NEXT-GENERATION BIOCERAMIC SEALERS: POTENTIAL COMPLICATIONS

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Aim: next-generation *bioceramic sealers* are innovative materials in endodontics, designed to interact with biological tissues due to their hydration capacity and release of bioactive ions. They offer advantages in sealing ability, dimensional stability, and biocompatibility over other endodontic sealers (MTA, epoxy resin-based). However, their effectiveness has been questioned due to clinical case observation. This work presents a case where bioceramic sealer led to an adverse reaction, compromising tissue integration.

Methods: in the clinical case analyzed, the sealer was applied following single cone obturation technique. Its characteristics, interaction with inflammatory exudate and tissue response were evaluated through clinical and radiographic assessments. The material's behavior was compared with existing literature to identify potential anomalies in its biological reactivity.

Results: differently from the expected bioactive properties, the sealer triggered a foreign body response, leading to persistent inflammation and lack of periapical tissue healing. The analysis revealed no mineral infiltration at the canal interface and an immune response at the apex, resulting in an unfavorable prognosis for the surrounding tissue and treated tooth.

Conclusions: although bioceramic sealers are considered highly biocompatible, this case highlights the risk of unexpected reactions compromising their effectiveness. Further studies are needed to clarify the mechanisms of interaction between these materials and biological tissues to prevent potential complications.

DEVELOPMENT OF A MACHINE LEARNING ALGORITHM FOR THE CLASSIFICATION OF JAW MOVEMENTS ACQUIRED BY OPTICAL JAW TRACKING SYSTEM

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Aim: the aim of this study was to develop a machine learning algorithm for the classification of mandibular movements and patterns with tracings acquired with an optical jaw tracking system to support the clinical objective diagnosis of Temporomandibular Disorders (TMD).

Methods: mandibular movements were recorded with an optical jaw tracking kinesiograph (Cyclops, Itaka) from a sample of 160 subjects, students of the C.d.L. in Dentistry of the University of Perugia. Records were divided into healthy or with TMD groups, converted from point clouds to 2D images, and used to set up two models: k-Nearest Neighbors (kNN) and Support Vector Machine (SVM), processed in Python. 80% of the dataset was used for training and 20% for testing the models.

Results: the results show that kNN achieved an accuracy of 75%, while SVM achieved an accuracy of 74.38%, demonstrating a good discriminating ability.
The analysis showed that increasing the dataset size significantly improved the performance of the models, suggesting the need to further expand the database to refine diagnostic accuracy.

Conclusions: the results obtained confirm the potential of machine learning algorithms as a support tool in clinical diagnosis, with the future perspective of reducing the rate of error by integrating new models and expanding the sample, considering enrollment a more heterogeneous group of subjects.

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AMELOGENIN-DERIVED PEPTIDES (SP AND LRAP) CYTOTOXICITY ON HUMAN DENTAL PULP STEM CELLS (HDPSCS): AN *IN VITRO* STUDY

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Aim: porcine Enamel Matrix Derivative (EMD), primarily composed of amelogenins, is currently used to treat bone defects in periodontal surgery. Recombinant amelogenin-derived peptides, such as SP and LRAP, appear to preserve or even improve the biological activity of the full-length protein, attracting interest in the field of bone regeneration. This preliminary study aimed to assess the cytotoxicity of SP and LRAP on human Dental Pulp Stem Cells (hDPSCs) as a foundation for future investigations into the osteoinductive potential of these peptides.

Methods: SP and LRAP peptides on hDPSCs were tested at different concentrations (1, 5, 10, 50 and 100 ng/mL). A stock solution was prepared by resuspending the peptides in Phosphate-Buffered Saline (PBS) at an initial concentration of 100.000 ng/mL. Viability tests were performed using the MTT assay.

