CLINICAL ORAL IMPLANTS RESEARCH

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# Influence of simulated bone–implant contact and implant diameter on secondary stability: a resonance frequency *in vitro* study

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**Abstract**

**Objectives:** This study tested the hypothesis of no differences in resonance frequency for standardized amounts of simulated bone–implant contact around implants with different diameters. In addition, it was evaluated if resonance frequency is able to detect a difference between stable and rotation mobile ("spinning") implants.

**Material and Methods:** Implants with diameters of 3.3, 4.1 and 4.8 mm were placed in a purposely designed metal mould where liquid polyurethane resin was then poured to obtain a simulated bone-implant specimen. By regulating the mould, it was possible to create the following simulated bone–implant contact groups: 3.3 mm (198.6 $mm^2$); 4.1 mm (198.8 $mm^2$); 4.8 mm (200.2 $mm^2$); 4.8 mm (231.7 $mm^2$); 4.8 mm (294.7 $mm^2$). Each group included 10 specimens. After resin setting, resonance frequency was measured. On the last group, measurements were repeated after establishing implant rotational mobility. One-way ANOVA tests with *post hoc* comparisons, a Pearson's correlation coefficient and a *t*-test for repeated measurements were used to evaluate statistically significant differences.

**Results:** Implants with different diameters but with the same amount of simulated osseointegration revealed no differences in resonance frequency. On the contrary, an increase of simulated bone–implant contact resulted in significantly higher resonance frequency. A clear direct linear correlation resulted between resonance frequency and simulated bone–implant contact. Furthermore, a significant difference resulted between resonance frequency measured before and after creation of rotational mobility.

**Conclusions:** Within the conditions of this study, the secondary stability was correlated with the simulated bone–implant contact. In addition, resonance frequency was able to discern between stable and rotation mobile implants.

Resonance Frequency Analysis is currently widely used to evaluate the stability of oral implants (Sennerby & Meredith 2008). Early studies showed how resonance frequency is influenced by the distance of the transducer from bone (Meredith et al. 1996, 1997a,b) and by the stiffness of the bone–implant interface (Meredith et al. 1996, 1997a,b; Friberg et al. 1999a,b). However, it is still not completely understood what factor is determining such stiffness. In fact, while many *in vitro* (Bardyn et al. 2009; Tabassum et al. 2010), animal (Ito et al. 2008; Su et al. 2009), cadaveric (Rozé et al. 2009) and *in vivo* (Miyamoto et al. 2005) studies demonstrated that at placement, such stiffness is strongly dependant on the thickness of the cortical bone, it is much less understood what is determining such stiffness for an integrated implant. The latter situation has been mainly investigated by trying to establish a correlation between histological parameters from animal (Schliephake et al. 2006; Strnad et al. 2008; Abrahamsson et al. 2009) or retrieved bone-implant human samples (Scarano et al. 2006; Degidi et al. 2010) and their resonance frequency. Because of the contradictory results obtained, it has been pointed out that histology, given its two dimensional nature, might be inaccurate in describing the interfacial stiffness (Schliephake et al. 2006). Closely related to secondary stability, it is the

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progressive loss thereof. Again, while some studies indicated that the technique might be useful to point out such condition (Friberg et al. 1999a; Glauser et al. 2004; Bornstein et al. 2009; Sennerby et al. 2012) and to possibly avoid taking action until safe secondary stability is re-established, others doubted this possibility (Huwiler et al. 2007). Hence, it appears useful to try to increase the insight into the determinants of secondary stability as measured by resonance frequency.

