



THE EFFECT OF EQUILBRATING MOUNTED DENTAL STONE CASTS ON THE OCCLUSAL HARMONY OF CAST METAL COMPLETE CROWNS

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Statement of problem. Fabrication of indirect complete crowns that are in occlusal harmony upon insertion remains a problem in restorative dentistry, and dental stone cast expansion may play a role.

Purpose. This 3-part investigation compared occlusal contacts in a simulated patient with a harmonious occlusion with centric occlusion equal to maximum intercuspation (CO=MI) and an inharmonious occlusion (CO≠MI) with mounted stone casts, and compared the occlusal contacts after fabrication of a posterior complete crown fabricated on equilibrated and nonequilibrated dental stone casts.

Material and methods. A dentoform mounted in a semi-adjustable articulator served as the simulated patient and control. In part 1, a single set of maxillary and mandibular ADA type IV and V derived dental stone casts were fabricated and mounted (CO≠MI), and occlusal contacts/near contacts were compared. In part 2, 10 type IV and 10 type V cast mountings (CO=MI) were compared. In part 3, 10 type IV cast mountings were fabricated for adjustment (experimental) and 10 for no adjustment (control). A mandibular ADA type IV gold alloy complete crown was fabricated and adjusted on each set of casts and then returned to the dentoform. Vinyl polysiloxane interocclusal records of all mountings were scanned for optical density, and contacts were used to quantify occlusal contacts as exhibiting contact or near contact. Data were analyzed with Kruskal-Wallis ANOVA and Mann-Whitney U tests ($\alpha=.05$).

Results. The cast adjustment protocol (intervention) was successful in eliminating the majority of the occlusal disharmony in the casts believed to be caused by the effects of stone expansion. Actual and near contact areas for cast mountings of the equilibrated simulated patient were significantly different from those of the simulated patient ($P<.001$; $P=.001$, respectively). Actual and near contact areas for inserted crowns fabricated from adjusted casts were significantly different from those of the simulated patient ($P<.001$; $P=.007$, respectively), but actual contact areas were not different from those of the simulated patient with no crown inserted.

Conclusions. In this study, occlusal contacts of a simulated patient (dentoform) could not be accurately replicated with mounted dental stone casts. A cast adjustment procedure can aid in fabrication of a crown with a more accurate occlusion. (J Prosthet Dent 2010;104:122-132)

CLINICAL IMPLICATIONS

A cast adjustment technique can be implemented in the laboratory phase of indirect crown fabrication to minimize necessary occlusal adjustment upon restoration insertion.

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Fabrication of indirect complete crowns that are in occlusal harmony upon insertion is a problem that remains to be solved. Even with perfect execution of all required techniques and pattern generations, inevitable errors are present in definitive impressions, definitive casts, and mountings, as well as in the investment and casting processes. Impression trays,¹⁻⁸ impression materials,⁷ and disinfection practices⁹⁻¹¹ are all factors that may significantly affect the accuracy of the resulting dental casts.

Expansion of gypsum products used to fabricate dental casts has long been suspected of causing inaccuracies.^{12,13} The American Dental Association's Specification No. 25 for dental gypsum products stipulates that final setting expansion measurements should be made 2 hours after mixing.¹⁴ However, it is known that ADA type IV and V dental stones show delayed linear expansion continuing for 72-120 hours.¹⁵ Expansion of different brands of dental stone with equivalent ADA classifications may be different.¹⁶ Additionally, degradation of surface quality on dental casts has been reported with some impression material/gypsum combinations.¹⁷ Müller et al¹⁸⁻²⁰ found that different interocclusal recording media were less accurate when measured on duplicated casts, suggesting that the specific accuracy of the recording material is influenced by the derived casts.

Another critical factor in the fabrication process for indirect restorations is the proper articulation of definitive casts, which primarily affects the occlusal surface. Studies of various interocclusal recording materials and techniques,²¹⁻²⁵ including no-record indirect hand articulation of the casts^{23,26,27} and quadrant style, dual-arch impression techniques,^{28,29} have provided insight. Nonetheless, regardless of how precisely cast articulation and mounting procedures are performed, it is probable that duplication of a patient's occlusion is still not 100% predictable. Addition-

ally, the authors have observed that casts from patients with interceptive occlusal contacts in centric relation (CO≠MI) can be mounted with more accuracy than those from patients with harmonious occlusal contacts (CO=MI).

Occlusal refinement techniques have been performed on dental casts to manage some of the inherent material/procedural inaccuracies. Davis described "supraocclusion" of the definitive restoration as the most commonly encountered problem.³⁰ He proposed adjusting the posterior teeth on the cast until the most anterior teeth known to contact intraorally also contact on the casts. Davies et al³¹ also discussed the phenomenon and described the adjustment procedures as "model grooming." Boyarsky et al³² found that dental cast occlusal adjustment procedures, performed by laboratory technicians after mounting, decreased the adjustment time of single posterior crowns at insertion. Although the "supraocclusion" phenomenon is known, and empirical cast adjustment methods seem to have been successfully used, no studies have been designed to objectify the application of the technique.

