

Accuracy of Direct and Digital Indirect Orthodontic Bonding Using 3D Printed Transfer Trays: An Ex Vivo Study on Fresh Human Cadavers

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Abstract: This ex vivo study compared the accuracy of direct and indirect bonding techniques using 3D-printed transfer trays in fresh human cadavers. Five fresh human cadavers were selected to simulate clinical conditions, including the presence of enamel and surrounding soft tissues. Intraoral scans were obtained to generate digital models, and bracket positioning was performed virtually using OrthoAnalyzer software. Roth .022" Mini Master brackets were virtually placed on upper and lower arches, followed by the fabrication of 3D-printed transfer trays for indirect bonding. Each cadaver underwent both bonding techniques (direct and indirect), followed by intraoral scanning. Digital models were superimposed to assess differences in bracket positioning in the vertical (occluso-gingival) and horizontal (mesio-distal) planes. A total of 58 teeth were analyzed, the greatest deviation between the techniques was observed in the vertical plane, particularly in the maxillary arch, with a mean difference of 1.03 ± 0.36 mm ($p = 0.008$). Horizontal deviations were less pronounced and statistically non-significant (0.73 ± 0.43 mm; $p = 0.14$). The smallest differences were 0.26 ± 0.11 mm (vertical) and 0.19 ± 0.13 mm (horizontal). Indirect bonding with 3D-printed trays showed superior accuracy in bracket positioning compared to direct bonding, especially in the horizontal plane. This technique may offer clinically relevant advantages in enhancing bonding precision in orthodontics.

Keywords: Orthodontic Brackets; Dental Bonding; Dimensional Measurement Accuracy; Cadaver; Models, Dental.



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1. Introduction

The accuracy of bracket positioning during orthodontic bonding is one of the most accurate bracket positioning is a critical factor in achieving optimal biomechanics during fixed orthodontic treatment. Misplacement can lead to undesirable tooth movements, extended treatment time, unnecessary repositioning, incorporation of compensatory wire bends, and suboptimal functional and aesthetic outcomes [1, 2]. Direct bonding remains the most widely used technique due to its simplicity and reduced chairside steps, as it does not require laboratory processing. However, it is inherently more prone to operator-dependent errors [3, 4]. In contrast, indirect bonding was introduced to improve bracket placement precision through laboratory-guided procedures, offering enhanced control and reproducibility [5, 6]. Early

versions of this technique, however, were limited by impractical workflows and inconsistent clinical outcomes [7].

The advent of digital orthodontics, particularly CAD/CAM systems and 3D-printed transfer trays, has significantly improved the feasibility and accuracy of indirect bonding [8, 9]. These systems allow for individualized virtual bracket positioning, predictable tray fabrication, and integration with diagnostic tools such as CBCT, ultimately reducing chairside time and improving efficiency [3, 10]. Despite these technological advances, clinical evidence remains inconclusive regarding the superiority of indirect over direct bonding, especially in settings where factors such as saliva, soft tissue interference, and intraoral movement may affect outcomes [11, 12]. In vitro studies using typodonts or resin models do not fully replicate the complexity of the human oral environment, while in vivo studies raise ethical concerns related to repeated bonding and debonding procedures.

To address this limitation, ex vivo models using fresh human cadavers offer a biologically realistic and ethically viable alternative. These models provide enamel surfaces, soft tissue architecture, and intraoral access conditions that more accurately mimic the clinical setting [13-15]. However, no previous ex vivo study using fresh human cadavers has directly compared direct and digital indirect bonding under simulated clinical conditions. Therefore, the present study aimed to evaluate and compare the accuracy of direct and digitally guided indirect bonding techniques using 3D-printed transfer trays in fresh human cadavers.

2. Materials and Methods

Five fresh human cadavers were selected according to the following inclusion criteria: presence of complete upper and lower anterior segments (central and lateral incisors, canines) and at least one premolar per quadrant, with teeth free from fractures, extensive caries, or restorations that could interfere with bracket positioning. Cadavers with missing teeth, severe crowding, or dental anomalies were excluded. At the end, the sample comprised a total of 58 teeth, divided among 5 cadavers. Intraoral scans of the cadavers' dental arches were obtained using a Trios scanner (3Shape, Copenhagen, Denmark). The scans were processed in OrthoAnalyzer software (3Shape, Copenhagen, Denmark) to generate three-dimensional (3D) digital models. Each cadaver underwent two bonding protocols: indirect bonding and direct bonding. On opposite quadrants, alternating sides between specimens to minimize positional bias.

