

# Osseointegration of Zirconia (Y-TZP) Dental Implants: A Histologic, Histomorphometric and Removal Torque Study in the Hip of Sheep

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## ABSTRACT

**Objectives:** The purpose of the present study was to demonstrate the biocompatibility and rigid osseointegration of a zirconia (Y-TZP) dental implant with acid-etched surface.

**Materials and methods:** Eight zirconia (Y-TZP) implants (CeraRoot, Barcelona) were inserted in the hip of an adult sheep. The animal was sacrificed after 2 months. Four implants were used for histology and histomorphometric analysis, and the other four implants were used for removal torque test (RTQ).

**Results:** The histological evaluation showed a direct bone to implant contact (BIC) with no signs of inflammatory or foreign body reaction. The histomorphometric analysis values ranged from 79.5 to 79.9% of BIC. The RTQ showed values ranging from 75 to 83 Ncm.

**Discussion:** In this animal study, the biocompatibility of CeraRoot zirconia (Y-TZP) implants was demonstrated with the histologic observations of healthy bone tissue in direct contact with the zirconia (Y-TZP) implant surface. Moreover, the histomorphometric analysis and the RTQ confirmed a rigid fixation of the implant within the bone. Comparative studies with acid-etched titanium implants are needed to further evaluate the characteristics of CeraRoot zirconia dental implants.

**Keywords:** Zirconia implant, Ceramic implant, Y-TZP, Removal torque test, Histology, Histomorphometric, Zirconium, Etched surface, ICE surface.

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**Source of support:** Nil

**Conflict of interest:** Drs Oliva are family member owners and developers of the CeraRoot implant system.

## INTRODUCTION

The biocompatibility of zirconia (Y-TZP) as a dental implant material has been demonstrated in several animal investigations.<sup>2,3,5-12,16</sup> Also, *in vitro* experiments showed that the material is capable of withstanding simulated long-term load; however, the mechanical properties of zirconium seems to be influenced by mechanical preparation of the material.<sup>9,16</sup> Implant research shows that a rough surface topography is desirable to enhance the bone integration process,<sup>17</sup> but the turning of zirconium rods results in a relatively smooth surface. In this line, Sennerby et al (2005)

demonstrated a better implant retrieval torque resistance of porous zirconium surfaces in rabbits and similar results were obtained by Gahlert et al (2007). In a systematic review, Wenz et al (2008)<sup>18</sup> concluded that: Osseointegration of Y-TZP implants might be comparable to that of titanium implants; modifications of surfaces have the potential to improve initial bone healing and resistance to removal torque. The clinical application of Y-TZP implants was reported in a 5-year follow-up study of 831 CeraRoot implants, reported a success rate similar to titanium implants.<sup>13,14</sup> The purpose of the present study was to demonstrate the biocompatibility and rigid osseointegration of the CeraRoot zirconia dental implant with acid-etched surface (ICE™).

## MATERIALS AND METHODS

### Experimental Animals

One sheep (7 years, 50 kg) was used in this study. The animal was kept in purpose-designed site and fed on a standard diet. Twelve hours before surgery the animals received no more feed while water was accessible *ad libitum*.

### Ethical Approval

The study protocol was designed following the European Communities Council Directive of 24 November 1986. Adequate measures were taken to minimize animal pain or discomfort. A local ethics committee approved the study protocol in March 2010 (Fundació Hospital Asil de Granollers).

### Implant System

In this study, CeraRoot implant type 12 (Oral Iceberg, Barcelona, Spain) was used. The ceramic raw material was first pressed and molded into cylinders and presintered, machined to final shape with CAD/CAM, and finally sintered to full density.

Eight CeraRoot 12 implants were placed in the sheep hip. Four implants were used for histomorphometric study, and four implants were used for removal torque test (RTQ).

## Surgical Procedure

All surgery was performed under sterile conditions in a veterinary operating theater. The animals were sedated by intramuscular injection (10 mg/kg) of ketamine, 1 ml atropine and 5 mg/kg azaperone.

## Animal Sacrifice and Retrieval of Specimens

Three months after surgery the animals were sacrificed with an overdose of pentobarbital and potassium chloride, the hip was dissected and bone blocks containing implants were obtained. All specimens were fixed in 10% buffered formaldehyde and stored at room temperature for 10 to 15 days.

