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Efficacy of Using Pure Beta-Tricalcium Phosphate on Osseointegration of Dental Implants (An Experimental and Radiographic Study)

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Abstract: ***Background*****:** Pure beta-tricalcium phosphate has been developed in atrial to achieve higher osseointegration of dental implants.

Aim of the study**:** This study was to evaluate the effect of pure beta-tricalcium phosphate on osseointegratin around dental implants.

Materials and Methods *:* 24 dual dental implants were implanted in tweleve male dogs aged 1-2 years and weighting 18-20 kg. Implants insertion were performed on the right and left iliac crest of all dogs, then each group of two main groups

Group I (control group: 12 implants were left without bone graft.

Group II (study group): 12 implants with pure beta-tricalcium phosphate (Cerasorb). Implants were evaluated histological and radiographic analysis were tested after 4 and 8 weeks post implantation. The 6 dogs to each group and 3 dogs every periods.

Results*:* The Pure beta-tricalcium phosphate (Cerasorb) exhibited the optimal predictability for osseiointegration and radiographic values than control groups. Histomorphometrically, the mean percentage area of the formed bone showed significant amount of bone formation around the implants of the study group when compared with control group. All data collected were analyzed using SPSS version 26.0 software. Statistical significance was at a *P* value 5%.

Conclusion*:* It can be concluded that Pure beta-tricalcium phosphate (Cerasorb) with implants improved early osseiointegration with superior biomechanical property.

Keywords: *Dental implants, pure beta-tricalcium phosphate, Osseointegration, bone density.*

Introduction

Dental implants are alloplastic materials embedded in the maxilla and/or mandible for the management of tooth loss and to aid replacement of lost orofacial structures as a result of trauma, neoplasia and congenital defects. Implant supported dental restorations have become a major option in the treatment of edentulous alveolar ridge for the past several decades. To achieve optimum treatment outcome with dental implants, sufficient bone should be available to support and stabilize them .(1)

Osseointegration has been defined as a direct structural and Functional connection between living bone and the surface of the implant. Osseointegration represents a lifelong process of bone formation and adaptation to function and repair consequently. Osseointegration is a prerequisite for long term clinical function.(2)

One of the most promising groups of synthetic bone substitutes is β-Tricalcium phosphate (β-TCP) is a synthetic porous ceramic which has shown osseoconduction and very favorable substitution in standardized bone defects. It has also good tissue tolerance with no immunological or toxic reactions, good biocompatibility, characteristics of absorption and is effectively replaced by natural bone in an appropriated period of time.(3)

The use of TCP particles with increased porosity has been promoted in order to increase the biodegradability. These particles exhibit a material structure with micromiso and macropores which is designed to enhance the degradation process. This structure allows for a reduced bulk density.(4)

The ß-Tri-calcium phosphate (Cerasorb) might promote faster and better healing. These observations were further supported by the difference between the measured values of the bone implant interface gaps dimensions.(5)

Also beta tricalcium phosphate in humans and concluded that (Cerasorb) proved to be an effective bone replacing material with osteoconductivity since it was capable of gradual disintegration and thereby providing space for the regenerating bone.(6)

Apart from being osteoconductive**,** there is strong experimental evidence that calcium phosphates also have osteoinductive properties. Although the underlying mechanism remains largely unknown, it has been shown that these alloplastic materials can stimulate osteogenic differentiation of stem cells in vitro and bone induction in vivo.(7)

The improved stability throughout the graft material seems to further improve the quality of the bone that will be regenerated due to reduced micro motion of the material, which may lead to mesenchymal differentiation to fibroblasts instead of osteoblasts. It is known that micro movements between bone and any implanted grafted material prevent bone formation, resulting in the development of fibrous tissue.(8)

The success of dental implants should be defined in terms of the underlying mechanisms affecting the formation, adaptation and repair of bone**.** Various grafting procedures can be used with different bone substitute.(9)

From the fore mentioned data it was of interest to the effect of Pure beta-tricalcium phosphate (cerasorb) on osseointegration.

Aim of the study: The present study was to evaluate the efficacy of pure beta-tricalcium phosphate on osseointegration around dental implants.

Materials and Methods

Animals:

Twenty four dental implants in tweleve mongrel male dogs aged 1-2 years and weighting 18-20 kg. All experimental procedures were performed according to a protocol approved by the ethical committee of faculty of Dentistry, Suez Canal University, Egypt.

Grouping of implants:

The implants were randomly and equally divided into two groups:

Group I: (Control group):

The implants were inserted with out pure beta- tricalcium phosphate (Cerasorb).

Group II: (Study group):

The implants were inserted with pure beta-tricalium phosohate at osteotomy site.

-Presurgical phase

Dogs was premeditated with an intramuscular (IM) injection of Chlorpromazine hydrochloride at a dose of 1mg/kg.[1]

Surgical procedure:

-General anesthesia was conducted by intravenous (IV) injection of thiopental sodium 2.5% solution until the main reflexes was abolished.[2]

-Using scalpel 24, an incision around 3 cm and dissection for the subcut for exposing the iliac crest.

