consuming clothes, the definition of COEFICIENT, morphometry and light microscopy of the slides, statistical analysis.

**Results:** By morphometric examination after 3 months the BIC in the series with splinting was 1.68 times higher compared to 1 month. studies, in a series of experiments without splinting – 1.9 times, after 3 months. the difference between implantation experiments with splinting components and without splinting is 1.6 times. During the functional study of the resonant – frequency analyzer, there is an increase in the ISQ in the second and third months after surgery, but this figure is higher in the study using the splint component. **Conclusions:** Stagnation of the shingle component in the case of intrinsic intraoperative functional juvenile implantation accelerates the dynamics of osteointegration, so that high indicators of the efficiency of the implant stability can be achieved.

KEY WORDS: Immediate – implantation, immediate loading, splinting, experiment, ISQ (Implant Stability Quotient), BIC (Bone to Implant Contact)

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### INTRODUCTION

As a result of monitoring and analysis of clinical and experimental studies of dental implantation success in early and long periods of observation, scientists were able to justify the feasibility of introducing into clinical practice not only classical but also modified implantation protocols with different subsequent loading times [1, 2]. At the 6th ITI Consensus Conference (International Team of Implantologists), held in Amsterdam in April 2018, a group of scientists led by German Gallucci developed a unified classification of protocols for the installation and loading of dental implants for all possible clinical situations. Based on the analysis of publications selected for monitoring that met the criteria of evidence, the calculation of implant survival rates was made to determine the success and predictability of each protocol. Immediate implantation with immediate loading (type IA) can be performed only by specialists with high manual capabilities and with extensive clinical experience in the presence of certain clinical conditions with the provision of primary fixation of the torque 25-40 N / cm, or ISQ> 70 c.u. (conventional units) provides protection of occlusal functioning of temporary restorations [3]. Today there is a tendency to reduce the duration of subsequent orthopedic rehabilitation. Immediate implantation and immediate intraoperative functional masticatory load

can reduce the duration of treatment and the volume of surgery and obtain a high functional and aesthetic result [4]. Today there are discussions about the peculiarities of the formation and level of reduction of peri-implant bone tissue in different conditions, in particular, under load and micromobility [5]. It was found that around the implants with load there is an increase in blood flow and vasomotor activity of the microcirculatory tract of supporting tissues, increase in bone volume and increase in torque, which is the optimal prognosis for accelerating the pace of osteointegration [6]. In the process of studying the dynamics of osseointegration, it was found that one of the reasons for implant disintegration is not timely loading, according to Professor Branemark, but the micro-mobility of the implant, which occurs as a result of this load [7]. Modern systematization of views on the differentiation of results of immediate loading and immediate implantation and their interpretation in terms of modern understanding of bone remodeling mechanisms expands the possibilities for discussion and argumentation of different protocols of dental implantation taking into account the initial conditions of the clinical situation [8]. Understanding the negative and early timing of implant loading under the influence of functional occlusal loading in conditions with expert analysis of morphological rearrangement of bone



Fig.1. Stages of implantation (Art Implant, Ukraine)

**Fig. 2.** Sections with implant area. Areas of bone and connective tissue in the apical (A) and cervical regions (B) of implants with and without splinting components in the apical (C) and cervical regions (D) of the implants Stained with hematoxylin and eosin. X 200.

tissue around implants is limited in basic research. There is also a lack of convincing experimental data to study the dynamics of osteogenesis at different times.

### THE AIM

Study of the dynamics of morphological rearrangement of bone under conditions of immediate occlusive functional load and the effect of splinting of implants with temporary orthopedic structures with the analysis of the coefficient of stability of implants during immediate implantation in the experiment.

### **MATERIALS AND METHODS**

*Operation on animals.* A series of experiments was performed on 6 male Duroc pigs at the age of 6 months and weighing 40-60 kg. The study was conducted in accordance with the general ethical requirements for the use of vertebrates in medical and biological experiments [10] and approved by the local committee on bioethics of Uzhhorod National University. Under the combined anesthesia (hexinal 5% -1g, relanium 10 ml, thiopental 0.5 g per 10 ml of saline) and infiltration anesthesia (Sol.Ultracain 2%) in the holes of the removed premolars and mandibular canines implants of the Art Implant system were installed (Ukraine,







**Fig. 4.** Comparative dynamics of the dependence of the coefficient of stability of implants on the mandible according to Osstell Mentor on the use of the splinting component and without splinting in the clinical protocol of implantation.

certificate of conformity № UA.TR.101-87 / SU-1019). This is a two-stage dental implant, the endosal part of which is made in the shape of a cone. The external thread is made along its entire length, gradually increasing from 0.16 mm in the cervical to 9.9 in the apical part, with a thread pitch of 1.25 mm. The profile of the threaded turns is made trapezoidal-clamping with an angle of 15 degrees (Fig. 1).