Virtual bracket placement was performed by a calibrated orthodontist following the software's stepwise protocol, including tooth segmentation, contact point identification, and long axis determination Roth .022" Mini Master brackets (American Orthodontics, Wisconsin, USA) were selected from the software library and positioned at the center of the facial axis of the clinical crown of each tooth. Once finalized, the virtual setups were exported for manufacturing of 3D-printed indirect bonding trays by the Compass 3D laboratory (Belo Horizonte, Brazil).

Direct bonding followed a conventional clinical workflow (Figures 1A to 1B). Initially, an adhesive resin (Transbond XT primer; 3M® Unitek, Minnesota, USA) was applied to the bracket base, brackets were manually positioned at the center of the facial axis of the clinical crown of each tooth, and excess adhesive was removed. Indirect bonding involved adapting the 3D-printed transfer tray over the arch with brackets preloaded with adhesive (Figure 1C). For both methods, brackets were photopolymerized for 3 seconds each on the buccal and occlusal surfaces using a Valo LED curing unit (Ultradent®, Ribeirão Preto, Brazil) (Figure 1D). After each protocol, brackets were removed and the enamel polished (Figure 1E).

Following each bonding protocol, the dental arches were rescanned with the Trios Color intraoral scanner (3Shape, Copenhagen, Denmark) (Figure 2A to 2D). The digital models of direct and indirect bonding for each cadaver were superimposed

using the “Surface 3-point” registration method in OrthoAnalyzer software, selecting identical stable anatomical reference points (Figure 3A to 3B).

Figure 1: Clinical orthodontic procedures on a fresh human cadaver. A. Application of adhesive resin to the bracket base (direct bonding). B. Placement of orthodontic brackets on the tooth (direct bonding). C. Insertion of a 3D-printed transfer tray (indirect bonding). D. Photopolymerization of brackets (direct and indirect bonding). E. Polishing after bracket removal (direct and indirect bonding).