-Implant site was prepared in step by step procedure using pilolt drill to 3.7 of diameter and 10 in length, to ensure an efficient and a traumatic widening of implant site.

Drilling speed was 1200 rpm in a standardized fashion according to manufacturer instructions.

Osteotomy sites were first drilled with a pilot drill to a depth of 10 mm .This was followed by the final drill for all groups. The sites of implantation were irrigated profusely with normal saline.

After preparation of the osteotomy sites the implants were placed into their final positions using the manual ratchet. Then, all incisions were closed in layers using suture 0 silk.[3]

Finally, the skin was disinfected again with disinfecting agent (Betadine).[4]

Post-operative care:

All dogss were injected with analgesic (voltarine) 75mg/kg once times a day.[5] As prophylaxis, antibiotic (Cefotaxime) 150 mg/kg was injected in all dogs for five days after the operative.[6]

Evaluation:

For evaluation of bone healing at different periods of healing (4 and 8 weeks postoperatively), dogs were sacrificed by overdose of thiopental sodium. Then the block samples of iliac crest with implants were sectioned by a saw altogether with removal of unnecessary remnants of bone and soft tissue, and were prepared for the subsequent examinations.

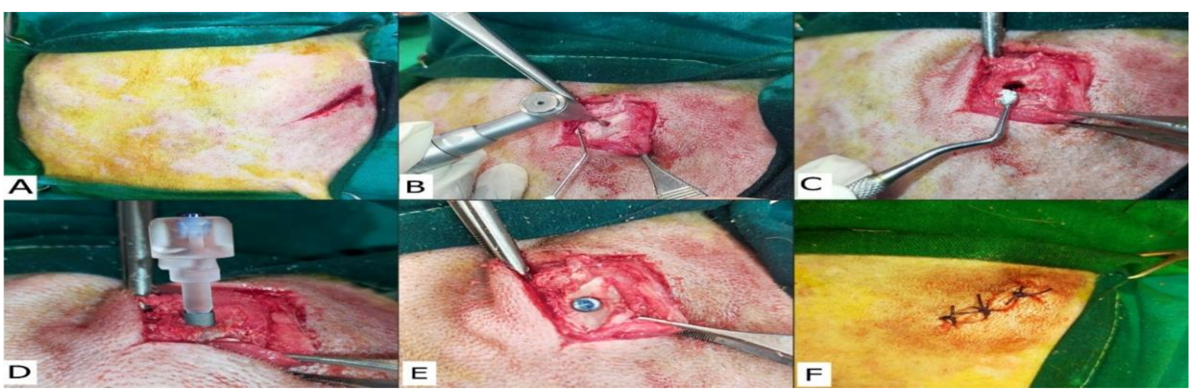

Fig. (1): Photographs showing the surgical Procedure **A:** Prepared area and Incision **B:** Reflection of the periosteum and drilling in the iliac crest **C:** Filling the site by the cerasorb bone graft **D:** Direction of the implant in iliac crest **E:** Cover screw of implant site **F:** Wound closure.

[1] Neurazine, Misr Co.pham, S.A.A.
[2] Nesdonal, Epico, Co.Egypt.
[3] Truglyde, India.
[4] Betadine,NileCo.Egypt.
[5] Novartis pharma.cairo.Egypt.
[6] Epico, Co.Egypt.

I. Histological analysis:

The specimens of the iliac crest with implants were dissected and immediately placed in 10% buffered formalin solution for 48 hours, decalcified in 50% formic acid and 20% sodium citrate solution for 4 and 8 weeks then the implants were removed carefully. Decalcified samples were washed for 24 h in 0.1M phosphate buffer (pH 7.2), dehydrated through graded ethanols, cleared with xylene, embedded in paraffin and serially-sectioned at 5 μm thickness. Some sections were stained with hematoxylin and eosin for observation by light microscopy and histomorphometric analysis.[10]

Histomorphometric analysis:

Slides were photographed using Olympus® digital camera installed on Olympus® microscope with 1/2 X photo adaptor; using 100 X objective. Then the resultd images were analyzed on Intel® Core I3® based computer using Video Test Morphology® software (Russia) with a specific built-in routine and area bone formation measurement.

II. Radiographic analysis.

The stored images of each dog was interpreted by one examiner at two different times so as to decrease intra and inter observer errors mean of the two trials. Each dog was radiographically assisted by (**Rayence radiograph)** at 4 and after 8 weeks post-operative by the control x ray system and fixed value (40 kv and 5 Mas) and image analysis of bone density by using idrisi software was recorded.

