ANATOMO-RADIOLOGICAL ASSESSMENT OF INCISIVE CANAL USING CONE BEAM COMPUTED TOMOGRAPHS

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ABSTRACT

Purposes: To determine the anatomical variability of the incisive canal (IC) and to clarify its characteristics, using cone beam computed tomography (CBCT) evaluation, in order to avoid intra- and post-operative complications in oral surgery.

Materials and Methods: The diameter, length and angulations of the IC and the width of the bone anterior to the canal were measured on 100 digital CBCT using the Galileos Implant 1.9 software. The patients' ages ranged from 14 to 54 years (53 males and 47 females). The data were analyzed using the Stat View 5.0 software.

Results: The anatomy of the IC shows a large variability in its morphology and dimensions, with the canal branching into up to four canals at the nasal level. The length of the IC varies from 4.94 mm to 26.13 mm with an average of 11.42 (± 2.71) mm with a statistically significant difference between genders. The ICs were classified into 6 categories according to their shape viewed on the sagittal sections, with two new shapes found. The incisive foramen has four shapes with a statistical correlation with the gender of the subject. The bone thickness anterior to the canal shows a statistical correlation with the gender of the subject and the presence or absence of the central incisor.

Conclusions: The present study highlights an important variability in the anatomy and morphology of the IC. Therefore, a CBCT analysis of this structure is essential to avoid any complications before any surgery in this area.

KEY WORDS: Incisive canal, anatomy, gender, complications, dental implants, cone-beam computed tomography, nerve injuries.

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INTRODUCTION

The incisive canal (IC), also known as the nasopalatine canal or anterior palatine canal, is a relatively long narrow structure located in the midline of the maxilla. The canal contains fibrous connective tissue, fat, and even small

salivary glands [1]. The terminal branch of the descending palatine artery and the nasopalatine nerve pass through this tunnel to communicate with the posterior septal branch of the sphenopalatine artery and greater palatine nerve, respectively [2]. This bony canal connects the

oral cavity roof with the nasal cavity floor. The canal continues in the oral cavity as a single incisive foramen posterior to the central incisor teeth (Fig. 1) and in the nasal cavity as the foramina of Stenson, which are frequently two in number. In some cases, two additional minor canals can be seen (foramina of Scarpa), which may transmit the nasopalatine nerve [3].

A thorough knowledge of the anatomical appearance of the IC and its variations is essential prior to any surgical procedure such as implant placement and local anesthesia in the anterior maxilla. Difficulties and anatomical limitations regarding the location of the IC in relation to the maxillary central incisor implants have been reported [4]. The implant contact with neural tissue may result in a failure of osseointegration or lead to sensory dysfunction [1]. The IC can also be involved in several pathologies, such as the nasopalatine duct cysts, and it may be difficult to decide through a radiograph whether any radiolucency in that area is a cyst or a large incisive foramen [5,6]. Much of this uncertainty has arisen because of two-dimensional (2D) representations of the three-dimensional (3D) anatomy of the IC in radiography. Innovations in imaging systems and the increased use of pre-operative CBCT evaluation have allowed us to obtain a more accurate and closer visual examination of these anatomical structures and pathologies [7]. Nevertheless, the anatomical variations of the IC are not very well documented in the literature and are often presented as case reports [3,8].

The aim of this study is to identify the anatomical characteristics of the IC in terms of its morphology, dimensions, relation to the central incisors and buccal bone thickness in order to avoid intra- and post-operative complications in oral surgery, and to search for any possible correlations of these variables with age and gender.

MATERIALS AND METHODS

Patient selection: The main source of data for this study was a group of patients presenting at the Outpatients and Implantology Department in Oral Medicine and Surgery at the Faculty of Dental Medicine, Monastir, Tunisia, between April 2014 and February 2015.

The study material included 100 CBCT CDs taken from patients during the pre-operative planning of implant placement in the incisor region. CBCT was performed with the Sirona GALILEOS Comfort unit and the playback of the acquired images was performed with the Galileos Implant 1.9 software. The inclusion criteria were a 100 digital CBCT involving the entire IC in all the three planes and the absence of any IC pathology or impacted teeth in the region. The patients were informed about the rationale behind the study and the methods applied, and an informed consent was obtained.