The importance of contact area on masticatory function³³⁻³⁹ and occlusal stability⁴⁰ is well known, and measurement techniques have been described. One technique uses a vinyl polysiloxane (VPS) material to record the occlusal contact relationship, an optical scanner to record light transmission and optical density through various known thicknesses of material, and imaging software to analyze and quantify areas of "actual contact" and "near contact" up to 300 μm .^{35,37,40-42}

The purpose of this investigation was threefold: (1) to qualitatively compare the occlusal contacts in a simulated patient with bilateral posterior interceptive occlusal contacts (CO≠MI) with respective "derived" mounted stone casts from 2 commonly used dental gypsum products; (2) to quantitatively compare the occlusal

contacts in a simulated patient with a harmonious occlusion (CO=MI) with respective derived mounted stone casts from 2 dental gypsum products; and (3) to quantitatively compare the occlusal contacts in a simulated patient after insertion of a single molar crown fabricated from adjusted and unadjusted derived definitive casts. The hypothesis was that derived casts would have induced interferences and that cast adjustment would compensate for these interferences and allow for a more harmonious occlusion.

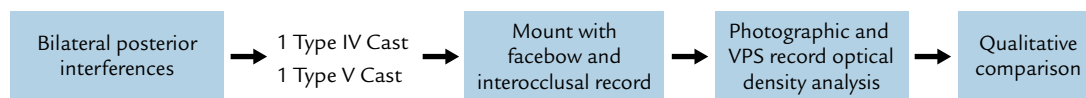
MATERIAL AND METHODS

This investigation was performed in 3 parts (Fig. 1). For all parts, a complete anatomic dentoform (M-1560; Columbia Dentoform Corp, New York, NY) with all 32 permanent teeth was arbitrarily mounted in maximum intercuspation (MI) in a semi-adjustable dental articulator (SAM 3; SAM Präzisionstechnik GmbH, Munich, Germany) to simulate an average patient position; this dentoform articulation served as the simulated patient (SP). Repeated procedural steps are described individually and referenced as: *impression protocol*, *mounting protocol*, and *scanning protocol*. All statistical tests were performed using a computer software package (SPSS 15.0; SPSS, Inc, Chicago, Ill).

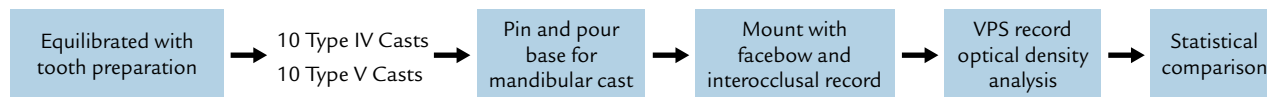
Part 1: Nonequibrated simulated patient

From the initial simulated patient (SP) mounting, the lower member of the dentoform was adjusted so that bilateral posterior interceptive occlusal contacts were created on a pair of molars in each arch (CO≠MI). A VPS (Blu-Mousse; Parkell, Inc, Edgewood, NY) interocclusal record captured the occlusal relationship of the dentoform, and the lower member of the dentoform was remounted. Two maxillary and mandibular complete arch impressions were made of the dentoform, and dental stone casts were fabricated according to the *im-*

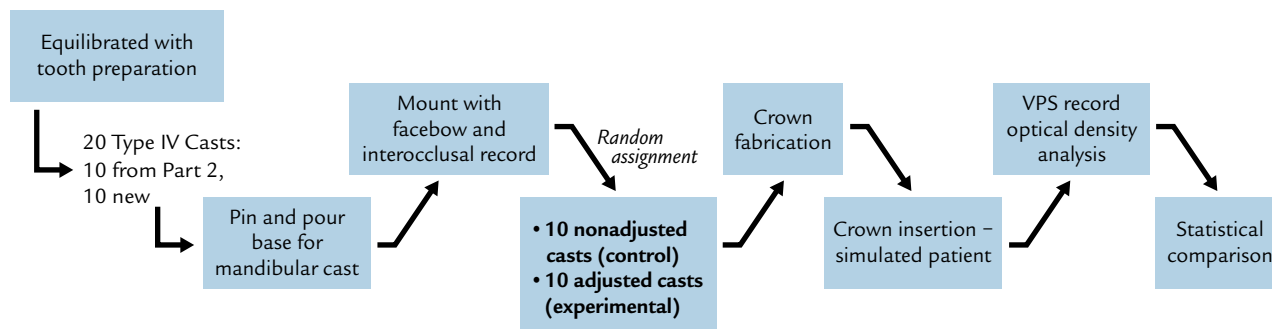
Part 1: Nonequilibrated simulated patient



Part 2: Equilibrated simulated patient



Part 3: Equilibrated simulated patient with inserted crown



1 Outline of material and methods.

pression protocol. One impression set was poured in a high-expansion, type V stone (DK) (Die-Keen, Modern Materials; Heraeus Kulzer, Inc, South Bend, Ind), and the other was poured in a low-expansion type IV stone (SR) (Silky-Rock; Whip Mix Corp, Louisville, Ky). Both sets of casts were mounted/articulated according to the *mounting protocol*. Red silk marking ribbon (Madame Butterfly Silk 3 5/8"; Almore Intl, Inc, Portland, Ore) was used to mark points of contact for the SP, DK, and SR casts, and all were photographed perpendicular to the occlusal plane at x1 magnification. Bilateral VPS (Blu-Mousse; Parkell, Inc) interocclusal registrations were made, scanned, and quantified according to the *scanning protocol*. Resulting data for "actual contact" (AC) and "near contact" (NC) were qualitatively compared.