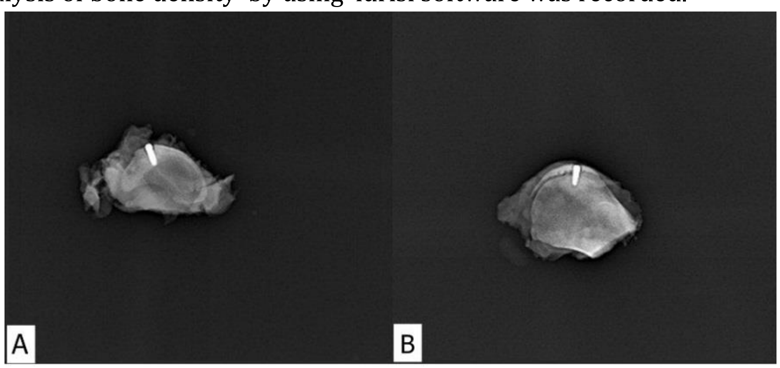

Fig. (2): Photographs showing the iliac crest bone radiograph **at 4 weeks. A**: Showing x-ray implant at control group. **B**: Showing x-ray implant at study group.

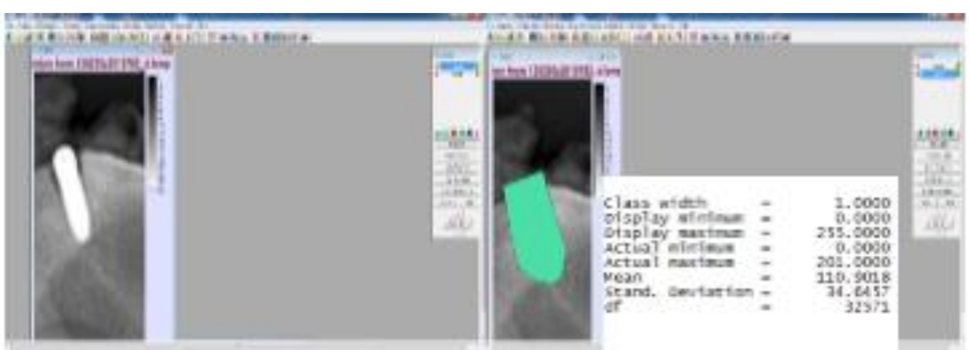

Fig. (3): Photograph showing control group evaluation of bone density around dental implant after 4 weeks.

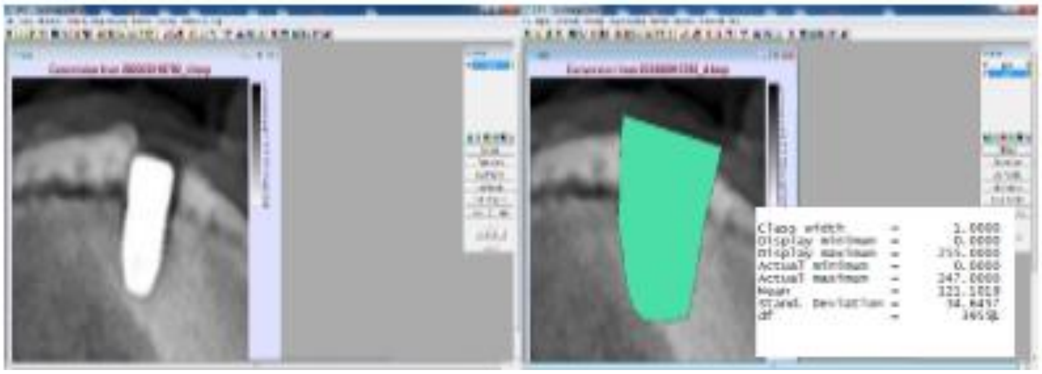

Fig. (4): Photograph showing study group evaluation of bone density around implant after 4 weeks.

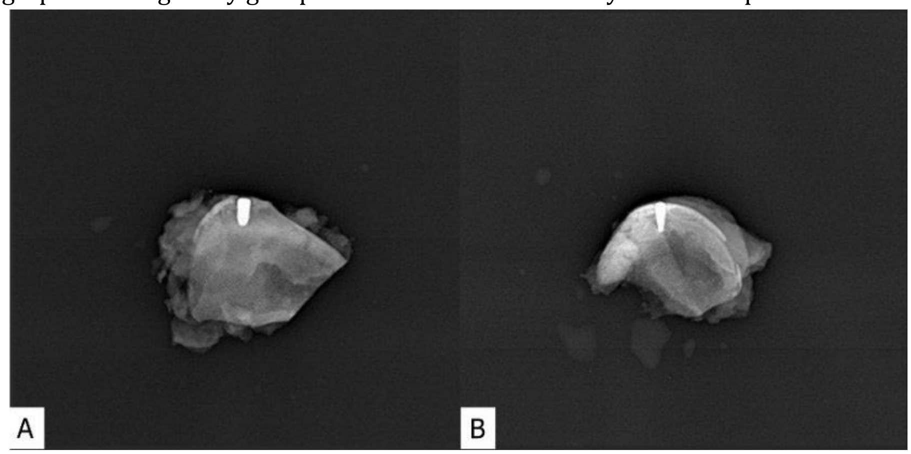

Fig. (5): Photographs showing the iliac crest bone radiograph **at 8 weeks**. **A**: Showing x-ray implant at control group. **B**: Showing x-ray implant at study group.