Results: treatment with SP peptide at concentrations of 50 and 100 ng/mL resulted in a reduction in hDPSC viability after 24 hours. Furthermore, SP 100 ng/mL decreased cell proliferation after 48 hours. No significant cytotoxicity was observed at any concentration of SP after 72 hours. LRAP reduced cell viability at concentration of 50 and 100 ng/mL after 24 hours. No significant differences in cell proliferation were observed in LRAP-treated cells after 48 and 72 hours.

Conclusions: the amelogenin-derived peptides, SP and LRAP, appear to be cytotoxic on hDPSCs at 50 and 100 ng/mL concentrations after 24 hours of treatment. Our future studies will now focus on their osteoinductive potential on hDPSCs.

TOLERABILITY OF SOFT TISSUES TO TWO DIFFERENT BIOACTIVE GLASSES FOR BONE REGENERATION: AN *IN VITRO* STUDY ON HACAT CELLS

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Aim: a crucial clinical challenge in modern implant dentistry is the treatment of bone defects and lacunae caused by trauma or disease. Due to the inherent limitations of autologous and heterologous grafts, synthetic materials have gained raising interest in guided tissue regeneration. Among these, bioceramics, and particularly bioactive glasses, have seen extensive use in recent years due to their well-known biocompatibility and regenerative potential. The aim of this study is to evaluate the tolerability of soft tissues to two different bioglasses: the gold standard 45S5 Bioglass® and the recently developed BGMS10.

Methods: this study involved *in vitro* testing using HaCaT cells, a spontaneously immortalized keratinocyte line. The tests assessed cell viability over ten days of culture and analyzed inflammatory responses through cytokine secretion profiles at three and five days. HaCaT cells were exposed to 45S5 Bioglass® and BGMS10 and outcomes were compared with control cultures.

Results: viability analysis revealed significant cytotoxicity from both bioactive glasses, with a statistically significant difference favoring BGMS10 over 45S5 Bioglass®. Inflammatory response analysis showed no significant differences between the control and study groups, indicating that contact with either material did not trigger inflammation.

Conclusions: BGMS10 demonstrated tolerability compared to 45S5 Bioglass® and holds promise for enhancing soft tissue healing. These findings, though preliminary, suggest that BGMS10's composition could provide advantages in clinical applications.

IN VITRO PRECLINICAL STUDY: DEVELOPMENT AND CHARACTERIZATION OF A HYALURONIC ACID AND PLATELET-RICH PLASMA COMPOSITE BIOMATERIAL FOR ENHANCING CELLULAR FUNCTIONS AND TISSUE REGENERATION

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Aim: this study developed and analyzed a composite biomaterial composed of Platelet-Rich Plasma (PRP) and Cross-linked Hyaluronic Acid gel (CHA) (Regedent, Zurich, CH). The aim was to determine whether incorporating CHA enhances platelet viability and modulates the release of their factors. Furthermore, PRP-CHA-conditioned medium was used to evaluate the adhesion and proliferation of human cells, i.e. gingival fibroblasts and osteoblasts.

Methods: PRP was collected from 10 healthy donors (5 males and 5 females) using the Idria G kit (Quantix, Milan, IT). PRP and CHA gel (at 1.8% and 0.9% v/v) were mixed to prepare PRP-CHA matrices. Additional matrices composed of PRP alone and PRP diluted in saline (NaCl 0.9%) were used as controls. All matrices were characterized, and platelet viability as well as the release of platelet granules were assessed. Next, platelet-conditioned media derived from PRP-CHA 1.8%, PRP-CHA 0.9%, PRP alone, or PRP+NaCl 0.9% were added at different volumetric percentages in place of Fetal Bovine Serum (FBS) in the culture media of human gingival fibroblasts and osteoblasts to assess their functions.

Results: PRP-CHA matrices improved platelet viability by enhancing their stability over time and allowing for a prolonged release of their bioactive factors. Moreover, PRP-CHA matrices upregulated genes encoding pro-proliferative and pro-migratory factors in fibroblasts and osteoblasts, enhancing their biological functions in a dose-dependent manner.