Measurements: The measurements were performed by one operator (an anatomy specialist) and for every parameter the mean of two measurements, separated by forty days, was calculated. Certain points of reference were used to complete the measurements:

IC nasal level: the line joining the anterior nasal spine and posterior nasal spine plane.

IC palate level: the horizontal plane passing from the incisive foramen.

Canal long axis: the line passing from the middle of the antero-posterior nasal foramen diameter to the middle of the antero-posterior incisive foramen diameter.

The main parameters measured were:

The number of openings of the IC at the nasal level (Fig. 2).

The canal shape in the sagittal plane: the selected slice was that in which we were able to see the whole of the IC. The antero-posterior diameter in the three levels of the IC (nasal, middle and oral) was compared and the sagittal shape was classified according to this comparison (Fig. 3).

The canal shape in the frontal plane: the slice selected in which we were able to see the whole of the IC in the frontal plane. It was classified according to the frontal division of the canal.

The IC division levels: these were assessed in the frontal plane based on the coronal ratio, which is the ratio of the canal length above the division to the entire length of the canal in the frontal plane (Fig. 4).

The canal angulation: this was measured (in degrees) in the sagittal plane, between the

vertical plane and the long axis of the canal using the measurement tool provided in the Galileos Viewer software (Fig. 5).

The canal length: this was measured (in mm), in the sagittal plane, between the IC nasal level (the middle of the antero-posterior nasal foramen) and the IC palatal level along the vertical plane using the measurement tool provided in the Galileos Viewer software (Fig. 6).

The antero-posterior and medio-lateral dimensions of the canal: for each level (nasal, middle and palate), the main IC dimensions (the antero-posterior and medio-lateral diameters) were determined in the axial reconstruction using the measurement tool provided in the Galileos Viewer software (Fig. 7).

The shape of the incisive foramen: this was determined at the palate level of the IC in the axial reconstruction by inspecting the contours of the IC foramen (Fig. 8).

The bone thickness anterior to the canal: this was evaluated (in mm) from the buccal limit of the IC to the facial buccal bone limit at three levels: nasal, middle and palate (Fig. 9).

All the data were first analyzed using descriptive statistics. The differences in the CBCT measurements between the patient subgroups were compared using either the t-test, one-way ANOVA, correlation Z test or simple regression test, as appropriate. A multivariate linear regression model was used to evaluate the association between the digital parameters. The significance level chosen for all the statistical tests was $P < 0.05$. All the analyses were performed using the Stat View 5.0 software.

RESULTS AND DISCUSSION

Among the 100 patients with an age range of 14 to 54 years, 53 were males (with a mean age 37.35 ± 11.78) and 47 were females (with a mean age 32.06 ± 11.18). The gender ratio was 1.12, proving the homogeneity of the sample (Fig. 10). Thirty-eight percent of the patients had a missing central incisor.

The main results comparing the different parameters, male and female and with the central incisor present or missing, are reported in tables 1 and 2.

In the present study, we observed that most

subjects had one opening at the nasal fossa. Thirty six percent of the patients had two openings, while 8% had three openings, and only 2% had four openings (Fig. 11). We found up to four foramina at the IC nasal level. Similar findings, in previous studies, were also reported observing up to four foramina at the IC nasal level (1, 7, 8). Sicher reported that there could be up to six separate foramina (9), the additional foramina being labeled the foramina of Scarpa (10). However, other authors observed only two foramina (2, 10). This variability in results could be due to sample differences and the various imaging techniques used in the different studies.

The mean antero-posterior diameter at the nasal foramen was 2.84 mm (± 1.29), the diameter ranging from 0.56 mm to 6.02 mm. The mean medio-lateral diameter at the foramen of Stenson was 2.62 mm (± 1.35), the diameter ranging from 0.45 mm to 6.85 mm. The greatest diameter was observed in the canals with a single opening at the nasal fossa and the shortest diameter was noted in the canals with four openings. No statistically significant differences between males and females and the different age groups with respect to the number of openings or average medio-lateral diameter of the foramen of Stenson were observed in the present study ($P > 0.05$). There was also no significant correlation between the antero-posterior and medio-lateral nasal foramen and the central status ($P = 0.75$).

Fig. 1 : A clinical view of the incisive foramen.

This figure illustrates the proximity between this structure and the alveolar crest of a central incisor missing intended to be replaced by an implant.