## Histological Preparation

The specimens were harvested and placed in 10% neutral buffered formalin. Upon receipt in the Hard Tissue Research Laboratory the implant, bone and soft tissue specimen were sectioned vertically in an anterior/posterior (mesial/distal) orientation according to protocol specifications. Immediately after sectioning specimens were dehydrated with a graded series of alcohols for 9 days. Following dehydration, the specimens were infiltrated with a light-curing embedding resin (Technovit 7200 VLC, Kulzer, Wehrheim, Germany). Following 20 days of infiltration with constant shaking at normal atmospheric pressure, the specimens were embedded and polymerized by 450 nm light with the temperature of the specimens never exceeding 40°C. The specimens were

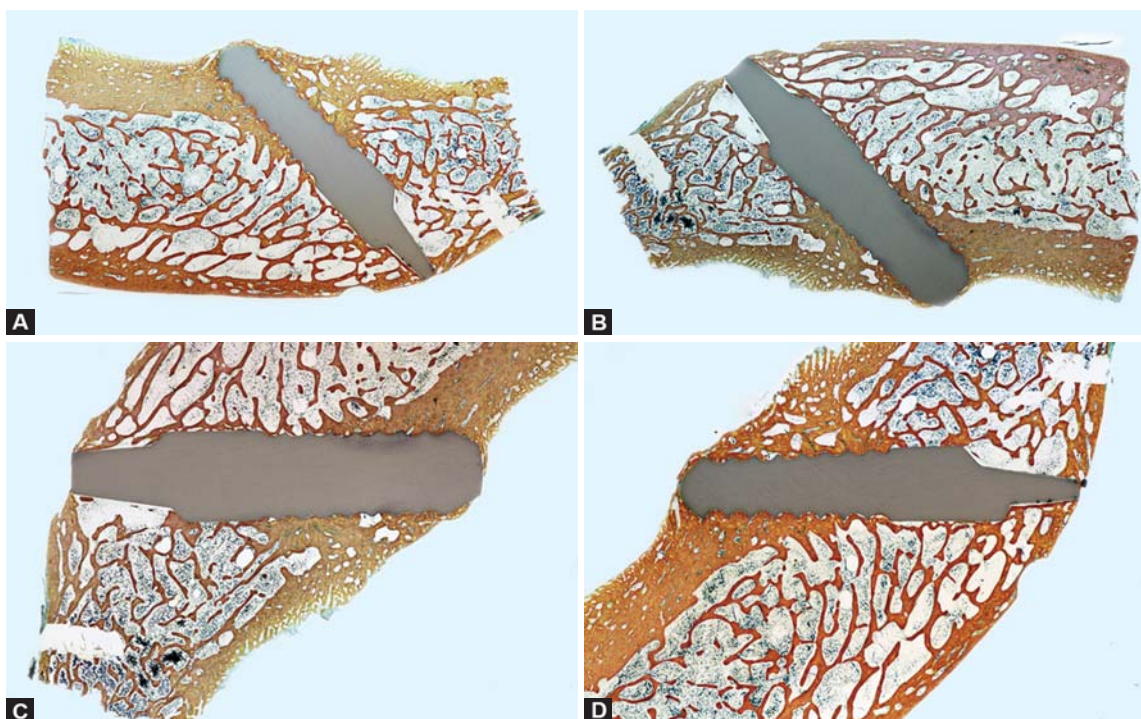
then prepared to by the cutting/grinding method of Donath<sup>4</sup> and Breuner (1982) and Rohrer and Schubert (1992).<sup>15</sup> The specimens were cut to a thickness of 150 µm on an EXAKT cutting/grinding system (EXAKT Technologies, Oklahoma city, USA). Following this, specimens were then polished to a thickness of 45 to 65 µm using a series of polishing sandpaper disks from 800 to 2400 grit using an EXAKT microgrinding system followed by a final polish with 0.3 µ alumina polishing paste. The slides were stained with Stevenel's blue and Van Gieson's picro fuchsin and coverslipped for histological analysis by means of bright field and polarized microscopic evaluation.

