Romanian Biotechnological Letters *Vol. 23, No x, 2018*

ORIGINAL PAPER

Experimental model for the study of autogenous mandibular bone grafts integration

Received for publication, November, 3, 2017
Accepted, February, 5, 2018

ALEXANDRU POLL [1], COZETA ANCA MINCULESCU [2], VANDA ROXANA NIMIGEAN [3], DANIELA BĂDIȚĂ [4], ROSALIE ADINA BĂLĂCEANU [5], DIANA LORETA PĂUN [6], SIMONA ANDREEA MORARU [7], VICTOR NIMIGEAN [8]

1,8. Anatomy Department, Faculty of Dental Medicine, "Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania. 2. The Special Motricity and Medical Recovery/Rehabilitation Department, Faculty of Kinetotherapy, National University of Physical Education and Sport, Bucharest, Romania. 3,7. Oral Rehabilitation Department, Faculty of Dental Medicine, "Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania. 4. Physiology Department, Faculty of Dental Medicine, "Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania. 5. Physiology Department, Faculty of Veterinary Medicine, University of Agronomical Science and Veterinary Medicine, Bucharest, Romania. 6. Endocrinology Department, Faculty of Medicine, "Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania.

Corresponding author: *Vanda Roxana Nimigean, Associate Professor, Head of Oral Rehabilitation Department, Faculty of Dental Medicine, "Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania, No 17-23 Calea Plevnei Street.*
Phone: +40721561848. E-mail: vandanimigean@yahoo.com

Abstract

This research was conducted in order to create an animal experimental model which would allow the study of the integration of autogenous bone grafts used in implant dentistry. When the augmentation of the available bone is necessary, using mandibular autografts is the first choice, with specialised literature presenting numerous clinical situations which have benefitted from surgical treatment of bone quantity deficits.

The material used in the study was represented by seven dried skulls of adult domestic dogs, Canis familiaris, which died by natural causes. In order to define the experimental model, we utilized two morphometric work methods, morphometry through CT (Computed Tomography) imaging, and morphometry performed directly on dried canine skulls. The measurements were performed in order to determine the dimensions of the mandibular and maxillary buccal cortical bones and the width of the alveolar processes.

Both imaging and direct morphometry revealed that the thickness of the buccal alveolar compact and the width of the alveolar process were significantly larger in the posterior mandible than in the lateral maxillary region, on both the right and left sides. On the basis of the results, we established that the posterior region of the mandibular body should be the donor site, and the lateral region of the maxillary body should be the recipient site. The experimental model involved the creation of a maxillary bone defect and its augmentation with an autogenous mandibular bone graft.

The present research is useful in order to select the adequate experimental model for a particular scientific purpose.

Keywords: implant dentistry, alveolar bone bioactive capacity, biomaterial-driven bone regeneration

Background

This article proposes an animal experimental model, morphologically similar to the human maxilla and mandible, which enables the study of the integration of mandibular corticocancellous autografts applied in maxillary bone defects, and, later, the study of the epithelial and connective tissues integration to dental implants in grafted areas.

Introduction

Using mandibular autografts is the most frequent alternative for the reconstruction of resorbed alveolar bone, in implant dentistry (A. POLL [1]).

Recent scientific advances in the field of implant dentistry have enabled the development of new biotechnologies, especially bone regeneration techniques using biomaterials (A. POLL & al. [2]). Experimental animal models are useful in the evaluation of biologic behaviour of such materials and clinical success of the procedures, as *in vitro* models cannot replicate the complexity of the human anatomy.

It would be ideal that an experimental model for the study of the integration of bone autografts to have, embryologically and morphologically, similar biological traits in the donor and recipient areas (A. POLL [1], S. ISAKSSON [3], B. KLINGE & al. [4]).