Both implantation protocols provided occlusal functional loading. On the one hand (I series of researches – 3 implants) additional installation of a temporary splinting orthopedic design was carried out that provided functional stability of implants and elimination of micromovements of each implant, on the other hand jaws (II series of researches – 3 implants) temporary orthopedic designs were not separate and provided functional stability for the micromobility of implants.

The animals received a dairy-vegetable diet for 5 days after surgery. Observations of the condition of tissues in the area of the operation were performed daily for two weeks, then until the end of the experiment – once a week.

Determination of the stability coefficient of implants. The Implant Stability Quotient (ISQ) was determined using an Osstell Mentor resonant-frequency analyzer (Integration Diagnostics, Sweden). The primary stability of the implants was also measured intraoperatively with a torque wrench from the Vitaplant system (Germany) with a measuring range from 10 to 40 N / cm in steps of 5 N / cm. ISQ (Implant Stability Quotient) was performed using an Osstell Mentor resonant-frequency analyzer (Integration Diagnostics, Sweden). The primary stability of the implants was also measured intraoperatively with a torque wrench from the Vitaplant system (Germany) with a measuring range from 10 to 40 N / cm in steps of 5 N / cm.

*Production of sections*. Animals were removed from the experiment 1 and 3 months after surgery. From the lower jaw of the animals were cut bone fragments with implants for the manufacture of sections. The samples were placed in saline according to the Technovit system, in 4% neutral formaldehyde solution. The dehydrated bone block was placed in a special form with positioning on the orientation of the cut plane. Epoxy resin was used under vacuum for 15 minutes to fix (StruersClioVac). The next day, 1 mm thick fragments were isolated using a hard tissue microtome

Investigated indicators	The bone to implant contact (BIC)	
	1 month	3 months
Implants with splinting components	44.8± 3.19	75.27±2.98
Implants without splinting components	24.25± 1.36	46.87± 2.387

**Table I.** Morphometric parameters of bone tissue on the surface of implants ( $M \pm m$ , BIC,%) with and without splinting by loading after 1 and 3 months after implantation

\*Note: Significant difference between the indicators in BIC between groups implants with and without splinting components for 1 and 3 months, and in terms of terms (p < 0.05, for U-test Manna -Whitney).

(Leitz 1600) along the longitudinal axis and glued to the slide surface using thermoplastic adhesive (IKAC-MAGHP 4). Grinding the slices allowed to thin them to 20 microns (StruersLabopol 35). Subsequently, the resulting sections were stained with hematoxylin and eosin. Qualitative characteristics of bone tissue (morphometric studies) were described in a light microscope "Optika B-383PL 40x1000xTrino" (approx. 10, vol. 20). Histological specimens were photographed with a digital camera "SIGETA M3 CMOS 14 MP USB 3.0".

Morphometric analysis of the bone-implant interface. The bone to implant contact (BIC, %) was determined in the preparations to assess the osseointegration. Morphometric studies were performed on the photographs taken under one magnification using Micros microscope (Austria) (x 200) and DSM 800 camera software (Ukraine). Morphometric studies were performed according to the recommendations of Albrektsson T, & Wennerberg A. [9].

Statistics. Numerical values were presented as mean (M) and its standard error (m). After testing the series by the Kolmogorov-Smirny method, numerical values of the implant stability coefficient were processed using the Wilcoxon T-criterion for paired samples at 1, 2, and 3 months. The Mann-Whitney test was used to assess the reliability of implant stability coefficient values when comparing groups of animals with and without splinting in two independent samples, as well as to compare indicators in the morphometric study (series of experiments at 1 and 3 months). The results of laboratory and clinical studies were processed by the methods of variational statistics with determination of the average value, its errors, the Student's t test for multiple comparisons, using Excel (MS Office 2010, Microsoft, USA) and STATISTICA 6.0 (StatSoft, USA). Differences of indicators at significance level p <0.05 were considered statistically significant.