Conclusions: PRP-CHA biomaterials enhance human cell functions and represent a promising tool for tissue regeneration.

ANALYSIS OF THE OPTICAL BEHAVIOUR OF CLEAR ORTHODONTIC ALIGNERS EXPOSED TO PIGMENTING BEVERAGES

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Aim: the aim of this *in vitro* study was to analyse the optical changes, in terms of transmittance, of the latest generation of orthodontic aligners to establish whether the possible alterations in transparency, which would occur after their exposure to food liquids with different pigmenting power and acidity, are actually the result of chemical interactions of a permanent type and therefore capable of structurally modifying the polymeric nature of the production materials.

Methods: 6 different commercial brands of orthodontic aligners were selected: Clear Correct®, FlexiLigner®, Invisalign®, Nuvola®, Spark™ and Sure Smile®. Of each, 5 new aligners of the upper arch, were considered for a total of 30 aligners. From each aligner, 2 samples were obtained by sectioning the right and left molar area into 5 x 5 mm squares, for a total of 60 samples (n = 10). On the samples of each brand, with a UV/Vis Lambda35 spectrophotometer, the light transmission was measured through a range of wavelengths between 210-1100 nm, before and after in vitro aging in 4 beverages with different acidity: coffee, black tea, red wine and cola, at a stable temperature of 37°C.

Results and conclusions: from the results obtained, in the orthodontic aligners examined, there was a decrease, albeit slight, in the transmittance values in all cases of prolonged contact with pigmenting drinks with different acidity, a reduction which however varies according to the agent considered but also to the peculiar chemical nature of each polymer blend, characteristic for each brand of aligners, often similar but with some peculiarities which are often difficult to investigate, because they are difficult to know.

EFFECT OF HYALURONIC ACID GEL ON CLOT STABILITY: AN *IN VITRO* STUDY

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Aim: this *in vitro* experimental study aims to investigate the effect of cross-linked hyaluronic acid gel (hyaDent BG, Regedent AG, Zurich, CH) on clot stability.

Methods: effect of cross-linked Hyaluronic Acid gel (xHyA) on clot stability was assessed using the thromboelastograph TEG 5000, measuring five parameters: Reaction time (R), achievement of a certain clot firmness (K), kinetics of clot development (angle), Maximum Amplitude (MA), and clot lysis percentage (LY30). The study involved ten healthy volunteers (five women and five men, aged 24-26 years) and analyzed four blood sample groups: blood (control group), blood with saline solution (FISIO), blood with 0.9% xHyA in saline solution (HA 0.9%), and blood with 1.8% xHyA in saline solution (HA 1.8%). Data were analyzed using the Friedman test, ANOVA, and post-hoc analyses with paired t-tests and Dunn's test.

Results: both HA 0.9% and HA 1.8% groups showed a significantly reduced LY30 compared to the control group. The HA 1.8% group also exhibited a significantly higher MA compared to the control group. No significant differences were found in R, K, or angle between HA 0.9% and HA 1.8% groups and the control group. The FISIO group showed a significant reduction in R, along with increased angle and MA, compared to the control group.

Conclusions: these findings suggest that xHyA, particularly at a concentration of 1.8%, can reduce clot lysis and enhance clot stability, which could be beneficial in clinical settings where clot formation and stability are essential for tissue healing and regeneration.

COMPARATIVE EVALUATION OF POLISHING PROTOCOLS FOR ONE UNIVERSAL SHADE NANOCOMPOSITE

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Aim: this study evaluates the impact of different polishing protocols on the gloss and surface morphology of one universal shade nanocomposite.