Using animals with experimental and scientific purpose is quite an old scientific practice, which is still developing due to its advantages [5]). Also, the introduction and development of new biotechnologies and therapeutic devices necessitate the use of adequate animal models, which present bone healing and morphology similar to that of the human species (N. MARDAS & al. [6]). Most studies regarding the integration of bone autografts used in implant dentistry, had as an experimental model the calvarian bones (the parietal bones) of various animals, these experimental models being very easy to repeat (M.A.W. MERKX [7]).

The experimental specimens used in such studies must be of a large enough size to allow for standard x-rays investigations, without other nearby anatomical structures overlapping. Also, these must be easily manoeuvred, have a known background, and well documented growth and development of the maxillo-mandibular structures. Thus, we have considered that a medium-sized domestic dog, *Canis familiaris*, is morphologically, volumetrically, and embryologically similar to the human model, and could meet these criteria (A. POLL [1]).

Other research regarding the integration of bone grafts used in implant dentistry described different experimental models, such as the experimental model using a rabbit as specimen (B.K. ATIYA & al. [8]), the experimental model using a rat as specimen (P. KORN & al. [9], D.M. LEVY & al. [10]), the experimental model using a goat as a specimen (D. ZOU & al. [11]), the experimental model using a pig as a specimen (C. OGUNSALU & al. [12]), the experimental model using a cat as specimen (A.M. SILVA & al. [13]).

The comparison of embryological development and morphological similarities in animals and humans is the basis for experimental studies. Even though the results of experimental studies cannot be transferred directly to the human species, the development of animal models in research means obtaining results with a high degree of relevance for human species (A. POLL [1]), X. STRUILLOU & al. [14], D.I. SALAVASTRU & al. [15]).

Achieving such an experimental model represents a biotechnological procedure useful for the assessment of bioactive capacity of the alveolar bone of the maxilla and the mandible.

Material and methods

Following documentation through specialised literature, we established that the animal which would best meet the requirements of the adequate experimental models for our purpose, the study of the integration of autogenous bone grafts used in implant dentistry, is the domestic dog, *Canis familiaris*. Due to the anatomical configuration of the canine maxilla and mandible, as well as the direct accessibility, necessary for future surgical procedures, we decided that the maxillary lateral region and the posterior mandible were adequate anatomically, one as recipient and the other as donor sites. In the process of establishing the experimental model, the measurements focused on these areas.

Seven dried skulls of adult dogs, which died from natural causes, were used for the study. The research was organised and completed according to the legislation in force.

The measurements were completed in order to determine the average dimensions of the maxillary and mandibular buccal bone plates, the average width of the maxillary and mandibular alveolar processes, and also to determine the nearby presence of anatomical obstacles like the maxillary sinus or the mandibular canal.

In defining the experimental model, we established two methods of working, consisting of morphometric determinations through CT (Computed Tomography) imaging of dried canine skulls and morphometric determinations performed directly on dried canine skulls.

Morphometry through CT imaging of dried canine skulls

The measurements (in mm) of the thickness of the buccal bone plates and the width of the alveolar processes were completed through CT, in the lateral maxillary region and in the posterior mandible. Thus, three buccal-palatal sections were completed through the maxillary alveolar process, at the level of the first three premolars, on both the left and right sides, and in the mandible, three buccal-lingual sections were completed through the alveolar process at the level of the last premolar and at the level of the last two molars, on both the left and right sides. Overall, a total of 84 sections were completed, 42 in the maxilla and 42 in the mandible, 21 on the right side and 21 on the left side.

These sections were also analysed according to the presence of nearby anatomical elements (maxillary sinus and mandibular canal).

The measurements were completed using the 6-slice configuration computed tomography scanner (Siemens Somatom 6 CT scanner), with a series of technical parameters, such as 0.85 pitch, 1.25 mm thickness of sections, 6x1 mm section collimation, 120 kV and 45 mA. The width of the sections represents the real thickness of the reconstructed images.