### RESULTS

Observations of the animals showed that after implantation, the healing of the wound was without complications, and no inflammatory reaction or suppuration was recorded.

*Histological studies.* One month later, around the implants with and without splinting components immersed in the lower jaw of pigs, the process of osteogenesis prevailed in the cervical region compared to the areas in the apical region. In the conditions of splinting in the implant neck area, the newly formed bone tissue penetrated between the

threads in most areas, which contributed to the formation of a tight contact at the bone-implant interface. The foci of connective tissue were found, located along the convex part of the thread of the implant surface and in the recesses. When evaluating implants without splinting components in the endosal part of the implant revealed enlarged areas of connective tissue, alternating with small foci of newly formed bone tissue. The density of osteocytes in the bone tissue was greater around the implants with splinting. The cells were located in narrow lacunae. In the conditions of implantation without splinting, osteocyte lacunas were enlarged. Areas of bone tissue demineralization were revealed.

In the area of the thread of the implants with and without splinting, the adjacent bone tissue had characteristic signs of remodeling. Foci of overlaying of the newly formed bone tissue on the maternal bone, resorptive cavities of different sizes filled with loose connective tissue, which occupied large areas in the series of the experiment without the splinting component, were revealed. For this period of study, the BIC of implants with splinting was small, but compared with implants without splinting, the BIC was 1.85 times greater (Table I).

Three months after implantation, the area of bone tissue around the implants with splinting components were significantly larger compared to implants without splinting (Fig. 2).

After 3 months of the study in the series of implants with splinting the BIC was increased 1.68 times in comparison with 1 month, in the series of experiments without splinting – 1.9 times. The BIC difference of the implants with splinting components was 1.6 times more for this period compared to the implants without splinting (Table 1). Areas of connective tissue were present in both series (Figs. A, B, C, and D). As at the previous term, the localization of such areas was predominant in the experiment series without splinting.

Frequency-resonance analysis in determining the coefficient of stability of implants. Having stagnated the nonparametric statistical T-criterion of Vilcoxon, we improved the dynamics of the stability of the implants on the lower slit behind the Osstell Mentor because of a flexible component and without a single prototype in the basic protocol In the course of the follow-up carried out for the additional frequency – resonance analysis, we took away the onset results and the efficiency of the stability of the implants on the lower slit (Fig. 3).

The obtained results show that compared to the initial data of ISQ at the time of implantation, its values decrease sharply during the first month, so for tooth 3.4 (with splinting component) the difference is  $-3.96 \pm 1.66$  (p < 0.02), and for tooth 4.4 (without splinting component), this difference is equal to the disintegration value of the implant –  $-8.14 \pm$ 1.57 (p < 0.02). From the second month of the study there is a moderate increase in the value of CSR, which is explained by the peculiarities of the dynamics of morphological changes in osteointegration processes and is consistent with the data of other authors [11]. According to the analysis of numerical indicators according to the Mann-Whitney U-test, which is used to assess the difference between two unrelated samples (in the following case, between implants with splint components and without splints for periods of 1.2 and 3 months. results (Fig. 4).

The results show that during the first month after implantation with the splint component there was a decrease in stability in the bone-implant connection, the average value of ISQ in the study for tooth 3.3 was  $65.23 \pm 2.08$ , for tooth  $3.4 - 63.12 \pm 1.88$ , and for tooth  $3.5 - 66.44 \pm 2.12$  (p <0.03); in the study without a tire, these values were respectively: for tooth  $4.3 - 63.04 \pm 2.08$ , for tooth  $4.4 - 59.08 \pm 2.12$ , and for tooth  $4.5 - 63.23 \pm 2.08$  (p <0, 03), which corresponds to the micromobility of the implant. There is an increase in the stability of the implant in the second and third months after surgery, but this figure is higher in the study using the splint component. The average value of ISQ for tooth 3.3 in the second month of implantation is  $75.16 \pm 3.31$  (p <0.04), tooth 3.5 in the third month of implantation  $- 78.42 \pm 4.75$  (p <0.05).