Methods: 20 composite samples (2 mm thick, 12 mm diameter) were divided into 4 groups (n = 5), based on the polishing protocol. Each sample had a control side (CTR), polymerized under Mylar and a test side (F/P) subjected to: Group A (TwistA, TwistB), Group B (TwistA, TwistB, LUCIDA), Group C (TwistA, LUCIDA), and Group D (Sof-Lex Spiral Wheels, LUCIDA). Gloss (GU) was measured with a glossmeter, and surface morphology was analyzed by SEM. Data were assessed using ANOVA and Tukey's test ($p < 0.05$).

Results: a significant difference was found among groups for Side F/P ($p < 0.001$), while no differences for Side CTR ($p > 0.05$). Groups A and D differed significantly between Sides F/P and CTR ($p < 0.001$; $p < 0.05$), while Groups B and C did not ($p > 0.05$). Group A (50.36±4.53 GU) differed from all others ($p < 0.001$), while B, C, and D showed no significant differences ($p > 0.05$).

Conclusions: polishing significantly affects gloss and surface morphology. Protocols including LUCIDA achieved the highest gloss, further enhancing the material's surface properties.

3D-FEA ANALYSIS OF THE MECHANICAL BEHAVIOR OF ALKASITE POSTERIOR RESTORATIONS *VS* POLYMERIC MATERIALS

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Aim: the present study evaluated the effect of the combination of different dental filling materials in Class I cavities under occlusal loading using three-dimensional Finite Elements Analysis (FEA).

Methods: six computer-generated and restored models of a lower molar were created in the CAD software and compared according to the biomechanical response during chewing load condition. Two adhesively bonded bulk restorative materials were evaluated with or without the presence of a base material below. A food bolus was placed on the occlusal surface mimicking the compressive occlusal load (600 N) during the static linear analysis. The maximum principal stress (tensile) was calculated as stress criteria in enamel, dentin and restoration.

Results: all models showed high stresses along the enamel/restoration margin with a similar stress trend for models restored with the same upper-layer material. Stress values up to 12.04 MPa (Alk) or up to 11.12 MPa (BF) were recorded at the enamel margins. The use of flexible polymeric or ionic base material in combination with bulk-fill resin composite or Alk did not reduce the stress magnitude in dentine and enamel. Class I cavities adhesively restored with bulk-fill resin composite showed lighter stress concentration as well as Alk.

Conclusions: with the limitations of the present study, the adhesively bonded Alkasite restorations showed a promising mechanical behavior with reduced stress magnitude when used above different base materials or as a bulk restoration for Class I posterior restorations.

COMPUTER-AIDED DESIGN AND FINITE ELEMENT EVALUATION OF RESIN COMPOSITE RESTORATIONS IN PREMOLAR PROXIMAL CAVITIES

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Aim: this study evaluated the stress distribution in five different class II cavities of premolar models restored with conventional or bulk-fill flowable composite by means of Finite Element Analysis (FEA) under shrinkage and occlusal loading.

Methods: an upper validated premolar model was imported in the software, and five class II cavities with different occlusal extensions and dimensions were prepared: horizontal cavity on the mesial surface, mesio-occlusal cavity, mesial cavity, tunnel type cavity and direct access cavity. The models were restored with conventional or bulk-fill flowable resin composite. The Maximum Principal Stress criteria was chosen to evaluate the tensile stress results.

Results: the lowest shrinkage stress value was observed in the direct access cavity restored with bulk-fill flowable resin composite (36.12 MPa). The same cavity, restored with conventional composite, showed a score of 36.14 MPa. The horizontal slot cavity with bulk-fill flowable showed a score of 46.71 MPa. The mesio-occlusal cavity with bulk-fill flowable had a score of 53.10 MPa, while with conventional composite this was 55.35 MPa. Higher shrinkage stress was found in the vertical slot cavity with conventional resin 56.14 MPa, followed by the same cavity with bulk-fill flowable 56.08 MPa.

Conclusions: results indicated that the use of bulk-fill flowable composite resin more significantly decreased the polymerization shrinkage stress magnitude. The larger the cavity and the volume of material necessary to restore the tooth, the greater the residual stress on enamel and dentin tissue.