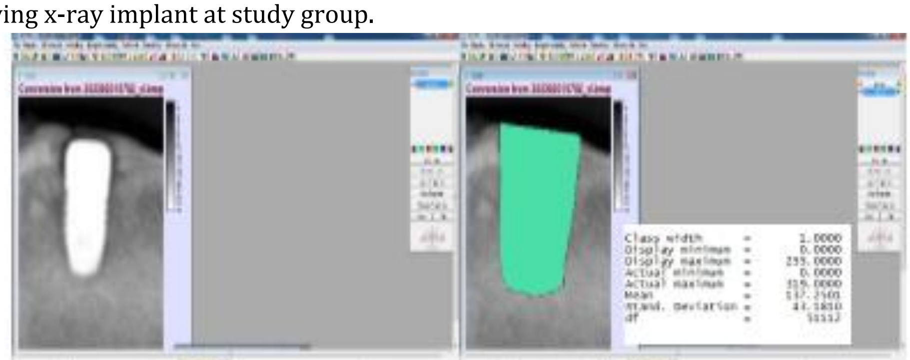

Fig. (6): Photograph showing control group evaluation of bone density around implant after 8 weeks.

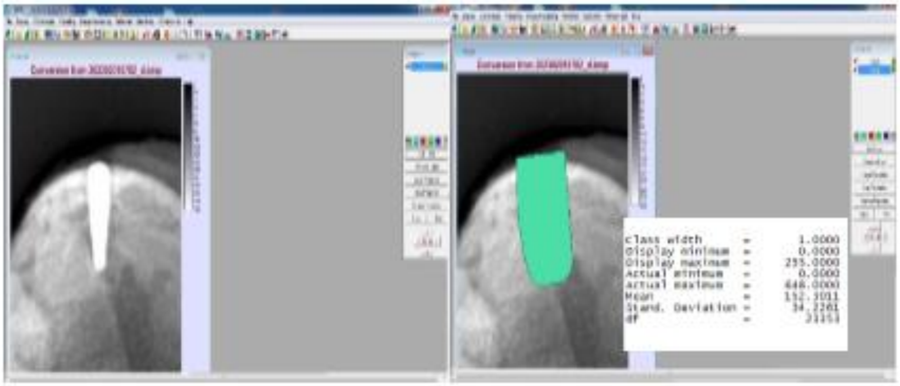

Fig. (7): Photograph showing study group evaluation of bone density around dental implant after 8 weeks.

<u>Statistical analysis:</u>

The data were collected and statistical analysis was done using the Statistical Package for the Social Software (SPSS, version 21 In., Chicago, I11, USA), Quantitative data were described using range (minimum and maximum), mean, and standard deviation. Significance of the obtained results was judged at Anova for histological analysis and Anova ant T test for removal torque for the 5% level.

Results

Histological analysis:

1. **Haematoxylin and eosin stain (H&E):**

At 4 weeks post implantation

Microscopic examination of bone at the site of the implant in the **control group** (**Fig. 8**) revealed thin less connecting bony trabeculae at both edges and at the base of the implant site with defective mineralization and wide marrow spaces meanwhile, **Study group** (implants with beta tricalcium phosphate bone graft (cerasorb). (**Fig. 9**) showed relatively thickened trabeculae with narrow marrow spaces and obvious mineralization.

At 8 weeks post implantation

Control group (**Fig. 10**) showed fibrous tissue filling the spaces at the implant site, more bone was formed at the base of the implant site with presence of active osteoblasts over the surface of the bone. **Study group I** (**Fig. 11**) showed more bone formation and less fibroplasia at the implant site.

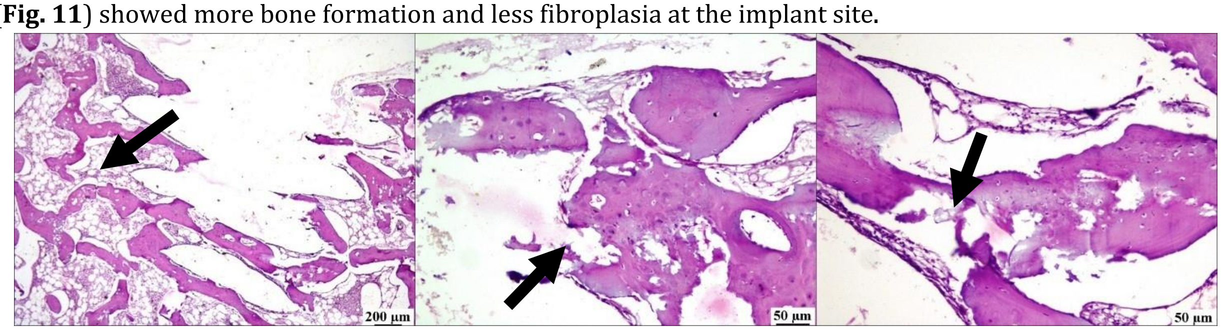

Fig. (8): Photomicrograph of **control group**, at **4 weeks A:** Showing thin less connecting trabeculae (arrow) at the implant site. **B:** Higher magnification showing bone necrosis at the edges of the implant with incomplete mineralization of the formed bone (arrow). **C:** Higher magnification showing thin boney trabeculae (arrow) at the base of the implant site. **(Haematoxylin and eosin stain)**