# DISCUSSION

One of the most important conditions in dental implantology is the creation of conditions for successful long-term and stable fixation of implants. The success of the implantation of non-biological implants into the bone depends on osseointegration, that is, the formation of a direct structural-functional connection between the implant and the bone. Data on the peculiarities of osseointegration, processes of the state of bone tissue, formation and resorption of bone around dental implants made of metals with different surfaces and qualities attract the attention of specialists in connection with the development of various protocols for installation and loading [12]. The process of osseointegration is a regulated cascade of intracellular and extracellular biological mechanisms, in the implementation of which various modes and periods of exercise play a significant role [13]. Fundamental studies of the state of the bone tissue around the implanted material are important for assessing the optimal timing of the early functional load on the implant, since the early loading function contributes to an increase in osseointegration; however, in the complex context of bone remodeling, disintegration of the implant is also possible. There is a study in which 29 articles were selected out of 889 meeting the inclusion

criteria, based on the assessment of various loading protocols (immediate, immediate non-occlusive, early and normal loading) of dental implants for marginal bone loss [14]. The lowest level of marginal bone loss around implants was recorded with immediate loading ( $0.05 \pm 0.67$  mm), and the highest for implants with immediate non-occlusive loading ( $1.37 \pm 0.5$  mm), that is. immediate loading protocol is a reasonable alternative to traditional loading protocol. In our study, under the conditions of implantation in the lower jaw of the bone of pigs Art Implant (Ukraine), studies were carried out in two directions – with splinting components and without splinting, with an assessment of the state of the peri-implantation bone by a histological method and using the technology for

an assessment of the state of the peri-implantation bone by a histological method and using the technology for determining the stability coefficient. There are two types of osteogenesis - contact, on the surface of the implant, and remote, which takes place in the peri-implantation area, and behind the researchers' data, the remotely located bone delivers signals that induce contact osteogenesis [15]. In this regard, two indicators are usually used to assess osseointegration in histological studies - BIC and an assessment of bone tissue formation at a distance from the implant [16]. The latter indicator can also reflect the effect of the implantation on the surrounding bone. In our study, we have demonstrated that under conditions of direct loading and stabilization, there is a significant increase in bone tissue on the screw surface of the implant and in the area adjacent to the thread. Decreases osteocytic osteolysis, which prevents bone demineralization. In the absence of splinting with physically conditioned micro-mobility of implants during loading, resorptive processes in the bone tissue increase both on the surface of the implant and in the areas adjacent to the screw region, extensive areas of demineralization are found, which affects the contact of the implant with the bone. We can assume that our two-stage intraosseous dental implant provides low-trauma insertion of the implant with a self-tapping screw into the bone bed of all bone types, provides contact osteogenesis, seals the structures of the cancellous bone without disturbing the structure of trabeculae and precervical resorption in the wound. The morphological studies carried out were confirmed by the data using the dynamics of the stability coefficient of implants in the lower jaw according to Osstell Mentor in conditions of using a splinting component and without splinting in the clinical implantation protocol.

# CONCLUSIONS

Stabilization of implants using a splinting component increases the direct contact of the implant with the surrounding bone due to the activation of osteogenesis in comparison with implants without splinting: after 1 month. BIC increases 1.9 times, after 3 months. – 1.6 times.

Stagnation of the shingle component in the case of intrinsic intraoperative functional juvenile implantation accelerates the dynamics of osteointegration, so that high indicators of the efficiency of the implant stability can be achieved.

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### **Conflict of interest:**

The Authors declare no conflict of interest.

### **ORCID and contributionship:**

Anatoliy M. Potapchuk: 0000-0001-9857-1407 <sup>A,E,F</sup> Yevhen L. Onipko: 0000-0002-3086-4657 <sup>A,B,D</sup> Vasyl M. Almashi: 0000-0002-2943-4844 <sup>B,C,E</sup> Ninel V. Dedukh: 0000-0003-0307-2328 <sup>A,D</sup> Oleksandr Ye. Kostenko: 0000-0002-0549-1561 <sup>D</sup>

#### **Conflict of interest:**

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# **CORRESPONDING AUTHOR**

Anatoliy M. Potapchuk Uzhhorod national university 60a Station St., 88000 Uzhhorod, Ukraine tel: +380509399457 e-mail: anatoliy.potapchuk@uzhnu.edu

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