IN VITRO MECHANICAL, BIOLOGICAL AND MICROBIOLOGICAL PROPERTIES OF AN AG-OMD ENHANCED COMPOSITE RESIN

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Aim: bacterial inhibition and prevention of secondary through ion-releasing materials are essential for direct composite restorations. The aim of this study is to evaluate the *in vitro* mechanical, biological and microbiological properties of silver nanoparticles (Ag) paired to an inorganic support (OMD) and added to composite resin.

Methods: an AgOMD Nanoparticle powder was added to a composite resin (1% weight). Three sample groups were prepared: 1% AgOMD, 1% OMD, and unmodified composite resin. A total of 15 cylindrical samples for the compressive test, 15 rods for the three-point bending test, 60 disks for biological tests and 30 disks for microbiological tests against *S. Mutans* and *S. Oralis* were produced.

Results: the same deformation at maximum strength was measured for all the samples, while AgOMD and OMD nanoparticles led to a reduction of elastic modulus of the samples with a reduction of maximum strength. The 1% AgOMD disks induced lower inhibition of cell viability compared to 1% OMD and Clean disks; all disks significantly reduced IL-1β and IL-6 secretion, with 1% AgOMD and 1% OMD having the best performance. Significant reduction of biofilm biomass was observed on AgOMD 1% for both *S. mutans* and *S. oralis*).

Conclusions: biological and microbiological tests have shown that the addition of silver nanoparticles has an inhibitory effect both on bacteria and inflammatory cytokines release. Further investigations should be performed to confirm the potential of bioactive composites and the difference in mechanical performances when compared to traditional composites.

THE EFFECTS OF LIGHT CRYSTAL DISPLAY 3D PRINTERS, STORAGE TIME AND STEAM STERILIZATION ON THE DIMENSIONAL STABILITY OF A PHOTOPOLYMER RESIN FOR SURGICAL GUIDE

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Aim: this *in vitro* study evaluated the dimensional changes of 3D surgical guides printed in-house using Shining 3D SG01 surgical guide resin.

Methods: five test bodies with different shapes and dimensions were designed using CAD software and printed with three LCD 3D printers: AccuFab-L4D, Elegoo Mars Pro 3 and Zortrax Inspire. Following standardized printing and post-processing protocols for SG01 resin, each printer produced 25 samples (5 per shape) for a total of 75 specimens. Dimensional measurements were performed using a digital caliper at four time points: after printing (T0), after one month of storage (T1), post- sterilization (T2) and one month post-sterilization (T3).

Results: all samples showed deviations from the CAD reference value of 12.25 mm at T0 and T2. Similar dimensional variations were observed at T1 and T3. Among the tested printers, AccuFab-L4D exhibited the highest dimensional stability, followed by Zortrax Inspire and Elegoo Mars Pro 3.

Conclusions: printer type, sterilization and storage duration significantly influenced the dimensional stability of surgical guides fabricated with SG01 resin.

IN VITRO BIOLOGICAL EFFECTS OF SWITCHING FROM TRADITIONAL SMOKE TO NEW ALTERNATIVE SMOKING DEVICES (IQOS AND ELECTRONIC CIGARETTE) ON ORAL HUMAN CELL POPULATIONS