Another aspect that seems to justify further investigation is the influence of implant diameter on resonance frequency. In fact, implants with different diameters showed the same stability in clinical studies where resonance frequency was used to measure secondary stability (Bischof et al. 2004; Degidi et al. 2009; Han et al. 2010). Conversely, other clinical studies (Zix et al. 2005, 2008; Karl et al. 2008; Kessler-Liechti et al. 2008; Bornstein et al. 2009) showed that resonance frequency was directly related to implant diameter and that wider implants reached higher secondary stability. When specifically analysing with a numerical approach, in a finite element analysis study (Pattijn et al. 2006), the impact of diameter on implant secondary stability, resonance frequency proved sensitive to changes in stiffness at the bone–implant interface and implant diameter resulted to be a factor of influence on implant stability. Therefore, given the contradictory data available on the role of implant diameter in determining resonance frequency, further study seems justified. In particular, it would be interesting to investigate such a relation in bone of lower density where implant stability might be more difficult to achieve and maintain (Jaffin & Berman 1991).

An original approach to investigate secondary stability and the influence of different diameters as well as different percentages of interfacial contact would be through the use of a polyurethane foam. Such a bone-simulating resin, in its block form, has been already used to eliminate the confounding effect of real bone interspecimen variability when investigating primary stability (Bardyn et al. 2009; Tabassum et al. 2010). The polyurethane resin, in its pourable form, might be also used to simulate osseointegration and thus secondary stability.

The aim of this study was therefore to test the null hypothesis of no differences in resonance frequency for standardized percentages of simulated bone–implant contact around implants with different diameters. The experimental set-up was designed to resemble soft bone mechanical characteristics. In addition, it was evaluated if resonance frequency is able to detect a difference between stable and rotation mobile implants.

## Material and methods

To precisely obtain predetermined amounts of simulated bone–implant contact, a mould was fabricated. This mould, a metallic cylinder with a height of 25 mm and an internal diameter of 25 mm, was composed of two halves that could be separated (Fig. 1a). On one of its bases, an implant holder was constructed using an insertion carrier (Institut Straumann AG, Basel, Switzerland) embedded in a resin disk so that an implant could be reproducibly placed in the centre of the mould (Fig. 1a). The mould contained 15 standardized perforations, divided on four perpendicularly disposed rows. Three rows had four holes and one had three holes. The distance between the perforations in the same row was 1 mm. Each of these holes was threaded to house screws with a diameter of 2.2 mm. By varying the number of screws in contact with the implant, it was possible to vary the amount of implant surface available for contact with the bone-simulating resin (Fig. 1c).

The resin used to simulate bone–implant contact was a closed-cell rigid pourable polyurethane resin (US Composites, West Palm Beach, FL, USA) with a density of 0.26 gcc and a compressive strength of 4 MPa as indicated by the manufacturer. Such resin is supplied in two components, A and B, to be mixed in a 1 : 1 ratio, and it expands during setting. An equal amount of 1.2 ml of both components was therefore measured with graduated syringes, mechanically mixed at 1500 rpm for 20 s and poured into the mould containing the implant. As per manufacturer's instructions for optimal resin expansion, care was paid to pour the resin when the ambient temperature was higher than 26°C. Care was also paid to prepare the resin when ambient humidity was in a 30–40% range (Szivek 1999). In addition, an insulating wax paste (Partall #2, Rexco, GA, USA) was used to avoid resin sticking to the mould walls. According to the manufacturer, the resin is fully set after 20 min, and therefore, this time was respected before considering the specimens ready for testing.

Resonance frequency was measured with the Osstell Mentor equipment (Integration Diagnostic AB, Gothenburg, Sweden). The smartpegs (Integration Diagnostic AB) used were Type 41 for 3.3 mm and Type 42 for 4.1 and 4.8 mm implant diameters. The smartpegs