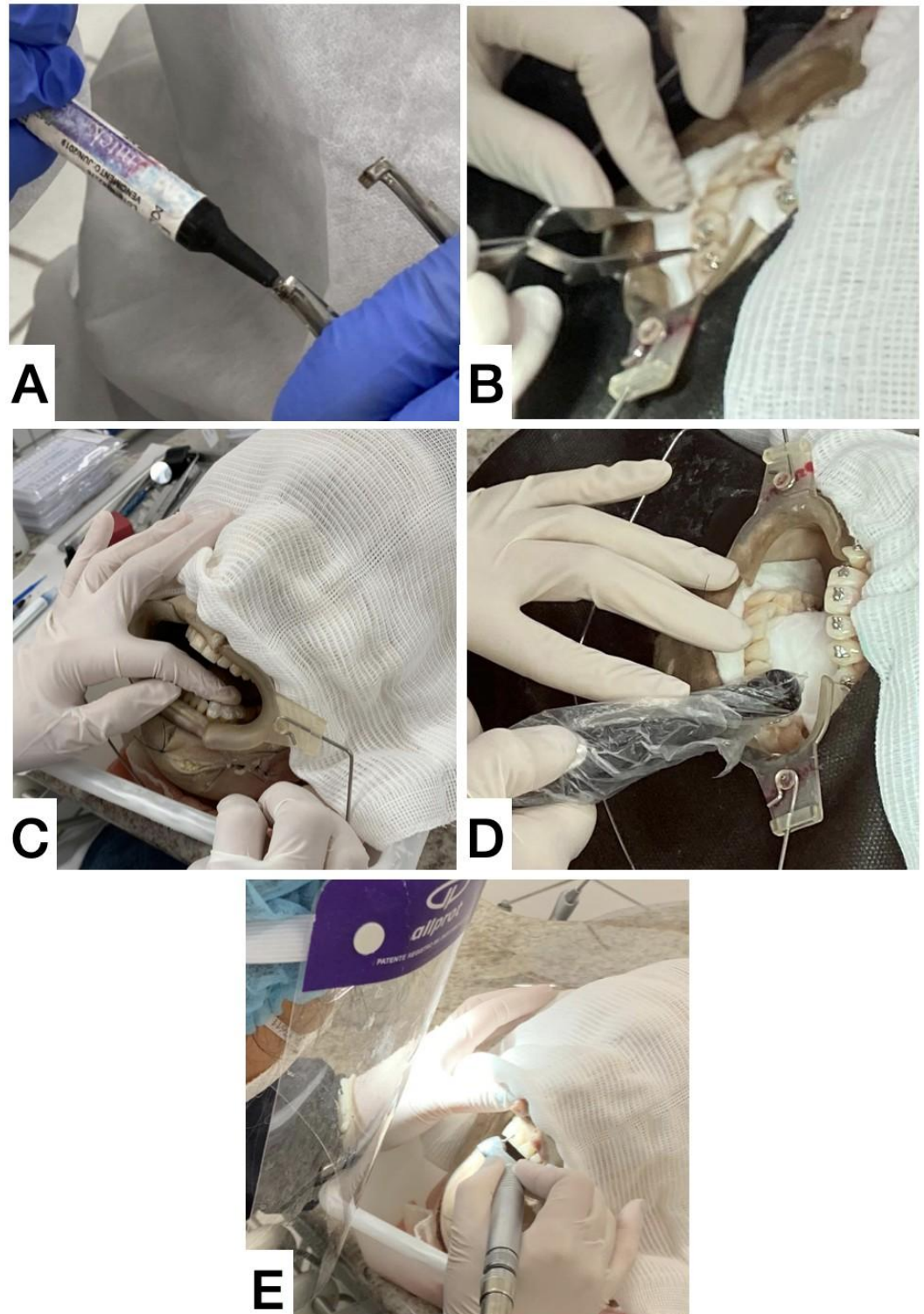


Figure 2: Digital intraoral scanning procedure. A. Intraoral scanning of a fresh human cadaver. B. Operator capturing the dental arch using a 3Shape intraoral scanner connected to CAD software. C. Computer screen displaying the processing of the 3D scan. D. Final 3D scan. E. Example of a 3D digital model with orthodontic brackets. F. Additional example of a 3D digital model with orthodontic brackets.