Part 2: Equilibrated simulated patient

The simulated patient (SP) occlusion was adjusted (equilibrated) so that all teeth occluded in a cusp-fos-

sa/cusp-marginal ridge relationship, and with equal intensity (CO=MI), as judged subjectively with 0.0005-inch shim stock (Artus Corp, Englewood, NJ). The left mandibular first molar was prepared for a complete coverage ADA type IV cast gold crown. Twenty maxillary and 20 mandibular complete arch impressions were made, and subsequent dental stone casts were fabricated according to the *impression protocol*. Ten impression sets were poured in DK and 10 in SR. After 72 hours of setting time, all mandibular casts were segmentally pinned (Axiopins; SAM Präzisionstechnik GmbH), and bases were poured in stone (Flowstone; Whip Mix Corp). Casts were mounted/articulated according to the *mounting protocol*. Bilateral VPS (Blu-Mousse; Parkell, Inc) interocclusal registrations were made for each mounted set of casts. All registrations were scanned and quantified according to the *scanning protocol*. Resulting data for AC and NC were statistically compared using the Kruskal-Wallis ANOVA ($\alpha=.05$).

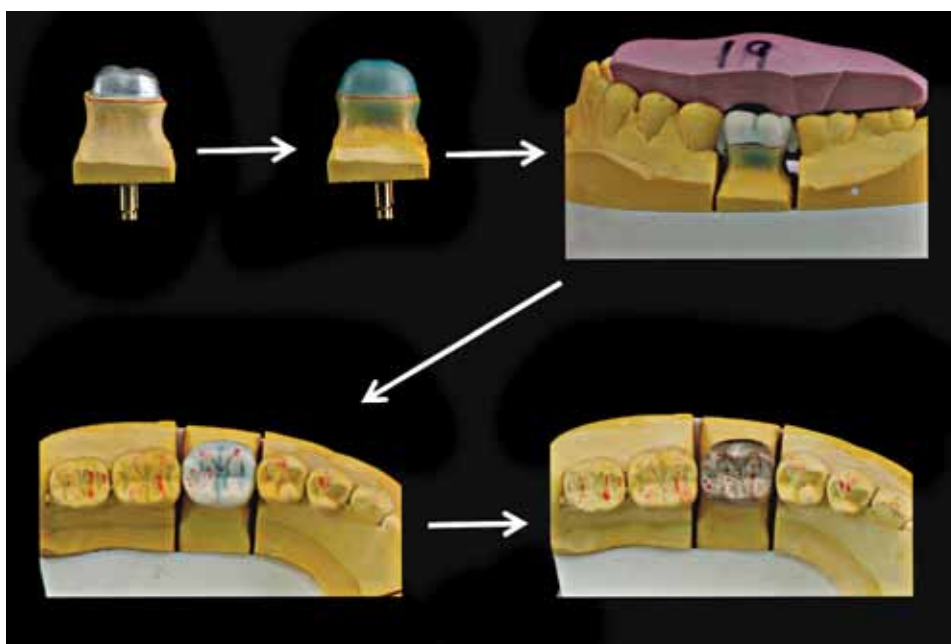
Part 3: Equilibrated simulated patient with an inserted crown

The 10 sets of Silky-Rock (SR) casts used in part 2 were also used in this part of the study, and 10 additional sets were fabricated using the identical protocol. Of the 20 sets of mounted casts, 10 were assigned using computer-generated random numbers to a control group, and 10 to the experimental group. All mandibular casts were sectioned so that the preparation die and the adjacent segments in the left quadrant were removable; the right quadrant remained rigidly fixed to the cast base. Two layers of die spacer (Tru-Fit; George Taub Products, Jersey City, NJ) were placed on all preparation dies to within 0.5 mm of the gingival finish line.

The centric locks on the articulator were released, the lower articulator member was positioned so that the casts were occluded into maximum intercuspation (MI), and the articulator incisal pin was reset to this position. Casts assigned to the experimental group ("experimental crown")



2 Definitive casts in experimental group adjusted prior to crown fabrication. Casts were adjusted to remove artifact CO-MI slide. Adjustment was complete when articulator pin contacted table.



3 Crown fabrication steps: (1) Dies dipped in coping wax. (2) Wax mold used to make “rough” pattern. (3) Cameo contours and occlusal tables of wax patterns refined. Castings were made from ADA type IV dental alloy using lost-wax technique with gypsum bonded investment material. (4) Occlusal contacts of castings were refined.

were systematically equilibrated until the incisal pin attained contact with its table (Fig. 2). Casts in the control group (“control crown”) received no occlusal adjustment.

A complete contour wax pattern (Geo Wax; Renfert GmbH, Hilzingen, Germany) was fabricated on each die. The wax patterns were sprued, invested (Cristobalite; Whip Mix Corp), and cast in type IV dental alloy (Ney-Oro 60; Dentsply Ceramco, Burlington, NJ) using the lost-wax technique. Each restoration was adjusted to its definitive die/cast to hold shim stock with the same firmness as the adja-