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Aim: the aim of the study was to evaluate the biological effects of switching from Traditional Smoke (TS) to alternative smoking devices on human gingival fibroblasts and oral keratinocytes.
Methods: TS (Gold Pocket, Marlboro), Electronic cigarette (E-cig) (Noir-Smooke) and IQOS (Philip Morris) were used. The smoking extracts, with concentration ranging from 100% to 3.12%, involved 4 h administrations for 4 consecutive days, and 4 h for 2 days with TS and for the next 2 days with medium (to simulate smoking cessation) or E-cig or IQOS (to simulate smoke switching). Cell viability (MTT assay), apoptosis and cell cycle (flow cytometry), and DNA damage (immunofluorescence) were performed on fibroblasts; cell viability (MTT assay), IL-1β and IL-6 protein level (ELISA) and gene expression (RT-PCR), and DNA damage (immunofluorescence) were performed on keratinocytes.
Results: TS-induced viability inhibition at low dilutions in fibroblasts was reduced by E-cig and IQOS switching. IQOS switch also partially reduced apoptosis and DNA damage. While in keratinocytes switch showed a recovery of the TS-induced effects with medium, not always evident with E-cig or IQOS. TS at high dilutions induced increased secretion of IL-1β, partially reduced after medium switching. TS-induced cell damage at 96h was partially recovered after switching with medium or IQOS, while with E-cig the damage was greater.
Conclusions: switching from TS to alternative smoking devices showed a reduced cell damage on fibroblasts compared to keratinocytes, and appeared to reduce TS-induced damage, particularly with IQOS.

COLORIMETRIC EVALUATION OF A 3D PRINTABLE RESIN *VERSUS* CONVENTIONAL COMPOSITE RESINS

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Aim: this study analyzes the color and translucency of a 3D printer resin, comparing it to different traditional composite resins. Color was evaluated with two different methods.
Methods: a hybrid ceramic-filled 3D printing Resin (3DR) (SprintRay™ Crown A3 Dentin) was compared to two traditional composites, A3 Body (A3B) and A3 Dentin (A3D) (Filtek™ Supreme XTE 3M ESPE). Samples were made at four thicknesses: 0.5, 1, 1.5, and 2 mm. SpectroShade-Micro spectrophotometer (MHT Spa, Italy) and Optishade colorimeter (SmileLine, Switzerland) were used to describe the colour using CIELCh color space. Translucency was calculated as the difference between color on white and black background ($p < 0.05$).
Results: with a perceptual threshold of ΔE1.2, all resins showed noticeable differences at each thickness. For A3 Body and 3DR, the increase in thickness resulted in an increase in chroma and a decrease in value and hue. A3D showed a general decrease in all three parameters. Translucency decreased with thickness for all materials. A3B showed the highest translucency, while A3D reported similar opacity to 3DR at 0.5 mm and 1 mm but was opaquer at 1.5 mm and 2 mm. Optishade detected greater differences in color and translucency than the spectrophotometer.
Conclusions: although 3DR is described as a dentin, its shade behavior is different from traditional composites. These differences must be taken into account especially when layering indirect restorations.

ANALYSIS OF MICROBIAL AND HUMAN FIBROBLAST COLONIZATION TO 3D-PRINTED PROSTHETIC RESINS ENRICHED WITH L18R SYNTHETIC PEPTIDE

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Aim: the uncontrolled spread of microbial biofilms in the oral cavity undoubtably plays a crucial role in the failure of prosthetic treatment. This study analyzes the biological response of microorganisms and Human Gingival Fibroblasts (HGFs-1) to two prosthetic resins with and without the incorporation of antibody-derived peptide L18R.

Methods: 3D printed samples were fabricated using two commercially available prosthetic resins (VarseoSmile Temp and Raydent C&B) and polished with different techniques. Surface characteristics were evaluated by atomic force microscopy and Scanning Electron Microscopy (SEM). Microbiological assessments of *S. sanguinis* and *C. albicans biofilms*, were conducted by SEM and confocal microscopy, with and without the addition of L18R. Gene expression analysis was performed to study genes involved in HGFs-1 adhesion and inflammatory response.

Results: samples presented changes in hydrophilicity and roughness due to the resin and polishing. All of them demonstrated acceptable biocompatibility, while Varseo showed better properties in reducing bacterial adhesion and interacting with HGFs-1. L18R antimicrobial activity was effective on both resins and significantly stimulated the encoding of proteins involved in HGFs-1 anchoring and proliferation.