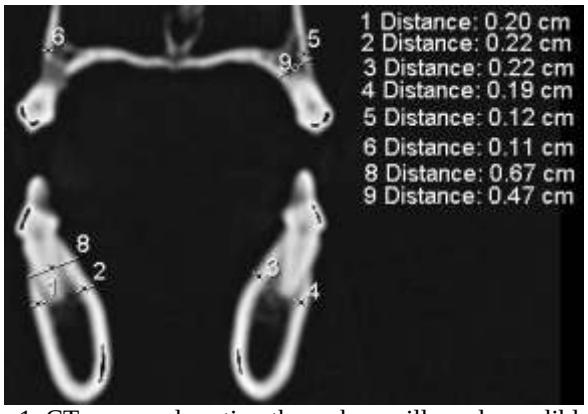

Figure 1. CT - coronal section through maxilla and mandible with the measurements of predetermined parameters.

Morphometry performed directly on dried canine skulls

The measurements and analyses were also completed on the seven dried canine skulls, which were sectioned, each maxilla having 6 bilateral buccal-palatal sections completed, at premolar level, obtaining, thus, a total of 84 sections, 42 on the right sides and 42 on the left sides. On the mandibles, 6 bilateral buccal-lingual sections were also completed, in the posterior region, at the level of the third and fourth premolars, the three molars and distal to the last molar, also obtaining a total of 84 sections, 42 on the right sides and 42 on the left sides.

These sections were completed using Ø 40 mm abrasive diamond blades on the edge, attached to a mandrel, using a dental micromotor, at regular speed.

The maxillary and mandibular sections were measured with a digital Workzone calliper (Globaltronics GmbH, Singapore).

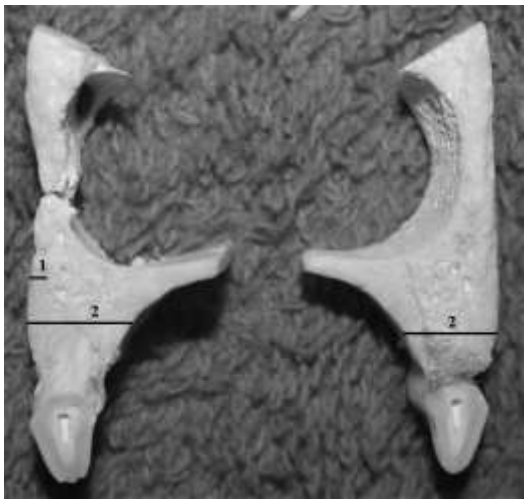

Figure 2. Buccal-palatal sections through the maxillary alveolar process in the premolar region: 1 - thickness of the buccal bone plate; 2 - width of the alveolar process.

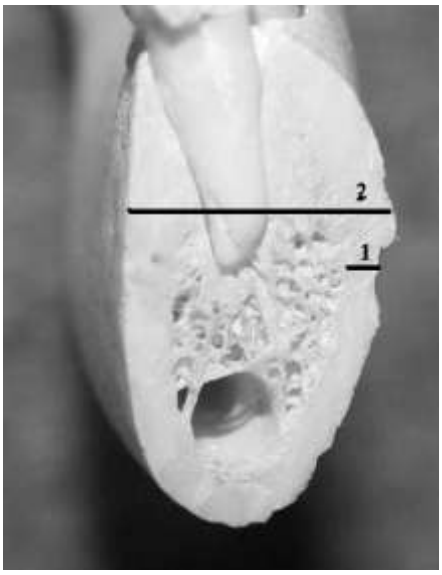

Figure 3. Buccal-lingual section through the mandible, in the premolar region: 1- thickness of the buccal bone plate; 2 - width of the alveolar process.

The data obtained were statistically analysed using Stata MP/13 software package. The average was calculated, along with standard deviation (SD), standard error (SE), 95% confidence interval on the average (95% CI), median, minimum and maximum values. The data were compared with the Student's t-test, which was used bilaterally, and the statistical significance threshold was established for $p \leq 0.05$.