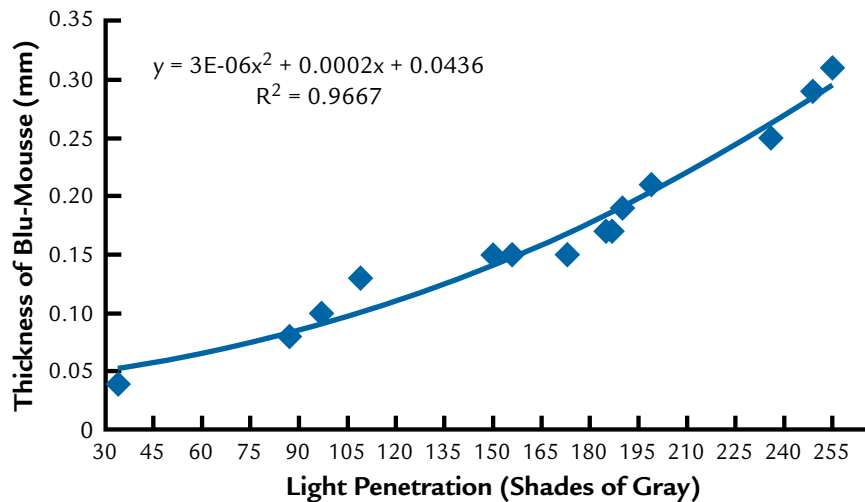
cent teeth (Fig. 3). The restorations were then inserted in the simulated patient (SP) individually. The intaglio of each crown was adjusted if necessary using optical magnification ($\times 4.3$), and an explorer was used to verify complete seating. Bilateral VPS (Blu-Mousse; Parkell, Inc) interocclusal registrations were made of the SP with each restoration in place. The records were scanned and quantified following the *scanning protocol*. AC and NC areas were compared using the Kruskal-Wallis ANOVA ($\alpha = .05$).

To determine the reliability of the interocclusal record fabrication and

scanning protocols collectively, 10 VPS records were made of the equilibrated SP and scanned according to the *scanning protocol*. Interclass correlation coefficient and Cronbach’s alpha statistical figures were determined for AC and NC areas.

Impression protocol

Standardized custom trays were fabricated with visible light-activated tray material (Triad VLC tray material; Dentsply Trubyte, York, Pa), with stops and positioning guides in the land area of the dentof orm casts



4 Calibration step wedge data. Line of best fit for relationship between thickness of VPS record and light transmission was described by quadratic equation with high level of correlation.

to allow 1-2 mm of relief. Trays were painted with tray adhesive (PVS; Kerr Corp, Orange, Calif) and allowed to set for 1 to 6 hours. A dual-phase VPS impression technique with an ADA type I and type III impression material (Extrude; Kerr Corp) was used. Separation time was 12 minutes after seating the impression tray. The impressions were poured within 15 minutes using custom standardized base formers. The casts were separated after 1 to 2 hours of setting time and were allowed to bench set for 72 hours. Each impression was poured only once, and dental stones were used according to the manufacturer's recommended powder/deionized water ratios.

Mounting protocol

A maxillary cast transfer record (Transfer Stand and Transfer Fork Assembly AX; SAM Präzisionstechnik GmbH) was made of the simulated patient on the articulator. Wax (Aluwax; Aluwax Dental Products Co, Alendale, Mich) was used on the transfer fork in a tripod fashion with the record trimmed to expose only cusp tip imprints. Maxillary casts were mounted using this "facebow recording" and mounting stone (Mounting Stone; Whip Mix Corp). The position of the cast was stabilized by hand on

the transfer fork until the stone had reached a snap set. A wax centric relation interocclusal record was made according to the method described by Wirth²⁴ and was used for all mounting procedures. The cast was held by hand and stabilized until the mounting stone was initially set. Magnetic split-cast mounting plates (Axiosplit; SAM Präzisionstechnik GmbH) were also used to facilitate mounting accuracy verification.

Scanning protocol

A double-sided flatbed scanner (Expression 1680; Epson America, Long Beach, Calif) was used for grayscale scans of each VPS record. The records were placed in the scanner in the same location and at the same setting and scanned at 600 dots per inch (dpi). Computer software (ImageTool 3.0; University of Texas Health Sciences Center at San Antonio, San Antonio, Tex) was used to perform optical density analysis of the transmitted light through perforations/near perforations from the first premolars through the third molars bilaterally. A calibration step wedge of 14 known thicknesses was fabricated and scanned so that a regression equation relating transmitted light to VPS thickness could be formulated based on the pixel grayscale values (Fig. 4).

Thickness was determined according to the following derived equation:

$$\text{Thickness} = 0.0436 + 0.0002(\text{GS}) + 0.000003(\text{GS})^2$$

The use of grayscale values (GS) allowed areas of near contact to be measured between 50 and 300 μm . Thicknesses of material $\leq 50 \mu\text{m}$ were classified as actual contact (AC); those between 50 and 300 μm were classified as near contact (NC). The pixel densities were quantified for each record as areas of AC and NC, respectively, in mm^2 .

RESULTS

Part 1: Nonequilibrated simulated patient

The respective actual contact (AC) and near contact (NC) areas were 2.17 and 12.44 mm^2 for the nonequilibrated simulated patient (SP), 2.31 and 11.74 mm^2 for the Silky-Rock (SR) cast mounting, and 3.06 and 14.75 mm^2 for the Die-Keen (DK) cast mounting. Visual comparison of the scanned interocclusal records revealed differences in all 3 records. Areas of actual contact (AC) as confirmed by shim stock and wax had no discernable differences, but there were differences in near contact (NC) areas between all 3 records. Although differences were apparent, they were

small and difficult to interpret, especially with only a single cast mounting per group. No statistical comparisons were made due to the small sample size (Figs. 5 through 7).