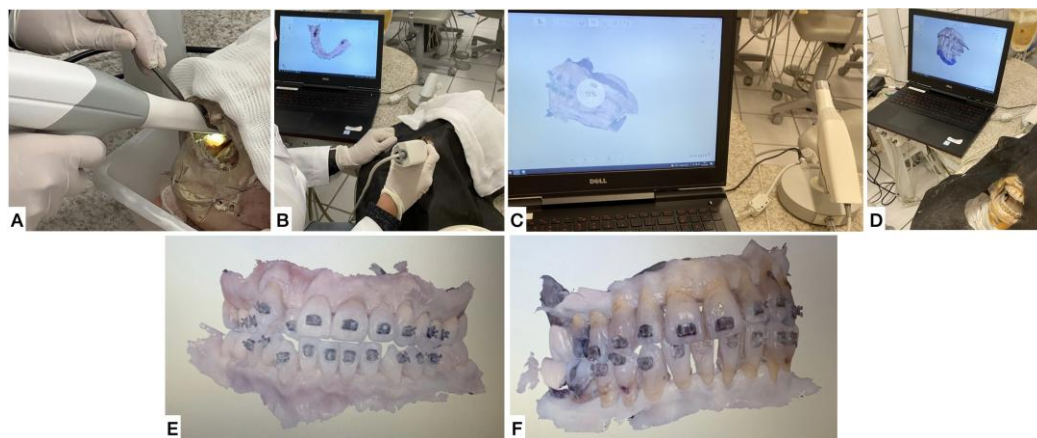
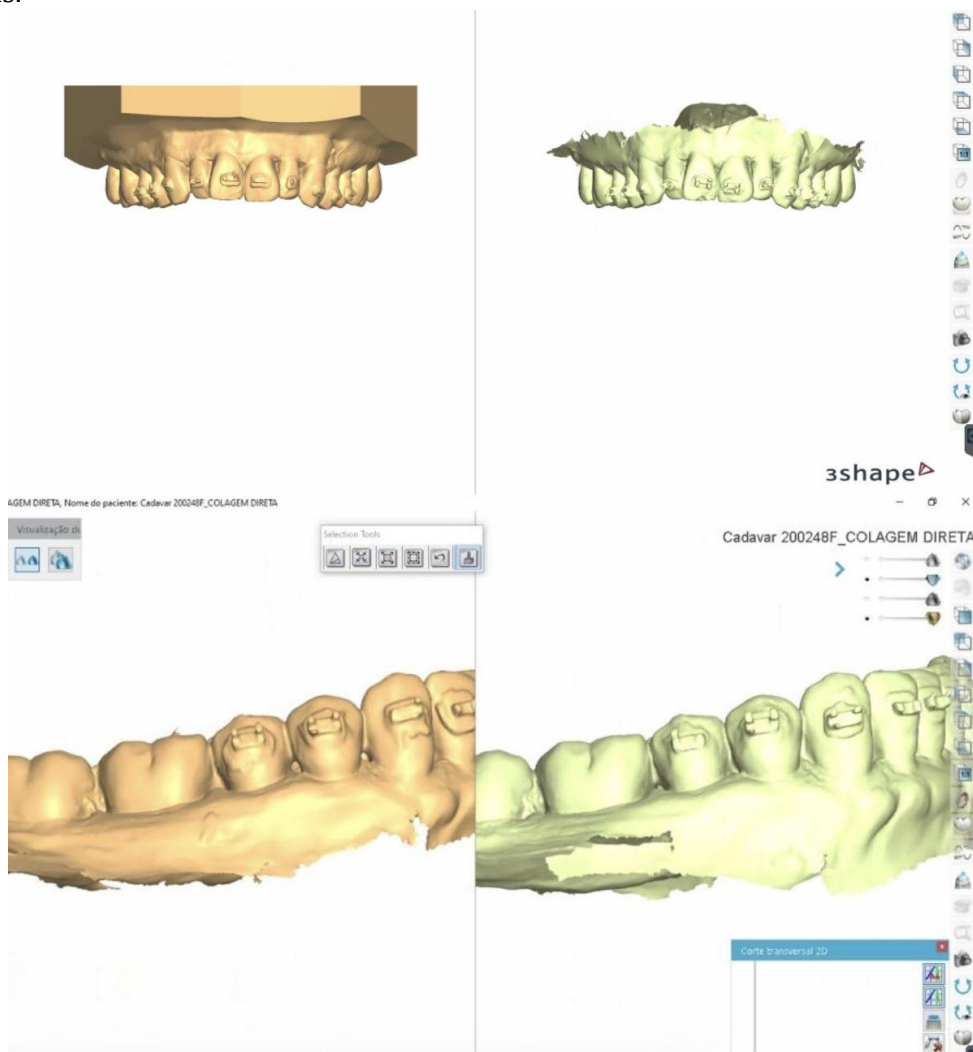


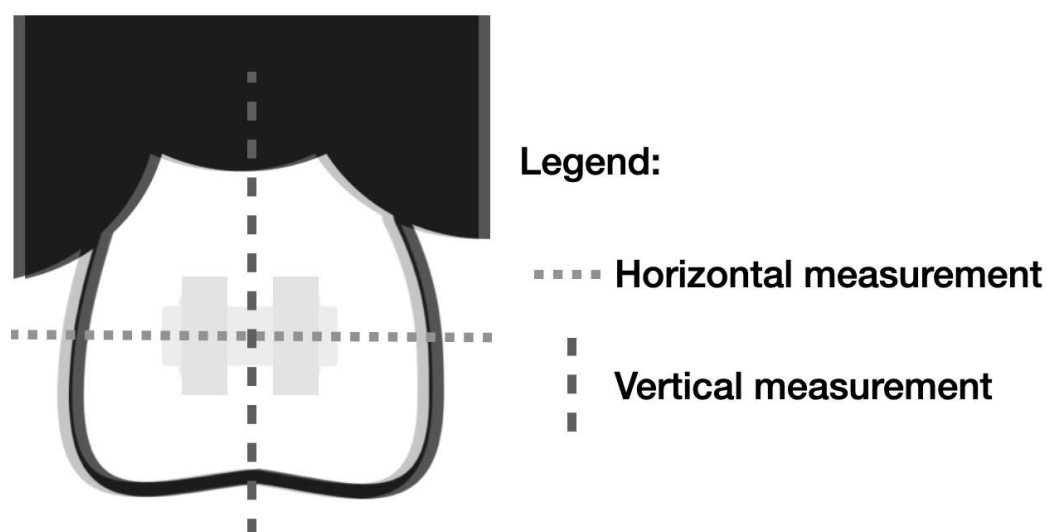
Figure 3: Comparison of 3D dental models using 3Shape software. A. Frontal view of maxillary arch models. B. Lateral view of mandibular arch models, with yellow representing direct bonding and green representing indirect bonding, for accurate analysis.