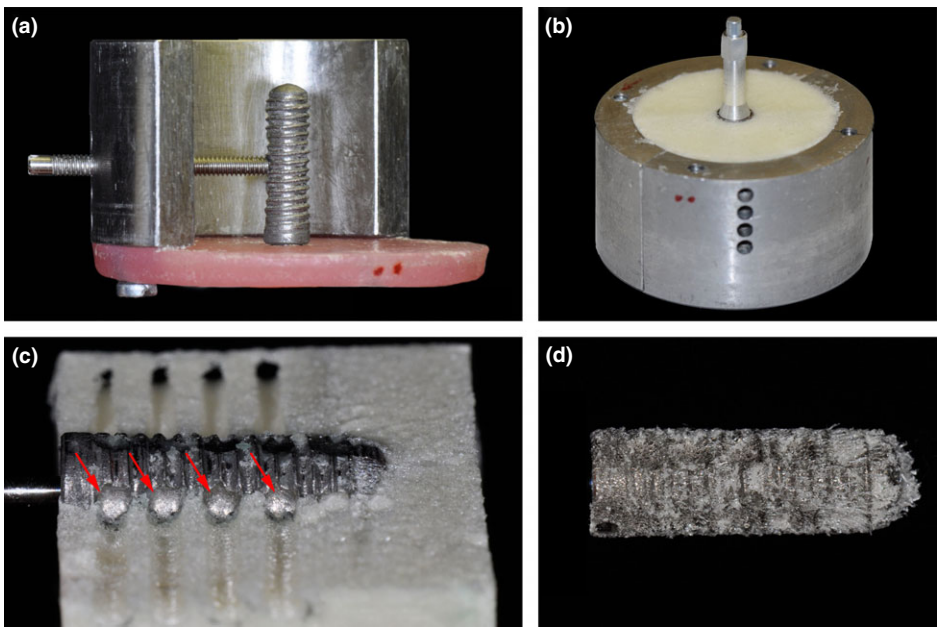


*Fig. 1.* (a) The empty opened mould with a screw in contact with the implant. The pink resin base,containing an implant carrier for reproducible implant positioning, is removed when the bone simulating resin is set. (b) The mould, containing a specimen consisting of simulated bone and the implant connected to the smartpeg and ready for resonance frequency measurement. (c) A specimen cut into halves to show the areas preserved from the resin contact thanks to the mould screws (arrows). The niches grinded onto the implant to eliminate the threads and to provide a uniform contact area with the screws are also visible. (d) An implant extracted from the resin after establishing rotational mobility. It is evident how the resin is still bonded to the implant and how the mobility is established because of a cohesive resin failure.

were mounted using the provided plastic driver. To avoid measuring the resonance frequency of the resin block itself instead of the interfacial one, the specimens were left inside the mould during measurements (Bardyn et al. 2009) (Fig. 1b). To ensure that the stability value was correctly identified, measurements were performed twice along the directions of the mould screws. The value used was the average of the most stable directional values obtained over all four measurements (Bardyn et al. 2009).

First of all, to predetermine the amount of surface available to simulate osseointegration, the surface of the implants to be used was defined. Three implants were used; they were all bone-level implants (SLA, Bone Level, Institut Straumann AG) with a diameter of 3.3, 4.1 and 4.8 mm and a 14 mm length. Each implant was photographed and then, using an image elaboration software (Photoshop, Adobe, San Jose, CA, USA), the outline of the implant profile was traced. Afterwards, by using a second software (ImageJ, http://rsb.info.nih.gov/ij), this outline was measured to obtain the length of the thread profile (Fig. 2). Finally, to calculate its surface and to bypass the difficulty of the thread presence, the implant was considered as a cylinder whose height and inferior base dimension were given from the previously taken measurements. It resulted that 3.3 implants had a surface of 198.6 $mm^2$, while 4.1 and 4.8 implants had a 249.2 $mm^2$ and a 294.7 $mm^2$ surface, respectively. In addition, it was calculated that each of the mould screws was diminishing the implant surface available for simulated osseointegration of 6.3 $mm^2$. In fact, to allow a flat surface contact between each mould screw and the implant, despite the presence of the implant threads, small niches were grinded on the implant in the area designated to contact the mould screw (Fig. 1c). Such modification was carried out with a bur with a diameter of 2 mm. The bur was deepened in the implant threads for half of its height, therefore producing a hemispherical niche whose surface was 6.3 $mm^2$, that is, half the surface of a sphere with a 2 mm diameter.