Results

The results obtained regarding measurements through CT imaging are presented in tables from 1 to 4, where N represents the number of sections.

Table 1. Thickness of the right buccal bone plate (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	21	0.70	0.04	0.20	0.61-0.79	0.40	0.68	1.10
Posterior mandible	21	1.73	0.04	0.19	1.64-1.82	1.50	1.70	2.20
Student's t-test, $p < 0.0001$								

Table 2. Thickness of the left buccal bone plate (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	21	0.70	0.04	0.19	0.61-0.79	0.38	0.70	1.00
Posterior mandible	21	1.69	0.04	0.19	1.60-1.78	1.50	1.70	2.10
Student's t-test, $p < 0.0001$								

Table 3. Width of the alveolar processes on the right side (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	21	6.01	0.17	0.76	5.66-6.36	4.70	6.00	7.90
Posterior mandible	21	7.77	0.22	1.02	7.30-8.23	5.90	7.60	10
Student's t-test, $p < 0.0001$								

Table 4. Width of the alveolar processes on the left side (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	21	5.92	0.16	0.75	5.58-6.27	4.70	6.00	7.50
Posterior mandible	21	7.79	0.20	0.91	7.38-8.20	5.90	7.60	9.50
Student's t-test, $p < 0.0001$								

The results obtained regarding direct morphometry on dried canine skulls are presented in tables from 5 to 8, where N represents the number of sections.

Table 5. Thickness of the right buccal bone plate (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	42	0.77	0.03	0.19	0.71-0.82	0.39	0.77	1.1
Posterior mandible	42	2.01	0.05	0.33	1.91-2.11	1.6	1.9	3
Student's t-test, $p < 0.0001$								

Table 6. Thickness of the left buccal bone plate (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	42	0.76	0.03	0.20	0.69-0.82	0.35	0.765	1.15
Posterior mandible	42	1.98	0.06	0.37	1.86-2.10	1.60	1.85	3.10
Student's t-test, $p < 0.0001$								

Table 7. Width of the alveolar processes on the right side (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	42	5.77	0.10	0.65	5.57-5.98	4.8	5.775	7.25
Posterior mandible	42	9.52	0.29	1.88	8.93-10.11	7	8.80	14
Student's t-test, $p < 0.0001$								

Table 8. Width of the alveolar processes on the left side (mm).

Bone region	N	Mean	SD	SE	95% CI	Minimum	Median	Maximum
Lateral maxilla	42	5.72	0.11	0.74	5.49-5.95	4.5	5.525	8
Posterior mandible	42	9.49	0.29	1.89	8.9-10.07	6.7	9	13.5
Student's t-test, $p < 0.0001$								

Discussion

No significant statistical differences were found both through direct and imaging morphometrics, regarding the thickness of the buccal bone plate in the target studied areas (anterior, medial or posterior) of the same specimen. The minimum thickness of the maxillary buccal bone plate was 0.35 mm, and the maximum thickness was 1.15 mm.

The width of the alveolar process expanded towards the posterior extremity of the target area. The minimum determined width of the maxillary alveolar process, in the predetermined area, was 4.50 mm, the maximum determined width was 8.00 mm, and the maxillary sinus (anatomical obstacle for implants placement) was located at a certain distance to the alveolar process. Similar results were also found in other morphometric studies (D.I. SALAVASTRU & al. [15], V. NIMIGEAN & al. [16]).

In the posterior mandible, there were significant dimensional differences regarding the thickness of the buccal bone plate, it becoming thicker towards the posterior, so that, distal to the penultimate molar, it had its greatest thickness, 3.10 mm, with the smallest thickness in the anterior region of the predetermined target area, 1.50 mm. The minimum width of the mandibular alveolar process, in the predetermined area, was 5.90 mm, and the maximum width was 14.00 mm. The mandibular canal was situated close to the buccal bone plate, and at 0.0-3.5 mm to the dental apices, distance which grew progressively from the molar region to the premolar region, on a similar trajectory to that found in humans. The fundamental difference between the two species regarding the trajectory of the mandibular canal in the posterior region of the mandibular body is that in the canine species, the mandibular canal is located near the buccal bone plate, whereas in humans, it is situated near the lingual bone plate, which could mean that this canal is an anatomic obstacle, deserving of special attention during bone graft harvesting in this region.