Since no tooth movement occurred in this ex vivo experimental setting, stable dental anatomical structures were selected as reference points for superimposition. Three points were chosen for each arch to perform the 3-point surface registration: in the upper arch, one point on the buccal cusp ridge of each second premolar and one on the mesial incisal edge of the right central incisor; in the lower arch, one point on the buccal cusp ridge of each first premolar and one on the cusp tip of the right canine. This approach, previously described in the literature [8], ensures reproducibility and minimizes potential variability related to soft tissues or tray seating. Furthermore, to reduce artefacts, all scans were obtained using the same intraoral scanner and standardized protocol both before and after bonding. The registration and subsequent measurements were confined to dental and palatal hard-tissue surfaces, thereby eliminating any influence of intraoral soft-tissue distortion, which is inherently lower in ex vivo cadaveric models compared to in vivo conditions.

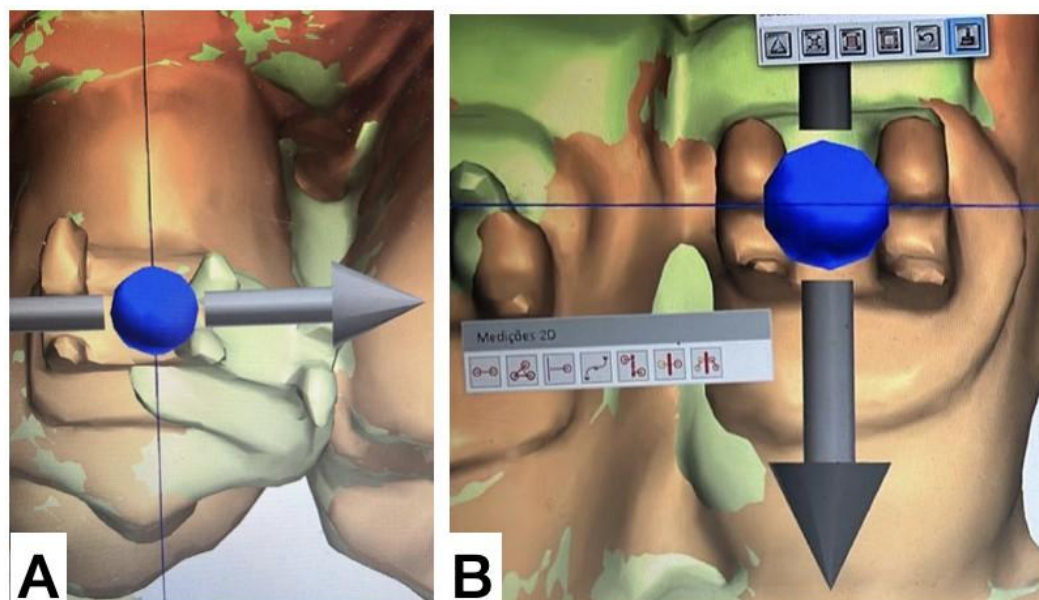
Bracket positioning differences were evaluated in the vertical (occluso-gingival) and horizontal (mesiodistal) planes. Measurement axes were defined according to the schematic in Figure 4, with vertical and horizontal reference lines intersecting at the center of the facial axis of the clinical crown of each tooth. Superimposed models were visualized in the software's measurement mode (Figure 5), enabling precise quantification of deviations between bonding techniques. All measurements were performed by a single calibrated examiner, with planes and points defined using the "2D Measurements" tool in sagittal, axial, and coronal sections. The bracket base served as the reference landmark in all analyses.

Figure 4: Schematic representation of horizontal and vertical measurements for bracket positioning on a tooth surface. The dashed vertical line represents the vertical measurement axis, and the dashed horizontal line represents the horizontal measurement axis.