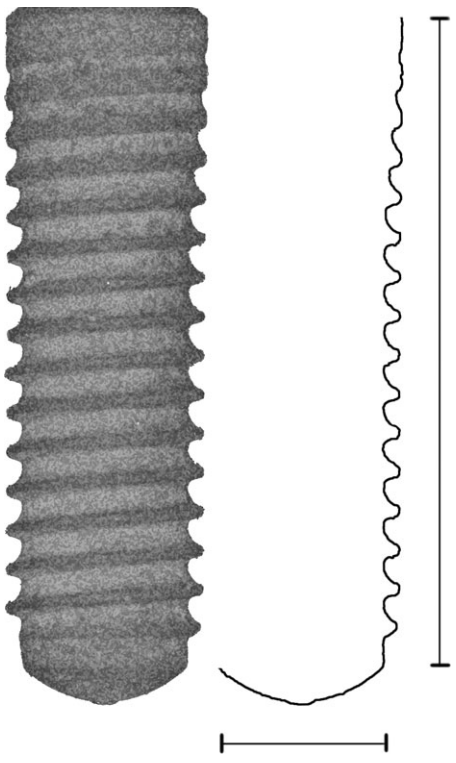

*Fig. 2.* Photography of one implant. The profile of the implant was traced and then measured to obtain the length of the base of the implant and its linear height. Such measurements were then used to calculate the surface of the implant side and its bottom areas that, after summation, determined the implant surface available for simulated osseointegration.

By adjusting the number of mould screws in contact with the implants, 5 groups were created with the twofold rationale of simulating the same amount of osseointegrated surface around implants of different diameters (3.3, 4.1 and 4.8 mm) and to simulate increasing osseointegrated surfaces around a 4.8 mm implant (Table 1). As such, when considering groups 1, 2 and 3, they had approximately the same simulated bone–implant contact at implants of different diameters. When considering groups 3, 4 and 5, they represented simulated bone–implant contact increases of approximately 10%, 20% and 30% at an implant with a diameter of 4.8 mm. Each group was composed of 10 specimens. To avoid stiffness asymmetries inside the simulated bone-implant specimens, the contacts of the mould screws were always distributed circumferentially around the implants. Because only three implants were used, the mould was opened after resonance frequency measurements, the implant was carefully cleaned from the resin and the inclusion process was repeated to create a new specimen.

Finally, after resonance frequency measurement, samples of group 5 were subjected to an inverse torque using a surgical ratchet (Institut Straumann AG). The aim was to create a cohesive failure into the resin to obtain a rotational mobility (Fig. 1d); however, care was paid to avoid lateral mobility. Resonance frequency measurement was then repeated on these modified specimens.

### Statistical analysis

After having tested the normality of data distribution (Kolmogorov-Smirnov test 0.714 $P = 0.68$) and the homogeneity of variances (Levene test 2.12 $P = 0.08$), an ANOVA test with Student-Newman-Keuls *post hoc* comparisons was calculated. To assess differences in resonance frequency for samples of group 5 before and after establishing rotational mobility, a *t*-test for repeated measurements was calculated. Finally, a Pearson's coefficient was computed to evaluate the correlation between resonance frequency and simulated bone–implant contact. The level of significance was set at $P < 0.05$. All data analyses were performed using statistical software (SPSS 19, IBM, Somers, NY, USA).

## Results

Resonance frequency results for each group are presented in table 1 and fig. 3. The ANOVA test was $F = 42.93$ ($P < 0.05$). *Post hoc* comparisons showed no significant differences in resonance frequency among the first three groups, representing implants with three different diameters but the same amount of simulated bone–implant contact. Conversely, when considering the last three groups featuring increasing amounts of simulated bone–implant contact, up to the full integration of group 5, significant differences resulted. Also, a significant direct linear correlation resulted between resonance frequency and simulated bone–implant contact ($r = 0.773$ $P < 0.05$) (fig. 4).