From a statistical perspective, both through imaging, and through direct morphometry, the thickness of the buccal bone plate was significantly greater in the posterior mandible compared to the lateral maxillary area, on both the right and left sides ($p < 0.0001$).

Also, from a statistical perspective, both through imaging and through direct morphometry, the width of the alveolar process was significantly greater in the posterior mandible compared to the lateral maxillary area, on both the right and left sides ($p < 0.0001$).

Correlating the results obtained, we established the posterior mandibular body as being the donor region, and the lateral region of the maxillary body, corresponding to the alveolar portion of the second or third maxillary premolars, as being the receiving region. Thus, we completed a canine experimental model to study the use of a mandibular autogenous graft for the augmentation of a maxillary bone defect.

What is more, other authors used in their studies canine experimental models in order to study bone regeneration in the case of autografts applied at the maxillary level (N. POUREBRAHIM & al. [17], A. ORYAN & al. [18]).

In the last decade many experimental studies on animal models were developed in order to increase the long-term performances of dental implants. These studies have shown that larger segments of bone autografts ensure a better conservation of bone volume and maintain bone height eight weeks after being applied (C. OGUNSALU & al. [19], C. OGUNSALU & al. [20], K. KON & al. [21]).

Unfortunately, there are no precise regulations between scientists regarding the basis of experimental research. The only criterion for the choice of a certain experimental research topic is its relevance, which cannot, however be judged in the short-term. A concept to systematically assess the relevance of the *in vitro* tests must be developed in order to increase quality and to finally achieve an evidence-based biomedical research (F.P. GRUBER and T. HARTUNG [22]).

Animal experimental models can be limited by scientific constraints regarding applicability to the human species, and due to the increase of regulation restrictions (A. KNIGHT [23], M. LEIST & al. [24], W. LILIENBLUM & al. [25]).

It may be recommendable that requests for animal experimentation take into consideration the differences between species, and include detailed arguments regarding the necessity of their completion. Also, prior to approval, the authorities should have the right to request the modification of the experimental protocol, for example, regarding the study design or the number of specimens (M. FOSTER and M. BRADDOCK [26], S.C. SARTORETTO & al. [27]).

Conclusions

There are studies that gravely undermined the hypothesis which proved that animal experimental models are irreplaceable for the progress of clinical research. However, overestimation of the clinical benefits of experimental research could be an error.

The canine experimental model can be considered appropriate for the assessment of the integration of biomaterials used for bone regeneration in implant dentistry.

This study should help researchers in the field select the most adequate experimental model for each scientific purpose.

Conflict of interests

The authors declare that they have no conflict of interests.

Acknowledgments

The research for this paper was conducted within Alexandru Poll's PhD Thesis, "Fundamental studies regarding biocompatibility of grafts used to augment maxillary and mandibular bone volume".

The stipulations in the European Council's Directive 86/609/EEC and Directive 2010/63/EU, regarding protection of animals used in experimental or otherwise endeavours were followed in this study. Also, the study was endorsed by the Ethics Committee of the Faculty of Veterinary Medicine in Bucharest and the study was performed in accordance with local laws and regulations.

Author contribution

Author #1 (Alexandru Poll), autor #4 (Daniela Bădiță), autor #6 (Diana Loreta Păun) and autor #7 (Simona Andreea Moraru) have equal contributions to this paper and thus are main authors.