## Histomorphometric Analysis

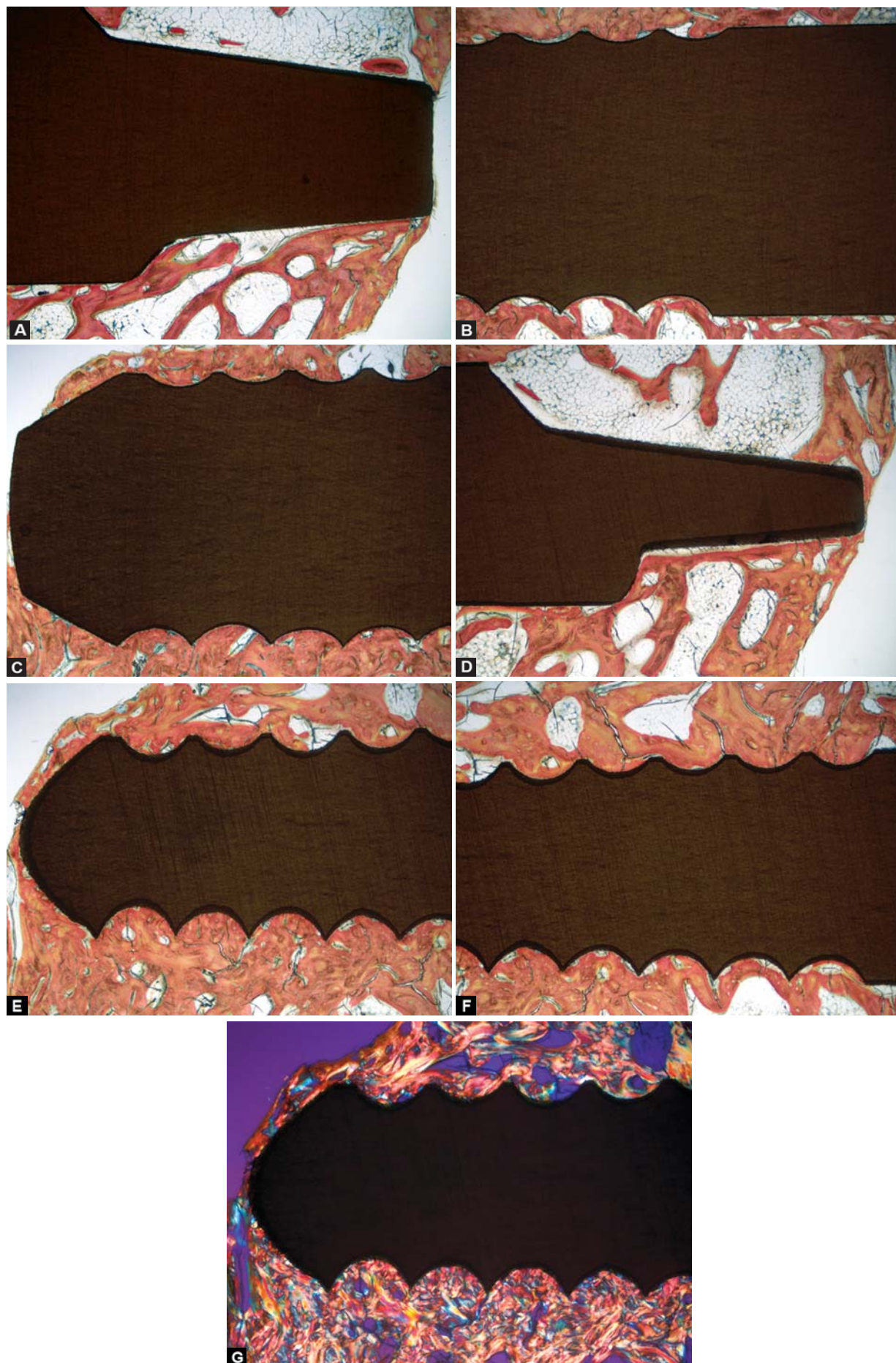
Following histological preparation, the specimens were evaluated histomorphometrically. All the specimens were digitized at the same magnification using a Nikon Eclipse 50i Microscope (Nikon Corporation, Japan) and a Spot Insight 2 mega sample digital camera (Diagnostic Instruments Inc, USA). Histomorphometric measurements were completed using a combination of spot insight program and Adobe PhotoShop (Adobe Systems, Inc.). At least two slides of each specimen were evaluated.