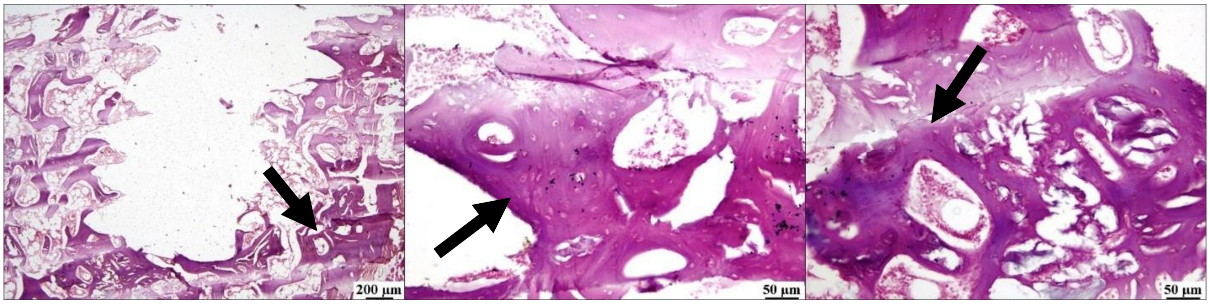

Fig. (9): Photomicrograph of **study group** , at **4 weeks A:** Showing relatively thick more connecting trabeculae (arrow) at the implant site. **B:** Higher magnification showing thick and connected bone trabeculae (arrow) at the edges of the implant site. **C:** Higher magnification showing thick and well-calcified bone (arrow) at the base of the implant site. **(Haematoxylin and eosin stain)**

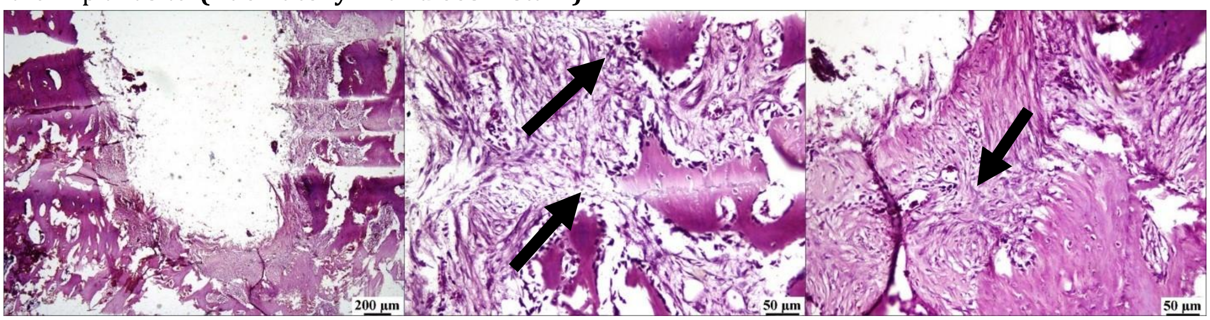

Fig. (10): Photomicrograph of **control group**, at **8 weeks A:** Showing fibrous union at the edges of the implant site. **B:** Higher magnification showing fibrosis with presence of active osteoblasts at the edges of the bone (arrow). **C:** Higher magnification showing fibrosis (arrow) with new bone formation. **(Haematoxylin and eosin stain)**

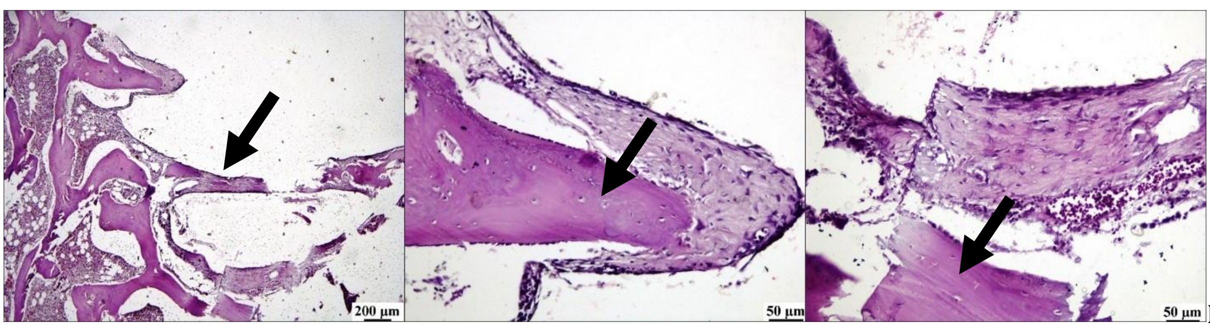

Fig. (11): Photomicrograph of **study group**, at **8 weeks A:** Showing mild fibroplasia (arrow) at the implant site. **B:** Higher magnification showing new bone formation (arrow) at the implant sides. **C:** Higher magnification showing newly formed bone (arrow) at the base of the implant site. **(Haematoxylin and eosin stain)**

The results in **table 1**, showed that there was statistically significant difference between the two groups after 8 weeks for % area and there is significant between control group and **study group** using one-way ANOVAs at P<0.05. The high mean values were recorded in **study group** bone followed by **control group** bone while **control group** was the lowest one after **4** and **8 weeks** (**Fig,12**)

a. Changes within each group

As regards changes within each group ,there was a statistically significant change in average area of bone formation with increasing time. The changes were 15. 80%, and 15.3% in GI and control group respectively.