References

1. A. POLL, *Studii fundamentale privind biocompatibilitatea grefelor utilizate pentru creşterea volumului osos maxilo-mandibular (Fundamental studies regarding biocompatibility of grafts used to augment maxillary and mandibular bone volume)*. PhD Thesis. "Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania, 52-87 (2015).
2. A. POLL, V.R. NIMIGEAN, D. BADITA, R.A. BALACEANU, S.C. CISMAS, P. PERLEA, S.A. MORARU, V. NIMIGEAN, *In vivo experimental model for the evaluation of dental implant integration*. Romanian Biotehnological Letters, 2017, DOI 10.26327/RBL2017.88
3. S. ISAKSSON, *Aspects of bone healing and bone substitute incorporation: an experimental study in rabbit skull bone defects*. Swed. Dent. J., 84 (Suppl.), 3-46 (1992).
4. B. KLINGE, P. ALBERIUS, S. ISAKSSON, J. JÖHNSON, *Osseous response to implanted natural bone mineral and synthetic hydroxylapatite ceramic in the repair of experimental skull defects*. J. Oral Maxillofac. Surg., 50(3), 241-249 (1992).
5. Http://ec.europa.eu/environment/chemicals/lab_animals/reports_en.htm
   Seventh Report from the Commission to the Council and the European Parliament on the Statistics on

the number of animals used for experimental and other scientific purposes in the member states of the European Union COM(2013)859/final. Retrieved 3th march 2014.