Conclusions: while smooth surface achieved using polishing paste and glaze did not reduce microbial adhesion on both resins, the treatment with L18R reduced the total biomass on the resins and displayed a non-cytotoxic behavior, significantly stimulating the expression of HGFs-1 genes involved tissue regeneration.

SURFACE PROPERTIES EVALUATION OF AN ELASTOMERIC MICELLE-BASED GLASS-IONOMER CEMENT

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Aim: to evaluate *in vitro* the surface roughness (Ra), water sorption (Wsp) and solubility (Wsl) of a Glass-Ionomer Cement (GIC) based on elastomeric micelle technology compared to conventional GICs.

Methods: a total of 30 disks (10 x 10 x 1 mm) were prepared from 3 GICs (n = 10), differing in composition [Deltafil (DMG); Fuji IX (GC Corp.); Equia Forte (GC Corp.)]. A surface varnish was applied only for Fuji IX and Equia Forte, as recommended by the manufacturers. Wsp and Wsl tests were conducted following the ISO:4049 guidelines. Ra was measured immediately after specimen preparation and after Wsp and Wsl testing, using a digital microscope. Data was collected and statistically analyzed ($p < 0.05$).

Results: Fuji IX exhibited the highest Ra, regardless of the testing period ($p < 0.05$). No significant differences in Ra were observed between Deltafil and Equia Forte ($p > 0.05$). Equia Forte demonstrated the highest water sorption among groups ($p = 0.001$), while no statistical differences were observed between the other materials ($p > 0.05$). Wsl values were similar among all the GICs ($p = 0.05$).

Conclusions: the newly introduced micelle-based GIC showed surface characteristics that were comparable to, or even better than, those of conventional GICs. This finding is particularly relevant clinically, given that varnish application is not required. Further studies are warranted to comprehensively assess its mechanical performance.

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BONE HEALING PATTERNS IN OSTEOPOROTIC PATIENTS: A STUDY ON TITANIUM SURFACES TREATED WITH VITAMIN D NANOPARTICLES

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Aim: to evaluate the osteogenic effect of grade 4 titanium discs functionalized with a vitamin D3 nanoemulsion on primary osteoblasts derived from trabecular bone of osteoporotic patients.

Methods: titanium discs (10 mm diameter, 2 mm thick) were coated with a 1 µg/mL nanoemulsion of vitamin D3 (corn oil, Tween 80, ultrapure water), prepared by ultrasound (100 W, 20 kHz, 10 min). Osteoblasts were isolated from osteoporotic bone via enzymatic digestion and cultured in osteoinductive medium (DMEM, 10% FBS, ascorbic acid, β-glycerophosphate, dexamethasone). Cells were seeded on treated and untreated discs. Osteogenic markers were evaluated by immunocytochemistry (osteocalcin, ALP), fluorescence microscopy (DAPI/FITC), and alizarin red S staining (OD 562 nm).

Results: nanoemulsion-treated surfaces induced a 36% increase in osteocalcin, 42% in ALP activity, and 48% in mineral deposition compared to controls. Morphology showed improved cell spreading, adhesion, and structured filopodia.

Conclusions: vitamin D3 nanoemulsion enhances early osteogenic differentiation in osteoporotic osteoblasts. This surface modification could improve implant integration in patients with poor bone quality.

THE INFLUENCE OF MATERIAL TEMPERATURE IN DENTAL LABORATORY DUPLICATION: EFFECTS ON THE ACCURACY AND ADAPTATION OF METALLIC REMOVABLE DEVICES

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Aim: the aim of this study is to optimize the duplication process to avoid distal prosthetic defects and improve the precision and adaptability of the custom-made medical device.

Methods: 20 models were used with two colloid casting methods and 3 key factors were evaluated:

a) model temperature: The models were treated with water at two temperature ranges: 10 models at room temperature 21°C and 10 models at 35°C;

b) colloid temperature: for 10 models the casting was performed at a temperature between 56-58°C, and 10 models at a temperature between 52-54°C;

c) cooling method: the first 10 models were cooled in water at room temperature 21°C, for the other 10 slow cooling was used with flasks immersed in water at a temperature of 35°C.