Table 1, %Area							
	4 weeks		8 weeks			T test	P value
	Mean	Std.Deviation	Mean	Std. Deviation	% change		
Control group	3.932[b]	0.114	4.701[c]	0.100	15.3	6.37	0.024**
Study group	6.486[a]	0.103	7.511[b]	0.102	15.80	19.07	0.003**
F test	618.22		544.51				
P value	<0.001**		<0.001**				
	**, means significant difference at P<0.001						

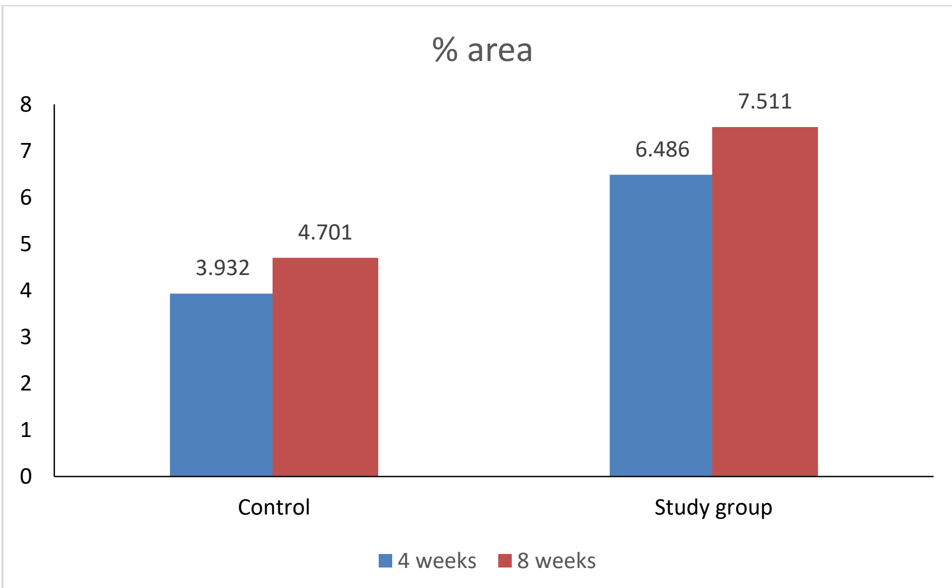

Fig. (12): Chart showing comparison of average area of bone formation around dental implant between both groups at 4 and 8 weeks.

<u>**Radiograph result:**</u>

The results showed descriptive statistics including the mean, standard division for bone osseointegration around implant using idrissi program on digital periapical radiograph.

Comparison between groups at 4 week and after 8 weeks:

The results in **table 2**, showed significant difference between **study group** and **control group** at **4 weeks** and after **8 weeks**. Generally, the high mean values were recorded in **study group** followed by **control group** . (**Fig.13**)

Table 2, Comparison between groups at 4 and after 8 weeks				
	Control group	study group	F	P values
At 4 weeks	127.73±10.70b	165.71±11.09a	22.50	<0.001**
At 8 weeks	141.46±12.55b	173.89±12.74a	18.19	0.006**
****, and different letters means significant difference at P<0.01**				

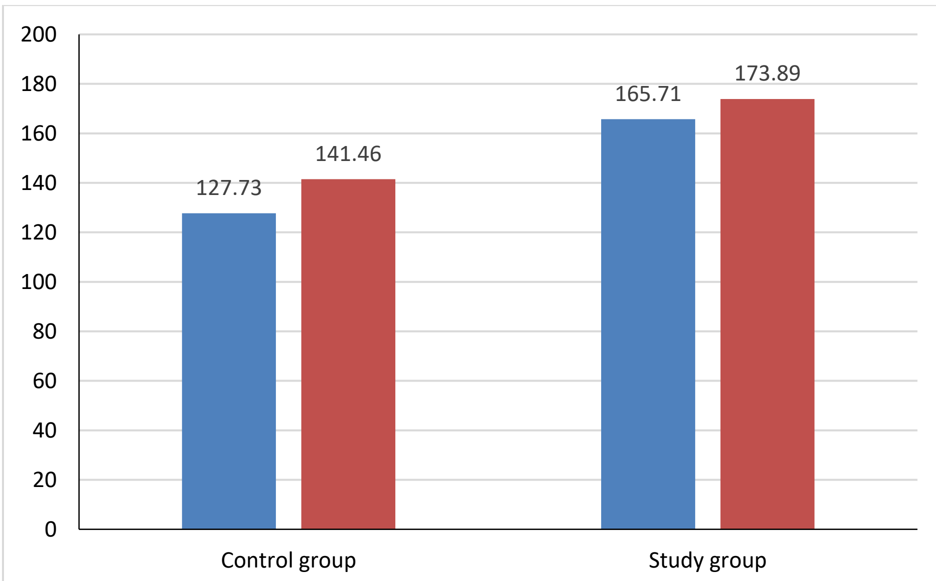

Fig. (13): Chart showing comparison between both groups of density around implant at 4 and 8 weeks.