6. N. MARDAS, X. DEREKA, N. DONOS, M. DARD, *Experimental model for bone regeneration in oral and cranio-maxillo-facial surgery*. J. Invest. Surg., 27(1), 32-49 (2014).
7. M.A.W. MERKX, *Autogenous bone and bovine bone mineral as grafting materials in maxillofacial surgery*. Benda BV, Nijmegen, 11-23 (2000).
8. B.K. ATIYA, P. SHANMUHASUNTHARAM, S. HUAT, S. ABDULRAZZAK, H. OON, *Liquid nitrogen-treated autogenous dentin as bone substitute: an experimental study in a rabbit model*. Int. J. Oral Maxillofac. Implants, 29(2), 165-170 (2014).
9. P. KORN, M.C. SCHULZ, U. RANGE, G. LAUER, W. PRADEL, *Efficacy of tissue engineered bone grafts containing mesenchymal stromal cells for cleft alveolar osteoplasty in a rat model*. J. Craniomaxillofac. Surg., 42(7), 1277-1285 (2014).
10. D.M. LEVY, C. SAIFI, J.L. PERRI, R. ZHANG, T.R. GARDNER, C.S. AHMAD, *Rotator cuff repair augmentation with local autogenous bone marrow via humeral cannulation in a rat model*. J. Shoulder Elbow Surg., 22(9), 1256-1264 (2013).
11. D. ZOU, L. GUO, J. LU, X. ZHANG, J. WEI, C. LIU, Z. ZHANG, X. JIANG, *Engineering of bone using porous calcium phosphate cement and bone marrow stromal cells for maxillary sinus augmentation with simultaneous implant placement in goats*. J. Tissue Eng. Regen. Med., 18(13-14), 1464-14678 (2012).
12. C. OGUNSALU, C. EZEOKOLI, A. ARCHIBALD, J. WATKINS, C. STOIAN, H. DAISLEY, C. LEGALL, S. LORDE, K. JACKSON, D. JAGGEERNAUTH, A. NELSON, N. MUNGAL, *Comparative study of osteoblastic activity of same implants (Endopore) in the immediate extraction site utilizing single photon emission computerized tomography: peri-implant autogeneous bone grafting with GTR versus no peri-implant bone grafting - experimental study in pig model*. West Indian Med. J., 60(3), 336-339 (2011).
13. A.M. SILVA, W.M. SOUZA, N.T. SOUZA, M.B. KOIVISTO, A. BARNABÉ PDE, S. POLÓ TDA, *Filling of extraction sockets with autogenous bone in cats*. Acta Cir. Bras., 27(1), 82-87 (2012).
14. X. STRUILLOU, H. BOUTIGNY, A. SOUEIDAN, P. LAYROLLE, *Experimental animal models in periodontology: a review*. Open Dent. J., 29(4), 37-37 (2010).
15. D.I. SALAVASTRU, V. NIMIGEAN, V.R. NIMIGEAN, A. POLL, S. MORARU, *Experimental model for the dental implant integration study*. Rom. J. Stomatol., 59(1), 24-27 (2013).
16. V. NIMIGEAN, V.R. NIMIGEAN, N. MARU, D.I. SALAVASTRU, D. BADITA, M.J. TUCULINA, *The maxillary sinus floor in the oral implantology*. Rom. J. Morphol. Embryol., 49(4), 485-489 (2008).
17. N. POUREBRAHIM, B. HASHEMIBENI, S. SHAHNASERI, N. TORABINIA, B. MOUSAVI, S. ADIBI, F. HEIDARI, M.J. ALAVI, *A comparison of tissue-engineered bone from adipose-derived stem cell with autogenous bone repair in maxillary alveolar cleft model in dogs*. Int. J. Oral Maxillofac. Surg., 42(5), 562-568 (2013).
18. A. ORYAN, S. ALIDADI, A. MOSHIRI, N. MAFFULLI, *Bone regenerative medicine: classic options, novel strategies, and future directions*. J. Orthop. Surg. Res., 9(1), 18 (2014).
19. C. OGUNSALU, A. ARCHIBALD, C. EZEOKOLI, *Emerging applications of an experimental single photon emission computed tomography: an analysis of 16 areas of interest in the pig's model*. West Indian Med. J., 61(9), 916-920 (2012).
20. C. OGUNSALU, A. ARCHIBALD, J. WATKINS, C. STOIAN, C. EZEOKOLI, H. DAISLEY, C. LEGALL, S. LORDE, K. JACKSON, D. JAGGERNAUTH, A. NELSON, N. MUNGAL, *Comparative study of the osteoblastic activity of two implant systems (Endopore versus Entegra) utilizing single photon emission computed tomography (SPECT): experimental study in pigs model*. West Indian Med. J., 62(2), 145-148 (2013).
21. K. KON, M. SHIOTA, M. OZEKI, S. KASUGAI, *The effect of graft bone particle size on bone augmentation in a rabbit cranial vertical augmentation model: a microcomputed tomography study*. Int. J. Oral Maxillofac. Implants, 29(2), 402-406 (2014).
22. F.P. GRUBER, T. HARTUNG, *Alternatives to animal experimentation in basic research*. ALTEX, 21(Suppl 1), 3-31 (2004).
23. A. KNIGHT, *Non-animal methodologies within biomedical research and toxicity testing*. ALTEX, 25(3), 213-231 (2008).
24. M. LEIST, S. KADEREIT, S. SCHILDKNECHT, *Food for thought... on the real success of 3R approches*. ALTEX, 25(1), 17-32 (2008).
25. W. LILIENBLUM, W. DEKANT, H. FOTH, T. GEBEL, J.G. HENGSTLER, R. KAHL, P.J. KRAMER, H. SCHWEINFURTH, K.M. WOLLIN, *Alternative methods to safety studies in experimental animals: role in the risk assessment of chemicals under the new European Chemicals*

Legislation (REACH). Arch. Toxicol., 82(4): 211-236 (2008).
26. M. FOSTER, M. BRADDOCK, *To guide the process. Part II: Animal models*. Screening, 3, 20-21 (2004).
27. S.C. SARTORETTO, M.J. UZEDA, F.B. MIGUEL, J.R. NASCIMENTO, F. ASCOLI, M.D. CALASANS-MAIA, *Sheep as an experimental model for biomaterial implant evaluation*. Acta Ortop. Bras., 24(5), 262-266 (2016).