***Table 1.*** **Description of the experimental groups and respective mean ISQ**

| Group N = 10 | Implant. diameter. (mm) | Implant. surface. ($mm^2$) | Mould screws in contact | Amount of simulated osseointegration ($mm^2$) | Mean ISQ | SD | Median |
|---|---|---|---|---|---|---|---|
| 1 | 3.3 | 198.6 | – | 198.6 | $52.8^{ab}$ | 2.6 | 52.8 |
| 2 | 4.1 | 249.2 | 8 | 198.2 | $51.5^{cd}$ | 2.5 | 50.5 |
| 3 | 4.8 | 294.7 | 15 | 200.2 | $53.3^{ef}$ | 3 | 54.5 |
| 4 | 4.8 | 294.7 | 10 | 231.7 | $56.9^{aceg}$ | 2.2 | 57 |
| 5 | 4.8 | 294.7 | – | 294.7 | $64.2^{bdfgh}$ | 2.4 | 64.5 |
| 5 after rotational mobility | 4.8 | 294.7 | – | 294.7 | $47.1^{h}$ | 4.4 | 46.3 |

Equal superscript letters indicate a statistically significant difference $P < 0.05$. Statistical comparison for group 5 after rotation mobility was calculated only versus group 5.

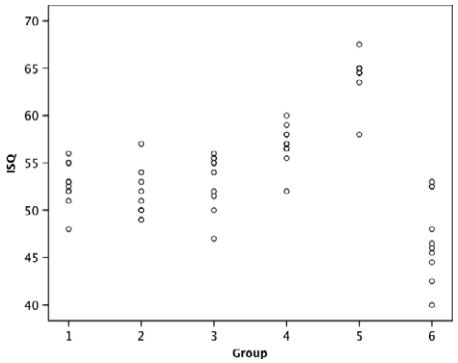


*Fig. 3.* Scatter plot of the data from all groups.

Finally, a significant difference ($t = 12.75$ $P < 0.05$) resulted for group 5 specimens between resonance frequencies measured before and after rotational mobility was established.

## Discussion

Resonance frequency is considered by some authors as an objective guide to assess osseointegration and therefore helpful to decrease implant failure risk (Sennerby & Meredith 2008). On the opposite, other authors questioned the quoted value of the technique (Aparicio et al. 2006), mostly because of the lack of data in support of its prognostic value to indicate developing implant instability. In any case, further research has been advocated to improve the clinical validation of this technique, whose merit and relevance might still be unrevealed (Koka 2006).

This study was started with the aim to investigate if resonance frequency is affected by variations in the quantity of simulated osseointegration around implant with different diameters. The present data showed that when implants with different diameters were in contact with the same amount of simulated osseointegrated interface, no significant differences in resonance frequency resulted.

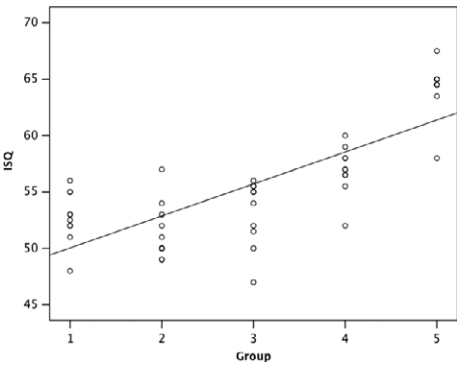


*Fig. 4.* Scatter plot of the data subjected to correlation analysis.

Conversely, when the simulated bone to implant contact was increased, a corresponding significant increase in resonance frequency was found. The tested null hypothesis is therefore accepted, as it appears that under the conditions of this study, resonance frequency is largely influenced by the amount of simulated bone–implant contact.