Statistical analysis was performed using SPSS Statistics, version 28.0 (IBM Corp., Armonk, NY, USA, 2021). The Shapiro–Wilk test was applied to assess normality, and the data were found to follow a normal distribution. Descriptive analysis was conducted using mean and standard deviation (SD). To evaluate mean deviations between groups, a one-sample Student's t-test was applied. A significance level of 5% was adopted, and clinical differences of up to 0.5 mm were considered detectable [16]. For a more comprehensive analysis, histogram plots were constructed. No sample size calculation was performed, as this was a pilot study.

Figure 5: Superimposition of 3D dental models illustrating bracket positioning analysis. The images show horizontal (left) and vertical (right) measurement tools within the 3Shape software, used to assess the spatial alignment between the superimposed models.



While the minimal detectable change reflects the sensitivity of our measurement system, the threshold for clinical acceptability is based on established orthodontic standards. A deviation of up to 0.5 mm has been adopted as a clinically acceptable limit for linear bracket positioning errors, in accordance with the Objective Grading System of the American Board of Orthodontics (ABO) and supported by recent studies assessing bonding accuracy [16].

3. Results

A total of 58 teeth divided among 5 cadavers are detailed in the descriptive data analysis, the mean differences between the bracket positions in direct and indirect bonding, both in the maxilla and mandible, in vertical and horizontal planes, were observed. The mean differences in millimeters (mm), standard deviations, and p-values are presented in Tables 1 and 2.

Table 1: Mean and standard deviation (in millimeters) of the differences between direct and indirect bonding, and one-sample Student's t-test for the maxilla.

Tooth	Mean	Vertical		Horizontal		
		SD	p value	Mean	SD	p value
11	1.03	0.36	0.008*	0.38	0.23	0.84
12	0.82	0.32	0.02*	0.25	0.12	0.9
13	0.67	0.3	0.17	0.54	0.68	0.44
14	0.66	0.65	0.28	0.35	0.34	0.82
15	0.79	0.53	0.14	0.4	0.26	0.77
21	0.97	0.29	0.006*	0.19	0.13	0.96
22	0.74	0.73	0.22	0.52	0.29	0.41

23	0.40	0.24	0.81	0.61	0.72	0.35
24	0.93	0.47	0.03*	0.35	0.26	0.88
25	0.26	0.11	0.9	0.31	0.08	0.9

Legend.: SD. standard deviation. (*): $p \leq 0.05$. Mean deviations of 0.5 mm were considered for the statistical test.

Table 2: Mean and standard deviation (in millimeters) of the differences between direct and indirect bonding, and one-sample Student's t-test for the mandible.

Tooth	Vertical			Horizontal		
	Mean	SD	p value	Mean	SD	p value
31	0.36	0.2	0.92	0.37	0.15	0.93
32	0.52	0.43	0.45	0.35	0.19	0.9
33	0.55	0.36	0.35	0.57	0.54	0.37
34	0.72	0.72	0.24	0.62	0.52	0.29
35	0.65	0.35	0.19	0.73	0.43	0.14
41	0.54	0.32	0.36	0.19	0.09	0,9
42	0.32	0.19	0.9	0.28	0.07	0,83
43	0.63	0.53	0.27	0.49	0.3	0.5
44	0.72	0.37	0.1	0.31	0.15	0.9
45	0.74	0.3	0.06	0.36	0.15	0.82

Legend.: SD. standard deviation. (*): $p \leq 0.05$. Mean deviations of 0.5 mm were considered for the statistical test.

In the maxilla, the largest mean discrepancy in bracket positioning between direct and indirect bonding was 1.03 ± 0.36 mm ($p = 0.008$) in the vertical (occluso-gingival) plane of tooth #11. All statistically significant vertical differences were found in the maxillary teeth, specifically tooth #12 (0.82 ± 0.32 mm, $p = 0.02$), tooth #21 (0.97 ± 0.29 mm, $p = 0.006$), and tooth #24 (0.93 ± 0.47 mm, $p = 0.03$), each exceeding the accepted mean deviation threshold of 0.5 mm. The smallest vertical mean value in the maxilla was 0.26 ± 0.11 mm ($p = 0.90$) in tooth #25. No statistically significant differences were observed in the horizontal plane for any maxillary tooth, with the largest horizontal mean of 0.61 ± 0.72 mm ($p = 0.35$) found in tooth #23 and the smallest (0.19 ± 0.13 mm, $p = 0.96$) in tooth #21.