Discussion:

Dental implants became a popular and reliable method for the restoration of missing tooth/teeth. Resorption of the edentulous alveolar ridge is a comparatively constant trait after tooth extraction. It is a rapid process during the first 6 months and then continue progressively, chronic and irreversible in a cumulative pattern.(11)
The specific goal of the current study was to study the assesment efficacy of using pure beta-tricalcium phosphate on osseointegration of dental implants through experimental studies. In the present study, the histologically stained sections (H & E) clearly showed the difference in osseointegration among both experimental groups.

Regarding to control group, the histological results in the present study concerning bone implant contact following the shape of the screw at four weeks period, showed newly formed woven with granulation tissue and dense inflammatory infiltrates surrounded by thin, newly formed woven bone trabeculae. Our results were agreed with ***El-bahrawy et al.,***(12) study who showed at there experimental work with rabbits that the bone contained loose granulation tissue with inflammatory cells infiltration at four weeks post-surgery. This experimental work was in agreement with the current study, which found that, at four weeks post-operatively there was newly bone trabeculae extending from the defect boundaries with abundant inflamed connective tissue, while eight weeks post-surgery revealed relatively thick bone trabeculae began to coalesce with each other. These events are also comparable to the findings of ***AlNashar et al.,***(13) in his experimental work at 12 weeks in rabbits.

In the present study, the absence of an excessive inflammatory response by the host tissue toward the graft particles at the two groups confirmed the biocompatibility of them which was in accordance with ***Cai et al.,***(14) at their experimental work at (2,4,and 8) weeks in rabbits.

In Cerasorb group (β-TCP) revealed more significant enhancement of osseointegration than control group at the same periods ($P<0.001$), Which declared variant degree of bone formation with a statistically difference at 4 and 8 weeks post operatively. After 4 weeks, the histological results demonstrated moderate thick bone trabeculae extended around implants, while at, at 8 weeks post- implantation, well-formed, more compact bone were established with much more amount of bone than control group.
It is worth mentioning that ß-TCP particles were showed relatively thickened trabeculae with narrow marrow spaces and obvious mineralization by histological and bone density by radiographic studies. While at eight weeks ß-TCP showed more bone formation and less fibroplasia at the implant site seen histologically.
The observations were not in accordance with the results of ***Horch et al.,***(15) who found that complete replacement of ß-TCP ceramic by autogenic bone could be proven radiographically after approximately twelve month.

Regarding to β-TCP group, our histological results found a new formed thin woven bone trabeculae at 4 weeks post implantation, while at 8 weeks post operatively, the bone trabeculae became thicker around implant**.** It was also found that the residual grafted material was surrounded or in contact with the newly formed bone trabeculae. The bone formation was significantly increase in comparison to the Cerasorb group and significantly decrease in comparison to the control group.

Using B-tricalcium phosphate (Cerasorb) in eight dogs was performed by ***Artzi et al.,***(16) They found that in addition of being biocompatible materials, complete bone healing was established in all grafted defects after twenty four months post operatively, and the B-TCP particles (Cerasorb) were completely resorbed. ***Elswify et al.,***(2) studied the same effect of Cerasorb around implant at rabbit models and concluded the same results ***Elswify et al.,***(2) This contraversary may be due to the longer follow up periods of their experimental studies than the current study.

In the current study radiographic evidence of osseointegration was accepted with the absence of any intervening radiolucency between the implant and the bone with an increase in radio-opacity around implants and disappearance of the cerasorb radio-opaque particles from the four to the eight weeks. In the control group improvement also took place but to a lesser extent as compared to the study group. Where focal areas of radiolucency were still obviously seen around the inserted implants four week post implantation indicating less increase in bone density. In this investigation, radiographic results were supported by statistical analysis of bone density. which confirmed that bone density was greater using cerasorb in the two successive observation time intervals. A progressive increase in the mean values of bone density was detected in the control group and the study group throughout the experiment. Our result agree with the result of ***El Swify et al.,***(2) Who works on forty two rabbits.

Meanwhile radiograph findings of ***Suba etal.,***(17) at their experimental study of twenty two dogs , revealed that the healing process leading to osseointegration was retarded in the control group at the different observation periods (2,4 and 6 weeks) as compared to the experimental groups at the same observation time intervals.
The study groups showed better results of contact between implant and bone surface, indicating that ß-Tricalcium phosphate (Cerasorb) might promote faster and better healing than control in all periods. These observations were further supported by the current analysis in which the difference between the measured values of the bone implant interface gap dimensions in both groups was statistically significant ($p<0.000$) at four and eight weeks post-operatively.