Part 2: Equilibrated simulated patient

The actual contact (AC) area was greater for the simulated patient (SP) than for the Silky-Rock (SR) cast, which was in turn greater than the AC area for Die-Keen (DK). The NC area was also greatest for the SP, followed by the DK and SR casts, respectively. There were significant ($P<.001$) differences in AC and NC between the 3 groups; post hoc tests showed that the differences were due to SP, which

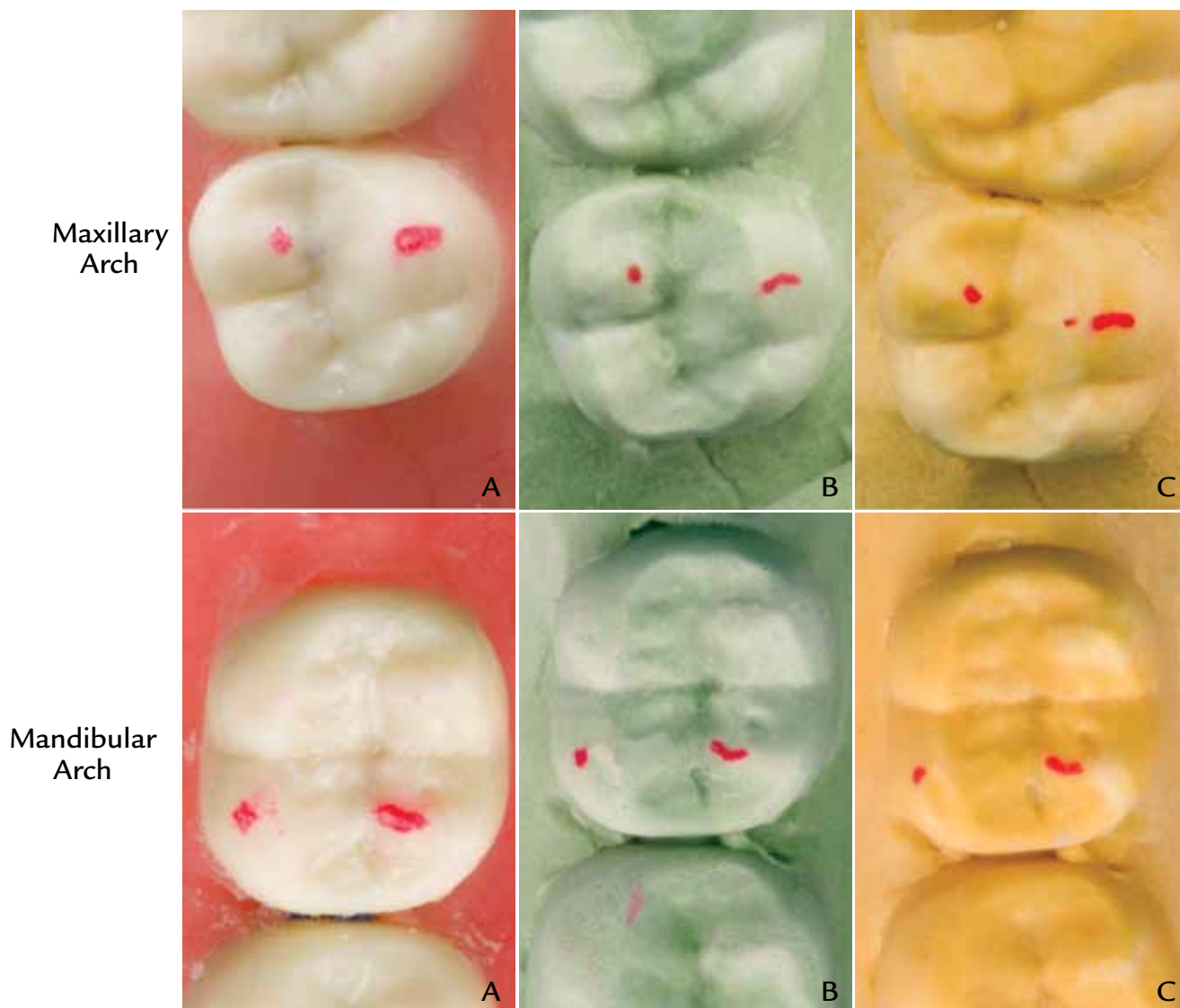
demonstrated significantly ($P<.001$) greater areas of contact and near contact than DK and SR. There were no significant differences in either AC or NC between the 2 gypsum products, SR and DK (Fig. 8) (Table I).