## RTQ Test Procedure

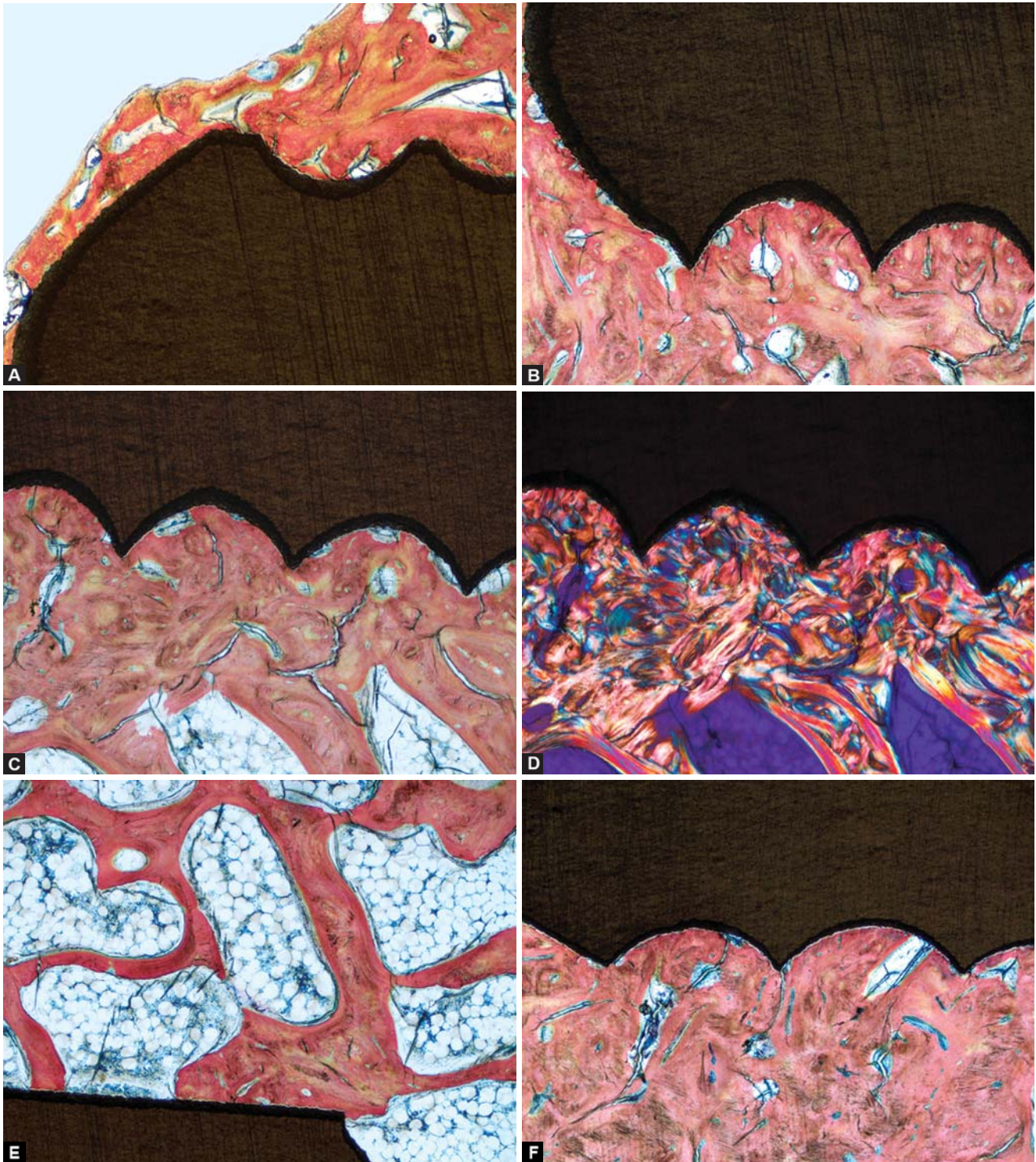
Four implants were subject to RTQ tests using a specially designed electronic device. The instrument involved an electric motor with a strain gauge mounted on a metal frame. The instrument was connected to the implant head of the



Figs 1A to D: Magnification 2.5x



**Figs 2A to G:** Magnification 20x



Figs 3A to F: Magnification 40x

ceramic implants. A fixed rotation rate was applied until failure of the bone-implant interface occurred. The peak RTQ was registered.