The experiment described was purposely set-up to study simulated secondary stability. In fact, while it has been shown that primary stability is mainly influenced by cortical bone thickness (Miyamoto et al. 2005; Rozé et al. 2009), less is known on secondary stability and its resulting stiffness as measured by resonance frequency. Theoretically, interfacial bone modelling and remodelling should increase the bone–implant contact and peri-implant bone density thus enhancing the immobility of the implant and its consequent interfacial stiffness. However, while implants retrieved from human bone after six months of healing showed a correlation with the histological bone to implant contact with resonance frequency taken before retrieval (Scarano et al. 2006), the same correlation could not be confirmed for implants retrieved after an early healing period (Degidi et al. 2010). Similar controversial data resulted from animal studies investigating the correlation between resonance frequency and histomorphometry. During healing, two studies failed to identify an association between significantly increasing peri-implant bone density or bone–implant contact and resonance frequency (Schliephake et al. 2006; Abrahamsson et al. 2009). On the contrary, proportionality was found in another study between bone–implant contact and resonance frequency changes during healing (Strnad et al. 2008). In any case, it has to be noted that for its two dimensional nature, histology might be not the best technique to characterize implant stiffness measured by resonance frequency (Schliephake et al. 2006). In addition, the variability that bone samples might display acts as a confounding factor when studying the correlation between bone–implant contact and resonance frequency (Bardyn et al. 2009). In fact, when resonance frequency was investigated with a numerical approach, an increasing degree of osseointegration was reflected by a trend of increasing resonance frequency (Natali et al. 2006). In this study, to obviate such shortcomings and to obtain standardized boundary conditions, the polyurethane foam approach was employed. This experimental set-up has been quite extensively applied in orthopaedics for testing the holding capacity of osteosynthesis screws (Szivek 1999). Previous studies in implant dentistry, using this resin in preformed blocks, investigated primary stability (Bardyn et al. 2009; Tabassum et al. 2010). However, the same resin can be used in a pourable form that, after setting, will adhere to the implant thus simulating osseointegration (Szivek 1999). In addition, the use of a mould allowed the control of the distribution of this resin onto the implant surface, to obtain reproducible specimens simulating different amounts of osseointegration. Unfortunately, it was not technically possible to simulate exactly the same amount of osseointegration for groups 1, 2 and 3; in fact, group 3 had a 2 $mm^2$ larger surface. However, this disparity did not produce significant differences in resonance frequency and, for the purposes of this study, groups 1, 2 and 3 were considered as equivalent with regard to the simulated surface of osseointegration. When using polyurethane resin, the bone–implant contact is influenced by the size of the cells in the resin (Szivek 1999). Because this is affected by pouring conditions, it is important to maintain the ratio between the components well controlled, to mix the components after mechanical stirring and to respect a working temperature (Szivek 1999). All this is necessary for optimal resin expansion and consistent mechanical conditions (Szivek 1999). Polyurethane resins are available in various densities that could simulate different bone densities. Accordingly, it would have been interesting to evaluate also the influence of different simulated bone mineralization on resonance frequency. However, this was not attempted in the present study because different resin densities also entail different foam cell size and ultimately produce a variation in the simulated bone–implant contact. Therefore, the density chosen for this study is one that simulates soft bone characteristics. This aspect could also account for the somewhat low ISQ recorded for specimens simulating full osseointegration, as for instance, the mean ISQ value of 52 for a simulated full integration of a 3.3 mm implant and 56.9 ISQ for a 4.8 one. In any case, there are no other studies in the literature assessing secondary stability of bone level type Straumann implants. On the contrary, it was observed with regard to 120 osseointegrated Straumann tissue level type implants placed in maxillary bone of supposedly lower density, that the mean ISQ was 52.5 (Zix et al. 2005). With regard to the present study, it seems more of importance to evaluate the meaning of the changes of ISQ reflecting variations in osseointegration rates, than their absolute values, and in particular, the direct correlation between simulated