Part 3: Equilibrated simulated patient with an inserted crown

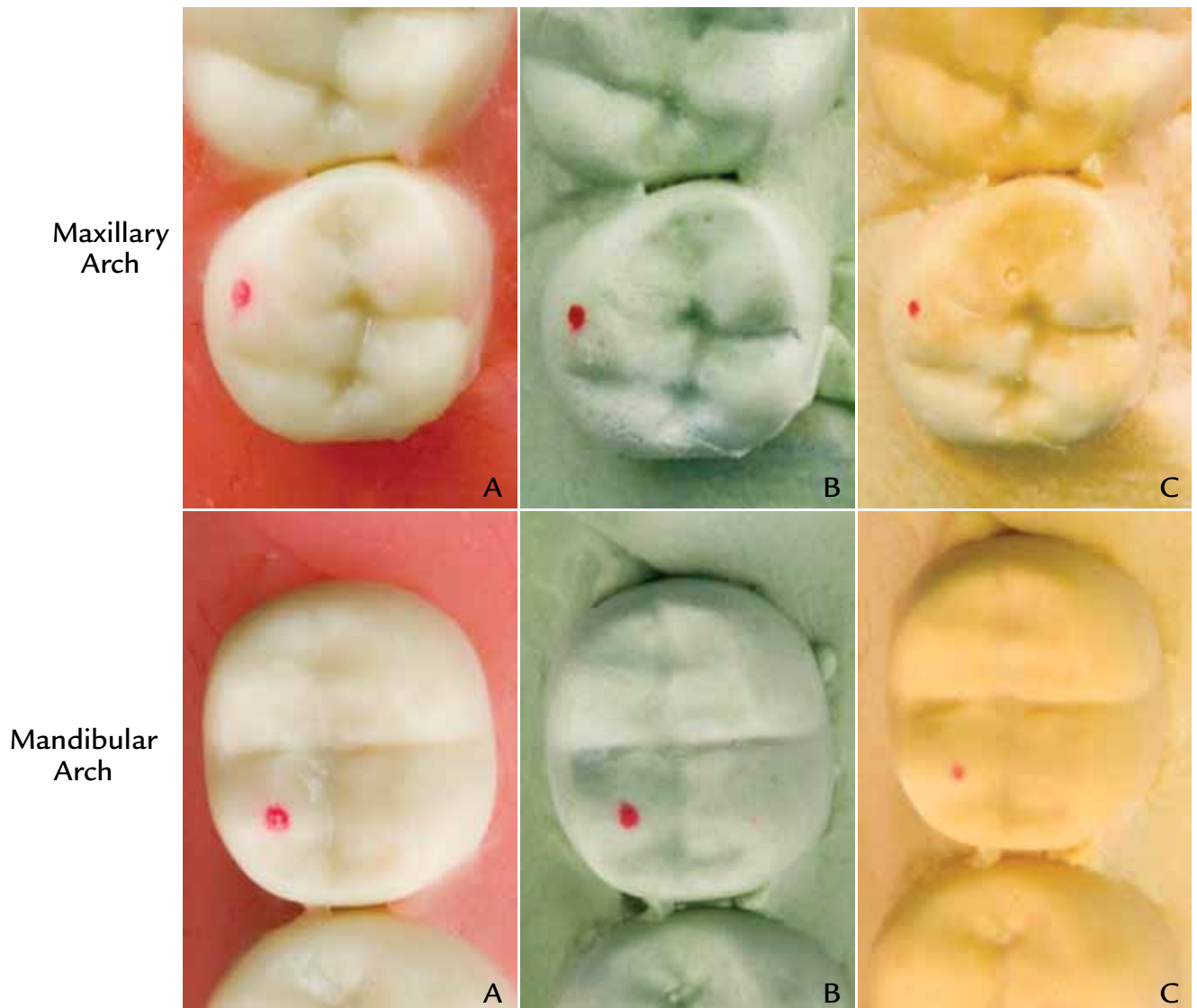
Areas of actual contact (AC) for the simulated patient (SP) with the control crown (no occlusal adjustment of the cast) were significantly smaller than the AC for the SP with no crown ($P<.001$), as well as the AC for the SP with the experimental crown with occlusal adjustment ($P<.001$). There were no significant differences

between the SP with no crown and the SP with the experimental crown. The areas of near contact (NC) demonstrated significant differences between all 3. Areas of NC for the SP experimental crown were significantly ($P<.001$) greater than the NC areas for the SP with no crown, which was in turn significantly ($P<.001$) greater than the NC areas for the SP with the control crown (Table II) (Fig. 9).

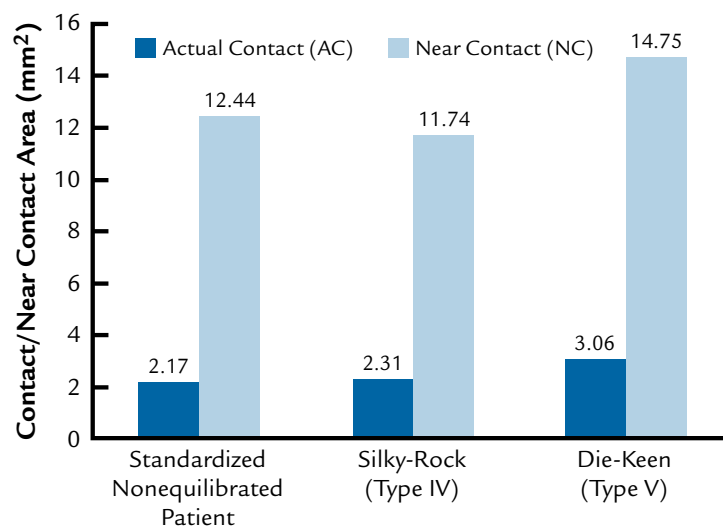
Methods used for obtaining and scanning interocclusal records showed a high level of reliability for AC and NC (interclass correlation coefficients: AC=0.976, NC=0.986; Cronbach's alpha: AC=0.998, NC=0.999).



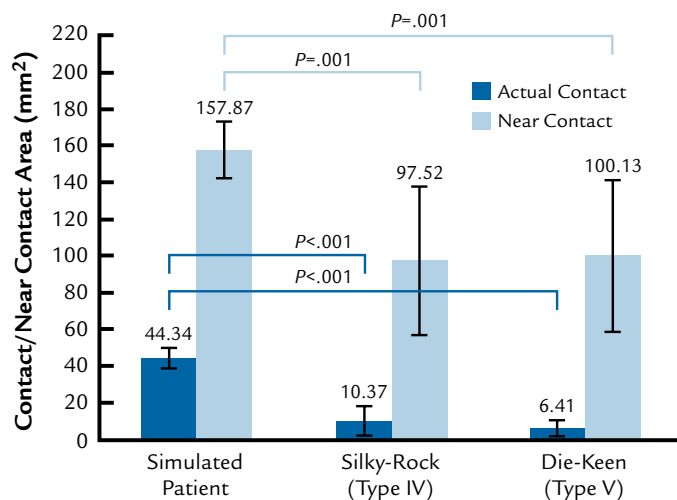
5 Right-side ribbon marking qualitative contact area comparison of (A) nonequilibrated simulated patient (SP) and cast representation with dental stones (B, Die-Keen (DK); C, Silky-Rock (SR)).