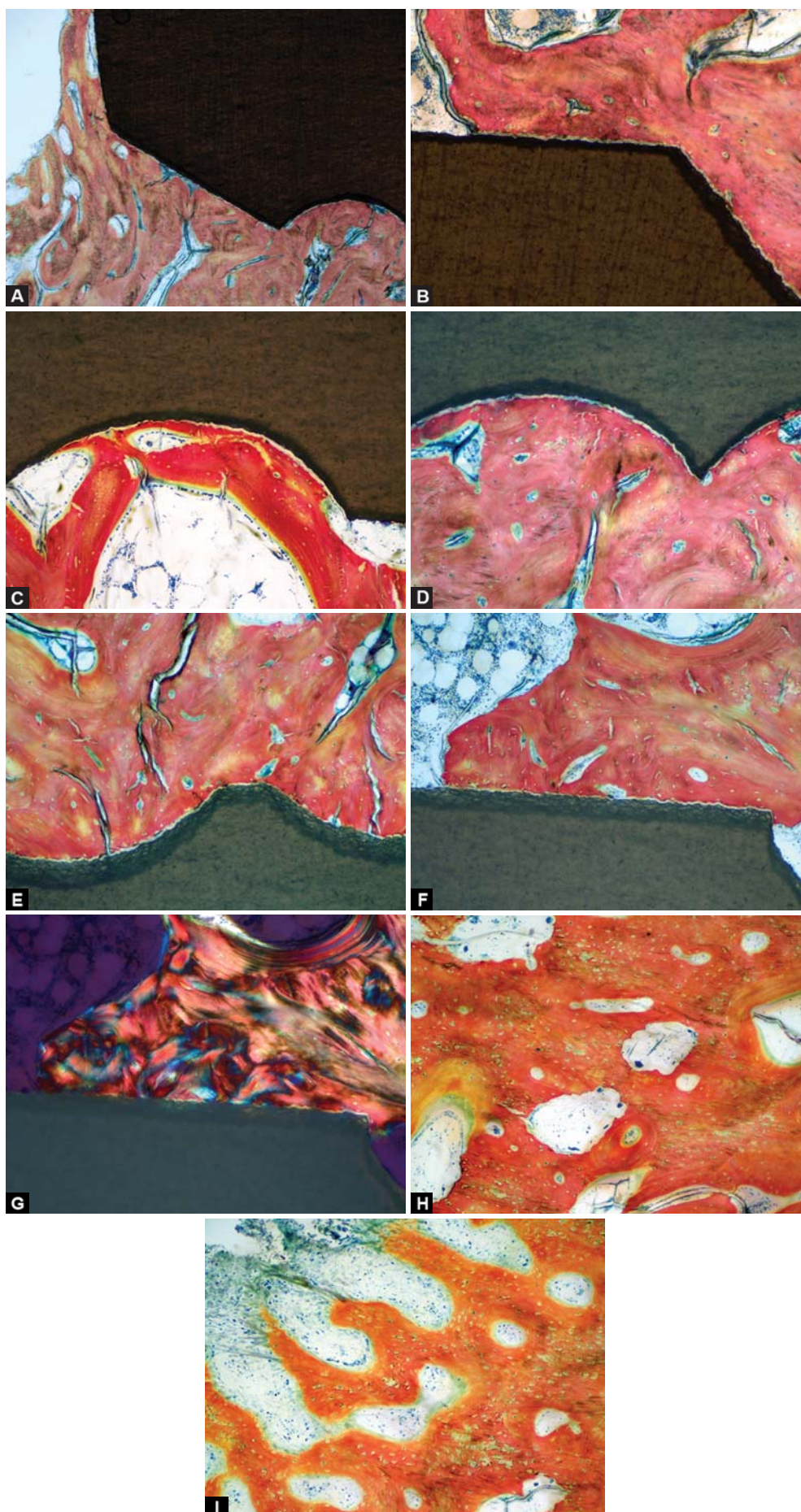
## RESULTS

### Clinical Observations

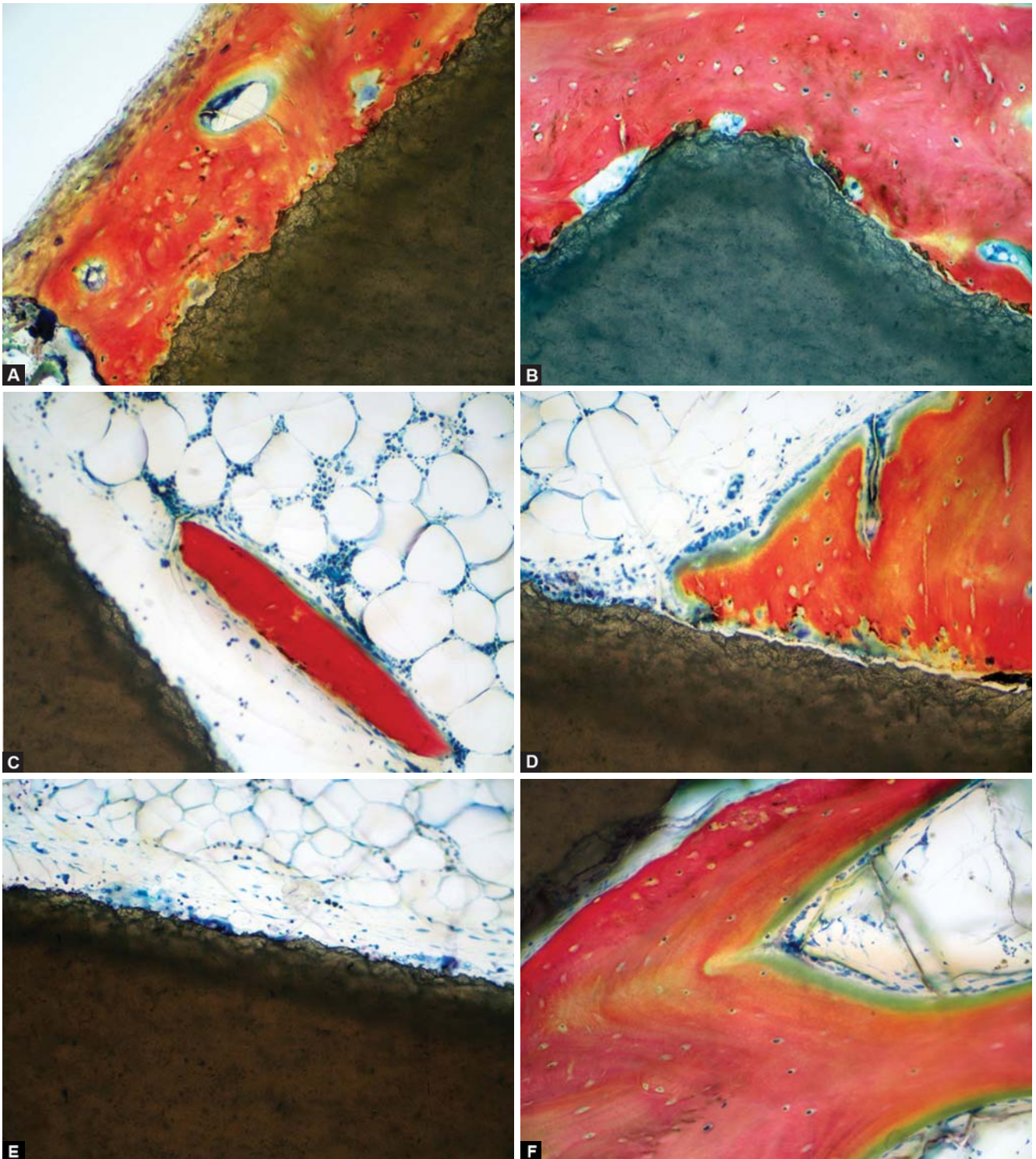
All implants achieved good primary stability. The healing period was uneventful, and the experimental sites healed well during the 12 weeks.