Fig. 2: Number of nasal openings determination.

B: CBCT horizontal reconstruction passed from the IC nasal level (1), determined in the CBCT sagittal reconstruction (A), showing two nasal foramina.

Fig. 3: IC sagittal shape determination.

CBCT sagittal reconstruction comparing the antero-posterior diameter (a, b, c) at the IC three levels: palate (e), middle and nasal (d).

Fig. 4: The IC division level determination.

CBCT frontal reconstruction showing the level of division of the IC which is the ratio between the IC frontal division length (B) and IC frontal length (A). a: IC nasal level. b: division plane level. c: IC palate level.

Fig. 5: IC angulation measurements determination.

Sagittal reconstruction illustrating the IC angulation (A°) between the vertical plane (b) and the canal long axis (c). a: IC nasal level.

Fig. 6: Canal length measurements.

CBCT sagittal reconstruction revealing length

measurement (b) between the middle antero-posterior nasal foramen at the IC nasal level (a) and the IC palate level (c).

Fig. 7: IC diameter determination.

Sagittal reconstruction (A) showing the different section levels on which measurements of the IC diameter were carried out. B, C, D: Axial reconstruction outcome from respectively the IC palate (b), middle and nasal levels (a). 1: antero-posterior diameter. 2: medio-lateral diameter.

Fig. 8: Shape of the incisive foramen determination.

The shape is determined by inspecting the contour of the incisive foramen in the CBCT horizontal reconstruction (B) passed from the aa' section (the IC palate level). A: CBCT sagittal reconstruction showing the whole of the IC.

Fig. 9: Bones thickness anterior to the IC measurements.

Sagittal reconstruction revealing bone thickness determination (a, b, c) at respectively the palate level (1), the middle level and the nasal level (2).

Fig. 10: Distributions of patients according to age and sex.

We can note in this histogram the homogeneity of the sample.

Fig. 11: The number of openings of the IC at the nasal level.

Axial CBCT reconstruction illustrating the different situations found. A: one opening; B: two openings; C: three openings; D: four openings.

Fig. 12: IC sagittal shape.

Sagittal CBCT reconstructions showing the six shapes of the incisive canal: A: Hourglass shape: 47% of cases. B: Inverted funnel shape: 38% of cases. C: Spindle shape: 6% of cases. D: Funnel shape: 4% of cases. E: "Y" shape: 3% of cases. F: Cylindrical shape 2% of cases.

Fig. 13: IC frontal shape and level of division.

Frontal CBCT reconstructions illustrating the different frontal shapes and level divisions of the IC. We note in figure A: An "I" shape without division and in the others figures different division levels with B: Nasal division level. C: Middle division level and D: Oral division level.

Fig. 14: Negative IC angulation.

Sagittal reconstruction revealing a posterior location of the incisive foramen compared to the nasal one.

Fig. 15: Different shapes of the incisive foramen.

Horizontal CBCT reconstructions revealing four different shapes of the incisive foramen.

Table 1: Summary table revealing comparative statistical analysis of dentate and edentulous subjects.

Parameters		Central incisor present	Central incisor missing	P (significant or not)
Age (years)		33.25(±12.2)	37.5(±16.9)	p =0.14
Medio-lateral diameter of the IC (mm)	Oral level	3.42(±1.22)	3.73(±1.33)	P=0.22
	Midpoint level	2.71(±1.26)	2.78(±1.35)	p =0.79
	Nasal level	2.52(±1.28)	2.76(±1.45)	p =0.38
Antero-posterior diameter of the IC (mm)	Oral level	3.74(±1)	3.74(±0.9)	p =0.8
	Midpoint level	2.49(±0.9)	2.78(±1.16)	p =0.5
	Nasal level	2.82(±1.33)	2.9(±1.22)	p =0.75
Length of the IC (mm)		11.37(±2.35)	11.51(±3.31)	p =0.79
Angulation (degree)		17.71(±7.22)	18.85(±6.70)	p =0.43
Shape of the canal in the frontal plane		$\chi^2=0.13$		p = 0.7
Shape of the incisive foramen		$\chi^2=4.5$		p = 0.21
Number of nasal foramen		$\chi^2=0.8$		p =0.84
Bone thickness (mm)	Oral level	7.99(±1.5)	6.99(±1.15)	p < 0.0001*
	Midpoint level	8.45(±1.47)	7.54(±1.5)	p =0.007*
	Nasal fossa level	11,58(±3.07)	10,07(±2.96)	p =0.31

Table 2: Summary table revealing comparative statistical analysis between male and female subjects.