In the mandible, no vertical or horizontal deviations reached statistical significance. The largest vertical mean was 0.74 ± 0.30 mm ($p = 0.06$) in tooth #45, while the smallest was 0.32 ± 0.19 mm ($p = 0.90$) in tooth #42. In the horizontal plane, the highest mean was 0.73 ± 0.43 mm ($p = 0.14$) in tooth #35, and the smallest values were 0.19 ± 0.09 mm ($p = 0.90$) for tooth #41 and 0.28 ± 0.07 mm ($p = 0.83$) for tooth #42. Overall, the most significant differences were concentrated in the vertical axis of the maxilla, particularly in the central and lateral incisors and first premolars, as illustrated in the histograms in Figures 6 and 7.

Figure 6: Differences between direct and indirect bonding in the maxilla along the vertical and horizontal axes.

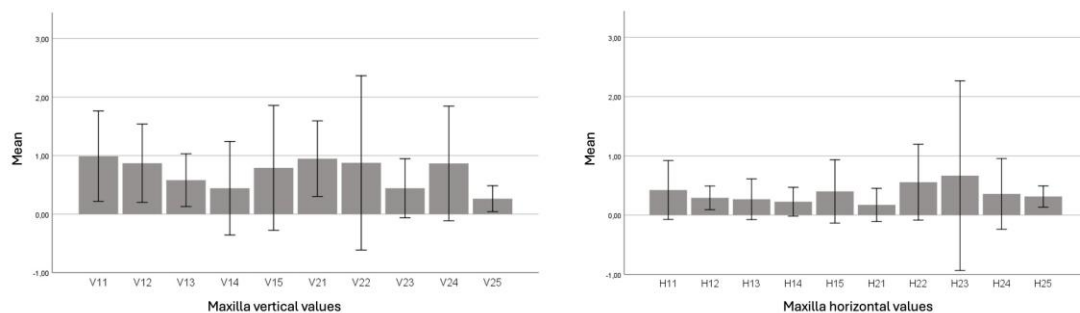
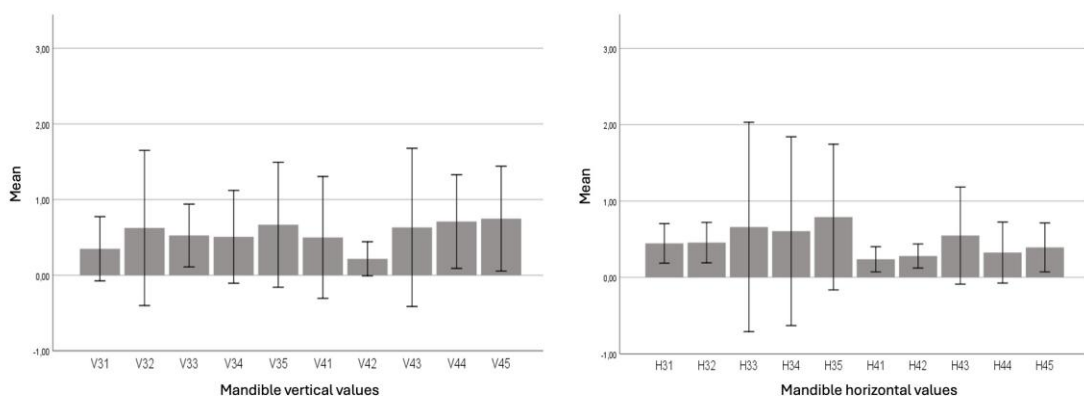


Figure 7: Differences between direct and indirect bonding in the mandible along the vertical and horizontal axes.



4. Discussion

This ex vivo study demonstrated that indirect bonding with 3D-printed transfer trays achieves superior accuracy in bracket placement compared to conventional direct bonding, particularly in the horizontal (mesio-distal) plane. Deviations greater than the clinically acceptable threshold of 0.5 mm were mainly observed in central and lateral incisors and first premolars, while horizontal discrepancies were smaller and not statistically significant. These findings indicate that the main clinical advantage of the digital indirect workflow lies in controlling horizontal positioning.