### Histological Analysis

The bone is much thicker around the implants than in the surrounding cancellous bone. The zirconia (Y-TZP) dental implants show excellent osseointegration. The low power (20x) photomicrographs, show the very solid bone contact (Figs 2B to F) and the area with less bone contact (Fig. 2A). Figure 2G, the low (20x) and medium power (40x) images (Figs 3A to F) show an implant that included most of the threads and attached bone. Figures 1A to 2D all show the



Figs 4A to I: Magnification 100x



**Figs 5A to F:** Magnification 200x

integration of most of the thread with vital bone. The polarized views (Figs 2G, 3D and 4G) show that the bone surrounding the implant is mature and remodeled. The medium power (40x) photomicrographs, Figures 3A to F show very good, continuous bone along the surface of the implant. The polarized view, Figure 3D, shows the junction of the bone and connective tissue area. The medium power images emphasize the excellent osseointegration as seen

very well in Figure 3B. This is normal and healthy cancellous bone. Figure 3E shows how the integrated bone on the implant blends with the thick, vital cancellous bone. The high (100x) (Figs 4A to I) and very high (200x) power images (Figs 5A to F) show in detail the intimate contact of the vital bone with the threads of the zirconia (Y-TZP) dental implants. Figures 4D and E show a thread surrounded by very solid trabecular bone. Figure 5B shows a good amount

of bone, including new bone formation, around the thread. The light green-staining connective tissue could have become osteoid and eventually formed bone as shown in Figure 5F (200×). In these photomicrographs, different staining qualities reflect the varying maturity levels of the different areas of the bone; darker red-staining bone is less mature than the lighter staining bone. We would assess this implant as being very well integrated into vital bone. From the histological point of view, implant appears to have an excellent osseointegration.

### Histomorphometric Study

The bone to implant contact (BIC) implant values ranged from 75.6 to 79.9%. The bone is much thicker around the implants than in the surrounding cancellous bone.

### RTQ Test

The RTQ values ranged from 75 to 83 Ncm (Table 1).

### DISCUSSION

Akagawa et al (1998)<sup>1</sup> examined the possibility of the long-term stability of osseointegration around partially stabilized zirconia (Y-TZP) implants with a one-stage procedure with different loading designs in the mandibles of eight monkeys. They did not observe a clear difference in the clinical features among the different types of support. Direct bone apposition to the implant was generally seen in all the groups. The bone contact ratio ranged from 66 to 81%, and the bone area ratio varied between 49 and 78% at 24 months after loading. Kohal et al (2004) investigated the osseointegration of loaded zirconia (Y-TZP) implants in comparison with titanium implants in the maxilla of six monkeys. The titanium implant surfaces were sandblasted with Al<sub>2</sub>O<sub>3</sub> and subsequently acid etched. The zirconia (Y-TZP) implants were only sandblasted. The mean BIC after 9 months of healing and 5 months of loading was 67.4% (SD: 17%) for the zirconia (Y-TZP) implants and 72.9% for the Ti-SLA implants. No statistically significant

difference between the different implant materials was detected. Gahlert et al (2007) investigated zirconia (Y-TZP) implants with either a machined or a sandblasted surface and compared them with sandblasted and acid-etched Ti-SLA implant surfaces in the maxilla of minipigs. The machined ZrO<sub>2</sub> implants showed statistically significant lower RTQ values than the other two implants types after 8 and 12 weeks, while the SLA implant showed significantly higher RTQs values than rough ZrO<sub>2</sub> surfaces after 8 weeks. The authors concluded that roughening the machined zirconia (Y-TZP) implants enhances bone apposition and has a beneficial effect on the interfacial shear strength. In addition, 13 implants were evaluated histologically for the apposition of mineralized structures after 4 and 12 weeks of healing. Even though the Ti-SLA surface revealed a stronger bone response after all healing periods in comparison with the zirconia (Y-TZP) implants, the results for sandblasted ZrO<sub>2</sub> implants were superior to those for machined ZrO<sub>2</sub> implants. In more recent publications Gahlert et al (2009, 2010) compared titanium implants and zirconia implants and did not find differences (although not significant) between the groups in terms of BIC and RTQ values.

### CONCLUSION

The present results show biocompatibility and osseointegration capacity for the CeraRoot zirconia (Y-TZP) dental implant with acid-etched surface, without signs of inflammatory or foreign body reactions. The histomorphometric analysis and RTQ test demonstrates a rigid fixation between the implant and bone.

### ACKNOWLEDGMENT

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**Table 1:** RTQ test and histomorphometric analysis (BIC)

	RTQ (Ncm)	BIC (%)
Sample 1	83	—
Sample 2	84	—
Sample 3	75	—
Sample 4	81	—
Sample 5	—	79.5
Sample 6	—	79.9
Sample 7	—	79.8
Sample 8	—	75.6
Average	80.75	78.7

(Average is not significant due to the small quantity of studied specimens)

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