6 Left-side ribbon marking qualitative contact area comparison of (A) nonequilibrated simulated patient (SP) and cast representation with dental stones (B, Die-Keen (DK); C, Silky-Rock (SR)).



7 Part 1 contact area group comparison of nonequilibrated simulated patient (SP) with Silky-Rock and Die-Keen dental stone casts.



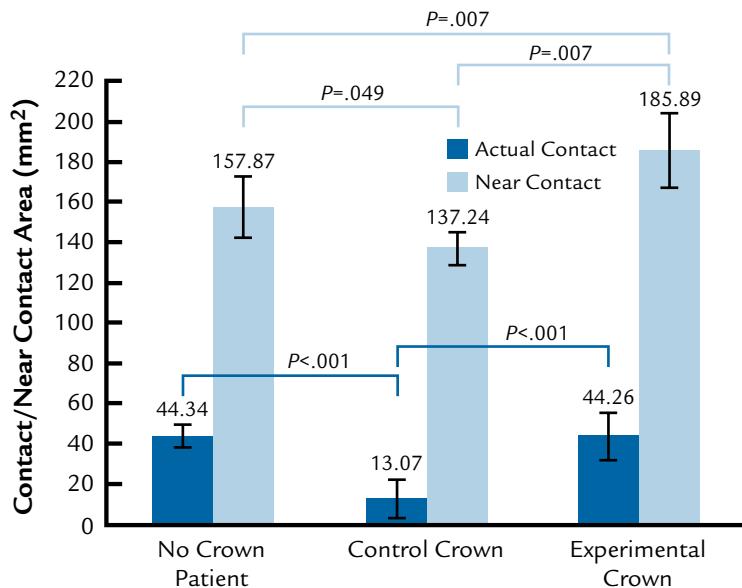
8 Part 2 group comparison showing equilibrated simulated patient (SP) statistical differences for actual contact ($P < .001$) and near contact ($P = .001$) with Silky-Rock and Die-Keen dental stone casts.

TABLE I. Part 2 quantitative comparison of contact areas for equilibrated simulated patient (SP), according to Kruskal-Wallis ANOVA ($P < .05$)

	Descriptive Statistics						Group Comparisons	Paired Group Comparisons		
	Simulated Patient (SP)		Silky-Rock (SR)		Die-Keen (DK)		SP vs. SR vs. DK	SP vs. SR	SP vs. DK	SR vs. DK
	Mean (mm ²)	SD	Mean (mm ²)	SD	Mean (mm ²)	SD	<i>P</i>	<i>P</i>	<i>P</i>	<i>P</i>
Actual Contact (AC)	44.34	5.54	10.37	8.10	6.41	4.37	<.001	<.001	<.001	.257
Near Contact (NC)	157.87	15.18	97.52	40.34	100.13	41.17	.001	.001	.001	.821

TABLE II. Part 3 quantitative comparison of contact areas for equilibrated simulated patient (SP) with crown, according to Kruskal-Wallis ANOVA ($P < .05$)

	Descriptive Statistics						Group Comparisons	Paired Group Comparisons			
	SP No Crown		SP Control Crown		SP Experimental Crown		No Crown vs. Control vs. Experimental	SP No Crown vs. Control Crown	No Crown vs. Control vs. Experimental	No Crown vs. Control vs. Experimental	
	Mean (mm ²)	SD	Mean (mm ²)	SD	Mean (mm ²)	SD	<i>P</i>	<i>P</i>	<i>P</i>	<i>P</i>	
Actual Contact (AC)	44.34	5.54	13.07	9.63	44.26	11.82	<.001	<.001	.650	<.001	
Near Contact (NC)	157.87	15.18	137.24	8.24	185.89	18.52	.003	.049	.007	.007	



9 Part 3 group comparison showing equilibrated simulated patient (SP) with crown statistical differences for actual contact ($P<.001$) and near contact ($P=.003$) areas.

DISCUSSION

This investigation intended to determine, in part 1, whether or not it is possible to duplicate a patient's centric occlusion and interceptive contacts on an articulator using 2 ADA-certified dental gypsum products. The location, size, and shape of the contacts were visibly similar for both stones, but with some small differences. It was difficult to determine the significance of any perceived differences in markings due to variability of the techniques and effect of the ribbon ink on different materials. It is reasonable to assume that any differences would be magnified as the number of contacts in the simulated patient (SP) increased.

In part 2, the finding that the equilibrated SP had significantly greater areas of actual contact (AC) and near contact (NC) when individually compared to the AC and NC areas of the 2 gypsum products indicated a difference in the ability to replicate a patient's occlusion on mounted casts, and supports the research hypothesis. Further, the data suggest that any differences in expansion between the high- and low-expansion gypsum products would not result in replica-

tion of the patient's occlusion with greater accuracy.