Cerasorb proved to be an effective bone-replacing material with osteo-conductivity due to its gradual disintegration providing space for the regeneration of new bone. Radiographic evidence of osseointegration was accepted with the absence of any intervening radiolucency between the implant and the bone ***Cooper et al.,***[18] In the present study, routine radiographic examination of the study group showed marked increase in radio-opacity around implants with disappearance of the B-TCP radio-opaque particles from the control group period. In the control group of the current study, improvement also took place but with a lesser extent as compared to the study group where focal areas of radiolucency were still obviously seen around the inserted implants and post implantation indicating less increase in bone density. At the same time, radiographic results were supported by statistical analysis of bone density, which confirmed that bone density was greater using cerasorb in the two studied successive observation time intervals. A progressive increase in the mean values of bone density was detected in the control group and the study group throughout the experiment. This increase was found to be highly statistically significant ($p<0.001$).

Collectively, the radiographical and histological analysis of our results indicated that pure beta tricalcium phosphate can improve and accelerate the bone formation around dental implants.

Conclusions:

From the results of the current study, the influence of delivery of Pure Beta-tricalcium phosphate on implant osseointegration in dogs; showed significant more bone formation around the implants of the study group when compared with control group due to its osteoconductive and biocompatible properties.

Recommendations:

Larger sample sizes and longer evaluation period should be used to confirm study results

References

1. Amine K., Gharibi A., Hsaine A., Kissa J. Effect of Bone Regeneration with Mineralized Plasmatic Matrix for Implant Placement in Aesthetic Zone. Case Reports in Dentistry 2007;52:70–88.

2.El-Swify A., Magda A., Mohamed Z., Helal M. Comparative study between bioactive glass and B–tricalcium phosphate (Cerasorb) on healing of induced mandibular bone defects in rats. Egyption Dental Journal 2009;55:895–897.

3.Fairbairn and Leventis. Protocol for bone augmentation with imultaneous early implant placement: a retrospective multicenter clinical study. International Journal of Dentistry 2015;5:8–18.

4.Giglio J., Giannunzio G., Olmedo D., Guglielmotti B. Histomorphometric study of bone healing around laminar implants in experimental Diabetes. Implant Dentistry 2000;9:143–149.

5.Kim Y., Kim A., Rodriguez E., Nowzari H. Risk of prion infection through bovine grafting materials. Clinical Oral Implants Research 2016;18:1095–1102.

6.Mazor Z., Mamidwar S., Ricci L., Tovar M. Bone repair in periodontal defect using a composite of allograft and calcium sulfate (DentoGen) and a calcium sulfate barrier. Journal of Oral Implantology 2011;37:287–292.

7.Motamedian S., Khojasteh A., Khojasteh A. Success rate of implants placed in autogenous bone blocks versus allogenic bone blocks: A systematic literature review.Annals of Maxillofacial Surgery2016;6:78–90.

8.Miron R., Zhang Q., Sculean A., Buser D., Shirakata Y. Osteoinductive potential of 4 commonly employed bone grafts. Clinical Oral Investigations 2016;20:2259–2265.

9.Suehirof ., Komabashiri N., Masuzaki T., Ishii M., Yanagisawa T. Efficacy of bone grafting materials in preserving the alveolar ridge in a canine model. Journal of Dental Materials 2022;15:2–7.

10.Goodacre CJ., Bernal G., Rungcharassaeng K., Kan JYK. Clinical complications with implants and implant prostheses. J Prosthet Dent 2003;90:121–32.

11.Pal U., Neeraj K., Dhiman R. Evaluation of implants placed immediately or delayed into extraction sites. Natl J Maxillofac Surg. 2011;2: 54–62.

12.El-bahrawy A., El-hamadan S., Sharshar A. Comparative evaluation of bone regenerating capacity using nanocrystalline hydroxyapatite and coral composite: A Canine Model Study. J Curr Vet Res 2021;3:37–42.

13.Alnashar A., Ahmad I. Evaluation the effect of hyaluronic acid on bone healing process:An experimental study in the rabbits. Eur J Pharm Med Res Artic 2017;4:98–101.

14.Cai Z., Zhang T., Di L. Morphological and histological analysis on the in vivo degradation of poly (propylene fumarate)/(calcium sulfate/β- tricalcium phosphate). Biomed Microdevices 2011;13:623–631.

15.Horch H., Sader R., Pautke C., Neff A. Synthetic pure-phase beta-tricalcium phosphate (Cerasorb) for bone regeneration in the reconstructive surgery of the jaws. Int J Oral Maxillofac Surg 2006;35:708-13.

16.Artzi Z., Weinreb M., Givol N., Rohrer D. Biomaterial resorption rate and healing site morphology of inorganic bovine bone and btricalcium phosphate in the canine : A 24 months longitudinal histologic study and morphometric analysis. Int J Oral Maxillofac implant 2004;19:357–68.

17.Suba Z., Tacs D., Matusovits D., Barabas J. Maxillary sinus floor grafting with S-Tricalcium phosphate in humans: Density and microarchitecture of the newly formed bone. Clin. Oral Implants Res 2006;17:102–108.

18.Cooper F. Biologic determinants of bone formation for osseo-integration: Clues for future clinical improvements. J Prosthet Dent 1998;80:439–449.