Parameters		Male	Female	P (significant or not)
Age (years)		37.35(±11.78)	32.06(±11.18)	p =0.08
Medio-lateral diameter of the IC (mm)	Oral level	3.72(±1.4)	3.32(±1.03)	P=0.12
	Midpoint level	2.83(±1.42)	2.63(±1.13)	p =0.44
	Nasal level	2.67(±1.46)	2.55(±1.2)	p =0.99
Antero-posterior diameter of the IC (mm)	Oral level	3.80(±1.09)	3.68(±0.79)	p =0.48
	Midpoint level	2.77(±0.88)	2.41(±0.61)	p =0.33
	Nasal level	2.91(±1.21)	2.78(±0.95)	p =0.86
Length of the IC (mm)		12.14(±2.85)	10.60(±2.39)	p =0.02*
Angulation (degree)		17.69(±6.84)	18.66(±7.25)	p =0.44
Shape of the canal in the frontal plane		χ^2 =0.21		p = 0.64
Shape of the incisive foramen		χ^2 =8.78		p = 0.03
Number of nasal foramen		χ^2 =3.65		p =0.3
Antero-posterior buccal bone thickness (mm)	Patients with central incisor present			
	Oral level	7.99(±1.5)	6.99(±1.15)	p =0.004*
	Midpoint level	8.45(±1.47)	7.54(±1.5)	p =0.01*
	Nasal fossa level	11,58(±3.07)	10,07(±2.96)	p =0.05*
	Patients with central incisor absent			
	Oral level	5.25(±2.16)	6.2(±1.78)	p=0.16
	Midpoint level	7.34(±1.85)	8.18(±1.39)	p=0.14
	Nasal fossa level	9.8(±2.85)	10.64(±2.58)	p=0.41

The ICs were classified into 6 categories according to the shape viewed on the sagittal sections (Fig. 12):

A cylindrical shape: this is formed by parallel labial and palatal walls of the IC (the same antero-posterior dimension in the three levels of the IC).

A funnel shape: this is formed by an increasing antero-posterior dimension of the IC from the nasal to the palate levels.

An inverted funnel shape: this is formed by an increasing antero-posterior dimension of the IC from the oral to the nasal levels.

An hourglass morphology: this is a shape in which the narrowest antero-posterior dimension of the IC is at the mid-level compared to the dimensions at the nasal and hard palate levels.

Spindle morphology: this is a shape in which the widest antero-posterior dimension of the IC is at the mid-level compared to the dimensions at the nasal and hard palate levels.

A "Y" shape: this is a morphological feature in which two canals are observed, an anterior small canal and a posterior canal.

The most commonly encountered shape was the hourglass shape, which was seen in 47% of

cases, and the least common one was the cylindrical shape (2% of subjects). However, in previous studies the cylindrical shape was the one most commonly found, being present in most subjects (1, 7, 8).

Two new shapes were found in the present study: the "Y" shape and the inverted funnel shape.

No statistically significant differences between the genders and between the different age groups with respect to the shape of the IC were observed ($p > 0.05$).

The evaluation of the different anatomical variations of the IC in the frontal plane resulted in the following findings: a single canal was identified in 55% of the subjects and variations of the Y type of the canal were seen in 45% of the cases. Authors previously described three shapes on the frontal plane ("I", "Y" and "II") [11]. In our study the "II" shape was not found. Similar findings with regard to the frequency of a single-canal configuration were reported in a recent study on 56 human cadavers, where microscopic computed tomography of the anterior maxilla was performed. The authors reported about 42.9% of the cases having a single type of canal [2].

Other study reported also that the "I" shape was the one most commonly observed [11].

The correlation between the frontal morphology and gender was not statistically significant ($\chi^2 = 1.8$; $p = 0.4$).

The ICs were divided into 3 categories:

A coronal ratio of 1-0.65: a division of the canal in the lower-third level.

A coronal ratio of 0.66-0.33: a division of the canal in the middle-third level.

A coronal ratio <0.33: a division of the canal in the upper-third level.