The observation of a slightly greater AC area for SR (10.37 mm²) in comparison to DK (6.41 mm²), while the NC areas were roughly equivalent, is interesting, and stone expansion may explain the difference in AC areas. The mechanisms by which this may occur include: (1) vertical expansion on a flat or inclined surface (cusp tip to fossa); (2) horizontal expansion on an inclined surface (inclined plane to inclined plane). In both situations, expansion may cause a slight vertical opening which could result in loss of AC area with a slight gain in NC area; this hypothesis is consistent with the present results for SR and DK as well as previous findings related to stone expansion for the 2 gypsum products.¹⁵

In part 3, the finding that the AC areas for the SP experimental crown were significantly greater than those for the SP control crown is important when evaluating the efficacy of the cast adjustment intervention. The mean AC areas for the SP experimental crown and SP no crown groups were almost equivalent, indicating that the adjustment intervention substantially increased the area of AC to near that of the simulated patient, and

supporting the research hypothesis.

It is interesting that the SP experimental crown had significantly greater areas of NC when compared to the SP no crown, while there was no difference in AC between the two. This observation would indicate that adjustment of the cast yields a greater gain of NC than AC when the crown is inserted, and that insertion of a crown that was fabricated from unadjusted casts will result in a level of hyperocclusion that yields a significant loss in NC area.

Although previous studies suggest that NC may actually be more important for mastication and occlusal stability,^{33,34,37,40} the current investigation suggests that AC is more important when attempting to accurately articulate 2 casts. The low values of AC area were believed to be the result of the effects of greater stone expansion, which manifest in the opening of the occlusal pin and a greater CO-to-MI slide artifact.

The cast adjustment protocol (intervention) followed in this investigation was successful in eliminating the majority of the occlusal disharmony in the casts believed to be caused by the effects of stone expansion, and supports the research hypothesis. Even under controlled in vitro conditions, crowns fabricated on unadjusted casts will be in significant hyperocclusion when inserted, and will cause occlusal disharmony if not adjusted, an effect that would be magnified with multiple restorations. Unfortunately, this adjustment comes at the expense of valuable chair time and loss of occlusal morphology. This finding is also consistent with the observations of Davis,³⁰ Davies et al,³¹ and Boyarsky,³² and adds support to the current authors' clinical observations that interceptive occlusal contacts in centric relation (CO≠MI) can be more accurately represented and verified on an articulator.

The scanner used in the current study presented limitations because the upper threshold for the scanned records is 50 μm and shim stock is 13

µm, and the AC areas for the scanned records may be exaggerated when compared with shim stock. This study was also done in vitro on a dentoform in simulated centric occlusion to decrease in vivo axis location variables, and the teeth were rigidly attached. Although clinically, the acquired, forward-postured, maximum intercuspal position may be the treatment position of choice for single tooth restorations in many patients, the effects of stone expansion will likely preclude the ability to articulate the casts in this position. Therefore, some form of cast adjustment will be necessary to attain the patient's "true" occlusal vertical dimension in the MI position. Future studies should replicate the control and intervention with casts mounted in an MI position to determine if equally satisfactory results can be obtained with respect to occlusal contact areas.

CONCLUSIONS

Within the limitations of this study, the following conclusions were drawn:

1. Casts made of a simulated patient dentoform with coincident centric occlusion (CO) and maximum intercuspal (MI) positions cannot be mounted to replicate the occlusal contact areas.

2. When derived casts from a simulated patient with coincident CO and MI positions are mounted, an artifact CO-MI discrepancy was created.

3. When single posterior crowns were fabricated on casts derived from the simulated patient with coincident CO and MI positions, the resulting castings were in hyperocclusion upon insertion into the simulated patient.

4. When derived casts were "adjusted" prior to crown fabrication, the castings were in near occlusal harmony with the remaining dentition upon insertion.

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NOTEWORTHY ABSTRACTS OF THE CURRENT LITERATURE

Effect of titanium surface characteristics on the behavior and function of oral fibroblasts

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Purpose: The purpose of this study was to evaluate the effect of different titanium surface characteristics on the behavior and function of oral fibroblasts as well as the deposition pattern of collagen within the extracellular matrix.

Materials and Methods: Titanium surfaces created by machining, acid etching with sulfuric acid (AE1), or acid etching with hydrofluoric acid (AE2) were analyzed using scanning electron microscopy (SEM) and energy-dispersive x-ray spectroscopy. Rat oral fibroblasts were cultured on different surfaces. Cell spread and morphology of extracellular matrix were evaluated using SEM. Attachment and proliferation of cells were examined by comparing the numbers of attached to detached cells and cell count, respectively. Gene expression was analyzed via reverse transcriptase polymerase chain reaction. Collagen production and deposition were examined via a Sirius red-based stain assay and confocal laser scanning microscopy.

Results: The machined surface showed a flat profile with isotropic grooves, the AE1 surface showed a uniformly microscale roughened surface, and the AE2 surface had a grooved profile with intermediate surface roughness. The AE2 surface contained fluoride atoms ($2.45\% \pm 0.44\%$ as F/Ti atomic ratio). Cell attachment was significantly weaker on the machined surface than on the AE1 and AE2 surfaces, whereas no differences were observed between the AE1 and AE2 surfaces. The cell counts on the machined and AE2 surfaces were higher, with a parallel orientation, whereas the cell count was lower and randomly distributed on the AE1 surface. The expression level of fibroblastic genes was similar among surfaces for all time points tested. Collagen production was highest on the machined surface, followed by AE2 and AE1 surfaces. Collagen deposition displayed a parallel pattern on the machined surface, while it was multidirectional on the AE1 and AE2 surfaces.

Conclusion: The surface characteristics of titanium affect attachment, spread, and proliferative activity of oral fibroblasts as well as the deposition pattern of collagen in vitro.

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