Our results are consistent with previous studies reporting improved precision with CAD/CAM-assisted indirect bonding [9,16,17]. As von Glasenapp et al. [9], we found that deviations were more pronounced vertically, likely due to the anatomical complexity of the maxilla and the influence of tray adaptation. While deviations below 0.5 mm may have limited clinical impact [17,18], the present data show that digital indirect bonding more consistently remains within or close to this threshold, reinforcing its value in esthetically critical regions and complex malocclusions. Conversely, earlier studies on conventional indirect bonding without digital support described variable accuracy and inefficient workflows [7], underscoring the role of 3D printing in overcoming such limitations.

This study has limitations such as the relatively small sample size and absence of longitudinal follow-up prevent conclusions about clinical outcomes such as bond failure [20,21] or long-term treatment efficiency [22,23]. All procedures were performed by a single operator, which reduces variability but may introduce bias. Furthermore, although cadaveric models provide greater clinical realism than typodonts

[24], they cannot fully replicate intraoral conditions such as saliva, thermal cycling, or patient movement.

Other additional factors influencing accuracy were noted by Pottier et al. [25] and Jungbauer et al. [26], authors emphasized the importance of tray design and manufacturing materials, while Li et al. [27] suggested that clinical experience may play a lesser role when digital transfer devices are employed. Nichols et al. [28] and Sabbagh et al. [29] also confirmed the reproducibility and reliability of indirect bonding protocols, supporting their clinical applicability. Furthermore, Xue et al. [30] and Zhang et al. [31] demonstrated that CAD/CAM-based trays and multilayer guide plates enhance bracket positioning accuracy.

From a biomechanical perspective, the improved accuracy of indirect bonding can be attributed to the guided positioning provided by transfer trays [6,9], which minimizes errors in determining the facial axis of the clinical crown and bracket height [1,11,16]. In addition, tray design helps reduce the effects of soft tissue interference [1] and limited visual access, challenges that are particularly relevant in posterior and maxillary regions [24]. In contrast, direct bonding remains vulnerable to vertical misplacement, even in the hands of experienced clinicians, due to anatomical variability, working angles, and lighting conditions [27]. By using fresh cadavers, our study reproduced these challenges under near-clinical conditions, offering results that may be more representative than those obtained with typodonts.

Clinically, enhanced horizontal accuracy with 3D-printed indirect bonding may reduce the need for bracket repositioning, minimize compensatory wire bends, and shorten overall treatment time. Even small deviations can compromise shear bond strength [20,32] and long-term stability [19,21,33], highlighting the importance of precise bonding protocols. Improved precision in esthetically demanding regions also has the potential to optimize smile outcomes and increase patient satisfaction, while the reproducibility of digital setups facilitates interdisciplinary communication and treatment planning [22,23,30].

Future studies should include a larger number of specimens to ensure adequate statistical power and external validity. Sample-size estimations indicated that as few as 3–7 paired teeth may be sufficient to detect clinically relevant differences of 0.5 mm under ideal assumptions. However, because teeth within the same cadaver are clustered and not fully independent, we recommend that future definitive trials include at least 10–12 cadavers, each contributing multiple teeth. This design would allow robust modeling of within-cadaver correlation, provide greater generalizability, and strengthen the evidence regarding the accuracy of direct and indirect digital bonding protocols.

Finally, the use of cadaveric ex vivo models provide a biologically realistic platform that bridges the gap between laboratory studies and in vivo clinical trials. As demonstrated in literature [34], such models allow for near-clinical testing of devices without ethical concerns. Our study extends this rationale to bracket placement accuracy and may serve as a foundation for future randomized clinical trials.

5. Conclusion

Indirect bonding with 3D-printed trays showed superior accuracy in bracket positioning compared to direct bonding, especially in the horizontal plane. This technique may offer clinically relevant advantages in enhancing bonding precision in orthodontics.

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Research Ethics Committee Approval: Data collection for this study began after approval from the Research Ethics Committee of the Pedro Ernesto University Hospital (CEP/HUPE) of the State University of Rio de Janeiro (UERJ) (Approval No. approval number 6.996.981 (CAAE: 80185124.2.0000.5259).

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Conflicts of Interest: None.

Supplementary Materials: None.

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