Most commonly, the IC division was in the nasal third level (64%), followed by the middle third level (31%). Only on 2 occasions was the division found to be in the lower-third level (5%) (Fig. 13). It had previously been reported that the dividing point of the IC in the coronal plane is at about the level of the upper fifth of the IC [2]. However, another study reported that the IC was, most commonly, divided within the middle-

third level (51%), and followed by a division in the upper-third level (47%) [8].

The IC division levels were not statistically significant difference between males and females ($\chi^2 = 2.11$ and $p = 0.54$). The same observation can be made in respect of the different age groups ($p < 0.05$). However, some authors previously reported in their studies a gender correlation with the level of the division but no similar correlation between the different age groups [8].

The mean angulation of the IC in our study was $18.15^\circ (\pm 7.02^\circ)$ varying from -4° to 34° . Liang et al previously reported a mean angulation of the IC from the vertical of $12.6^\circ (\pm 8.9^\circ)$ and Arpita et al a mean angulation of $27^\circ (\pm 8.03^\circ)$ from the vertical [1,8].

Some subjects demonstrated negative values i.e.; the incisive foramen could be located posterior to the nasal foramen (Fig. 14). This result was in contrast with the study of Arpita et al in 2013, who reported that the incisive foramen was in all the cases located anterior to the nasal foramen [8].

No statistically significant correlation was found between the angulation and the buccal bone thickness at the different levels, gender and central status did not having a significant effect on angulation. In accordance with these results, some authors previously reported similar findings [8,1,12].

The canal length ranged from 4.94 mm to 26.13 mm, the mean being $11.42 (\pm 2.71)$ mm. Song et al reported the length of the IC to be 12.0 mm (8.4-15.8 mm) in dentulous maxillae (2). Mraiwa et al reported a mean length of $8.1 (\pm 3.4)$ mm, Liang et al in their study assessed the length of the IC as being $9.9 (\pm 2.6)$ mm and for Arpita et al, the mean length was found to be $10.08 (\pm 2.25)$ mm [1,7,8].

Among the different age groups, no statistically significant differences in the length of the IC were observed ($p = 0.44$). Some authors previously reported that age had a significant influence on the length of the IC, with a decrease of the mean value in measurements relating to older patients [11]. This finding was also reported in a recent study involving 207 subjects using CT scans before dental implantation [12]. In this study, a

not statistically significant correlation was found between gender and the length of the IC. Similar findings were reported by others [8].

The central incisor (missing or present) did not show a significant correlation with the length ($p=0.7$). In the literature, the status of the central incisors significantly influences the length of the IC [11,13]. It was also previously reported that the period of the incisor loss statistically influences the IC length [11,13,14].

Due to the absence of any such relevant information (many of the patients did not remember exactly the period of the incisor loss) this parameter was not analyzed in this study.

However, there was a highly statistically significant difference ($p=0.004$) in the length of the canal between males (mean 12.15 (± 1.96) mm) and females (mean 10.61 (± 1.88) mm). Similar findings were previously reported [1,8]. The longer length of the IC in the measurements of male subjects could be attributed to the relatively larger cranio-caudal dimension of the face observed in males as compared to females [8].

The mean antero-posterior and medio-lateral diameters of the IC were respectively 3.07 (± 0.89) mm and 2.97 (± 1.08) mm. The average antero-posterior and medio-lateral diameters at the IC nasal level were respectively 2.84 mm (± 1.08) and 2.62 mm (± 1.05). At the midlevel they were 2.60 mm (± 0.77) and 2.73 mm (± 1.77), and at the palatal level they were 3.74 mm (± 0.76) and 3.55 mm (± 0.99). The differences in the values between males and females and among the different age groups were not found to be statistically significant. Similar findings were previously reported [1,8,12]. However, other authors found that the canal diameter increased with age [14].

A statistically significant correlation between the antero-posterior and medio-lateral diameters was found ($p < 0.0001$). There was a not statistically significant correlation between the IC diameter and the central incisor whether present or missing ($p=0.49$). An increase in the canal dimensions in edentulous patients had been reported in 207 CT scans examination [14].

Based upon these findings, some authors suggested that the canal diameter, and similarly the maxillary sinus, became larger with tooth

loss [14]. On the contrary, other studies conducted could not support this hypothesis [1,8,13]. The canal diameter did not change with the dental status in either gender, no statistical difference being noted in respect of the canal diameter. For this reason, more studies in this area are needed.