bone–implant contact and resonance frequency. When considering that the mean ISQ values recorded here were obtained from simulated bone–implant interface specimens, it seems nevertheless prudent to underscore that they should not be directly translated to the clinical situation where bone anatomy is different. In particular, one difference would be the lack of a distinct cortical layer of the experimental specimens. However, because the study was meant to investigate soft bone conditions, where a distinct cortical layer is often lacking (Ulm et al. 1999), the latter does not appear a serious limitation to the validity of the correlation between simulated bone–implant contact and associated resonance frequency. Moreover, despite the mould allowed for reproducible resin architecture at the implant interface, the purposely created voids deviate from the architecture of osseointegration. However, this seemed compatible with the aim of the study to assess differences in resonance frequency for predetermined and standardized degrees of secondary stability. A further limitation of the study is that because only one implant per diameter was used, they were repeatedly scraped from the resin thus smoothening their surface roughness. Consequently, although implant roughness contribution to implant stability is controversial (Sennerby & Meredith 2008), it is unlikely that the present experimental set-up took it into account. Despite the limitations of this model might render difficult to extrapolate a clinical relevance from these experimental data, some considerations and comparison with the clinical literature might be attempted.

When considering the stability of implants with different diameters, contradictory results are found in the literature. Some studies stated that larger-diameter implants of equal length achieve superior secondary stability (Zix et al. 2005, 2008; Karl et al. 2008; Kessler-Liechti et al. 2008; Bornstein et al. 2009), while others could not demonstrate such a difference (Bischof et al. 2004; Degidi et al. 2009; Han et al. 2010). In the light of the present data the results of the literature could be explained by noting that when their osseointegrated surface was reduced, 4.8 mm diameter implants revealed the same level of stability as 3.3 mm and 4.1 mm implants. As a consequence, it could be speculated that the degree of looseness of the bone architecture might play a role in determining the level of secondary stability. In any case, given the same architecture, wider-diameter implants would result to be more stable than thinner ones because they can potentially engage a larger amount of osseointegrated interface. Therefore, considered the reported high inter- and intraindividual variability in trabecular connectivity (Ulm et al. 1999), and in light of the present experimental results, the seemingly contradictory relations between implant diameter and secondary stability found in different studies could be interpreted as due to the clinical variability of trabecular architecture determining the bone–implant contact. It is likely that a correlation between diameter and secondary stability might be established only in larger samples that could compensate for the above-mentioned variability. As a matter of fact, the majority of the studies demonstrating the correlation between implant diameter and resonance frequency response included larger sample sizes than the studies where it did not appear. Finally, the present data also seem to suggest that resonance frequency is of little use in comparing the stability of different implants while, as previously stated (Sennerby & Meredith 2008), it would be better suited to follow changes of stability over time.

A last point to consider was the difference in resonance frequency found for group 5 specimens subjected to repeated measurements before and after establishing implant rotation mobility. This was a condition where the simulated osseointegration was disrupted in form of a cohesive failure of the resin resulting in rotational, but no lateral mobility. As such, it could simulate the state of an implant whose peri-implant bone, having not yet completed its remodelling, features a suboptimal load bearing ability. Such a state has been described as a cause of failing implant, where the implant in its healing phase, although clinically stable, is progressively losing its stability, for instance because of uncontrolled loading from a poorly relieved denture (Friberg et al. 1999a; Sennerby et al. 2012). Similarly, it has been described that an implant during its healing process can "spin" if subjected to torque (Valderrama et al. 2007; Bornstein et al. 2009). The safest clinical conduct in such a situation would be to take adequate rescuing action (loading postponement, reduction or removal). In the present study, implants whose simulated bone interface was purposely damaged, causing rotation mobility, systematically showed lower ISQ values than their integrated counterparts. Therefore, resonance frequency might be useful to identify this condition before, for instance, in the attempt to tighten an abutment, applying any torque that could further disturb a healing process still in act. However, it should be also pointed out that, at present, any normal range for osseointegrated Straumann bone level implants is missing, and therefore, further studies are necessary to establish a guideline ISQ that, when not reached, should elicit clinician's attention because he is potentially facing a spinning implant.

In conclusion, under the present experimental conditions, resonance frequency is directly related to simulated bone–implant contact and, as a result, larger implants achieve higher secondary stability than narrower ones. Resonance frequency could also be of help in identifying implants that, when subjected to a torque force, will show spinning.

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