The morphology of the incisive foramen was divided into four shapes. Most of the patients (62%) had an oval shape, twenty one percent of the subjects had a heart shape, sixteen percent had a clover shape and only one percent of the patients had a pike shape (Fig. 15). The shape of this foramen has not previously been described in any other study. The shape of the incisive foramen showed a statistically significant correlation with gender ($\chi^2 = 8.78$ and $p = 0.03$). The heart shape is most commonly observed in male subjects, while the clover shape is commonly observed in female subjects. However, the oval shape was observed in both genders. We found only one male with a pike shape.

The incisive foramen shape also showed a statistical correlation with the frontal shape: the clover and the heart shape were commonly associated with the "Y" shape. However, the oval shape was commonly associated with the "I" shape ($\chi^2 = 33.96$ and $p < 0.0001$).

The mean diameter of the incisive foramen in the present study was 3.64 mm (± 0.82). This value was lower than that reported in the previous study (4.6 mm) [7]. However, it was comparable to others studies 3.4 mm for Liang et al and 3.6 mm for Arpita et al [1,8]. The diameter of the incisive foramen is usually considered to be below 8 mm; when it exceeds 10 mm, pathological conditions should be considered [1]. In the present study, the diameter of the incisive fossa ranged from 1.43 mm to 6.42 mm, in accordance with the previously accepted values [7]. The mean diameter did not show any statistically significant differences between males and females ($p = 0.12$) or among the different age groups. The dimensions of the buccal bone demonstrated an increasing width from the oral level to the nasal level, with corresponding mean values of 6.77 (± 1.9) mm in the oral level (the measurements ranging from 0.92 mm to 12.33 mm), 7.67 (± 1.87) mm in the midline (ranging

from 2.65 mm to 11.94 mm), and 10.56 (± 2.96) mm in the nasal level (ranging from 2.24 mm to 17.51 mm). Similar findings were previously reported with corresponding values of 6.5 mm at the oral level, 6.59 mm at the middle level and 7.6 mm at the nasal level [11].

A statistically significant correlation was found between the antero-posterior buccal bone thickness and the missing central incisor at the oral level ($p < 0.0001$) and midline level ($p = 0.007$). The central status significantly influenced the width of the buccal bone, with decreasing values for patients with missing central incisors. Bornstein et al evaluated the effect of the dental status on the buccal bone width and reported that the width of the buccal bone is highest in cases when both centrals are present compared to subjects with one or two missing central incisors [11]. Same findings had been reported [12]. However, in our study, a not statistically significant correlation was noted between the antero-posterior bone thickness at the nasal level and the missing central incisor ($p = 0.31$). This result can be explained by the absence of any influence of the central incisor in this level because these measurements were carried out at a level higher than the incisive root limit (in the basal bone).

A gender significant difference was noted at the three levels of the buccal bone thickness for the patients with a present central incisor ($p = 0.0004$ at the oral level, $p = 0.03$ at the middle level and $p = 0.05$ at the nasal level). The same result could not be observed in patients having a missing central incisor. Similar findings were previously reported [8,11]. This result could be attributed to the larger dimension of the maxilla in males compared to females.

CONCLUSION

This study highlights the anatomical variability of the IC in relation to several parameters. New findings are reported in this study, such as the incisive foramen shape and its gender and frontal shape correlation. Two new sagittal shapes have been discovered. The length of the IC shows a significant difference between male and female subjects. The central incisor state (present or absent) reveals a significant correlation with the bone thickness. The anatomical

variations in terms of dimensional and morphological parameters are significant, emphasizing the role of 3D imaging and the assessment of this anatomical landmark in the treatment planning of this area for implant placement (such as IC displacement, canal occlusion by bone graft, using the canal as a preparation site, or vestibular bone graft). In the assessment of pathologies in this region (for example, the differential diagnosis between a cyst and the normal canal morphology); and in preventing inadvertent complications while operating in the anterior maxillary region (any implant contact with the IC may lead to implant loss or postoperative sensitivity). Based on these findings, limited CBCT imaging, as an adjunctive radiographic method, is of great value in terms of determining the dimensions and morphology of the IC before dental implant surgery, especially when one or both central incisors have been missing for a long period of time.

Conflicts of Interests: None

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