Results: the first 10 models obtained showed the formation of micro bubbles, while the slowly cooled ones produced uniform duplicates. Castings above 54°C showed contraction, while those between 52-54°C ensured greater precision. Rapid cooling caused surface defects and dimensional inconsistencies in the final artifact in the distal part, while the slow one preserved the structural integrity without altering the metal structure.

Conclusions: alterations from the temperatures recommended by the manufacturers influence the adaptation and precision of the prosthesis. The final cooling phase is crucial to control contraction. Following the guidelines on the use of gelatin and the correct management of the duplicating flask is essential to obtain precise and defect-free custom-made medical devices.

THE DOSAGE OF BONE BIOMARKERS ON SALIVARY MATRIX TO SUPPORT THE EVALUATION OF PATIENTS TREATED WITH RAPID MAXILLARY EXPANDER: PRELIMINARY STUDY

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Aim: orthodontic treatment with Rapid Palatal Expander (RPE) stresses palatal suture of maxilla with a variation of bone biomarkers. In the study, changes in the concentration of Parathyroid Hormone-related Protein (PTHrP) marker of bone regulation, type I collagen N-terminal Propeptide (PINP) marker of bone deposition, Tartrate-Resistant Acid Phosphatase (TRAcP) marker of bone resorption were evaluated in saliva.

Methods: 48 patients aged 8 to 10 years with dental and skeletal Class I relationships, maxillary transverse deficiency were enrolled. The study was conducted at the Dental Clinic of Foggia in collaboration with the Clinical Pathology Unit of the Polyclinic-Bari. Saliva samples were collected at five different time points: (T0) before application, at (T1)15, (T2)30, (T3)45, (T4)60 days after the start of expansion. Statistical analysis was performed with the Shapiro-Wilk normality test and U -Mann-Whitney test.

Results: a statistically significant difference was not found for PTHrP and TRAcP. PINP showed a statistically significant difference between T0, T2, T3 confirming that the variations at the level of the palatal suture are biologically evident in saliva when there is the peak of collagen deposition between the 30th and 45th day and coincide with the beginning of new bone formation.

Conclusions: the use of RPE leads to a significant increase in PINP, with collagen deposition and the beginning of bone formation. This preliminary study on saliva has highlighted how biochemical changes support and guide the clinician in the effectiveness of the treatment.

IMPLANT PROVISIONAL LONGEVITY: *IN VITRO* WEAR RESISTANCE IN 3D-PRINTED AND CAD/CAM RESIN MATERIALS

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Aim: this *in vitro* study investigated the wear resistance of two materials used for interim prostheses in complete-arch implant-supported fixed prostheses.

The null hypothesis was that there were no significant differences between wear resistance of the two materials.

Methods: two interim resin materials were evaluated: three-dimensional (3D) printed resin material (Varseo Smile Crown plus-VSC) (Test-1) and CAD/CAM milled resin material (Huge PMMA Block), (Test-2). A total of 22 specimens were prepared (N = 11 each). After thermomechanical aging (240,000 cycles, 50 N load, 5°C-55°C thermocycling, CS-4.4 chewing simulator), the two interim resin materials were scanned with a confocal microscope Sensofar S Neox 3D profilometer to generate 3D models of the surfaces. The specimens were compared, considering the Area (mm^2), the Volume (mm^3) and Max h/d (µm) of the impression for each specimen. The statistical analysis was performed with One-Way Anova and Dwass-Steel-Cristchlow-Fligner pairwise comparisons.

Results: there was no statistically significant difference of wear resistance between the tested materials.

Conclusions: in this *in vitro* investigation, 3D printed interim resin showed comparable mechanical behavior to traditional CAD/CAM milled resin.

EFFECT OF 2-STEP SILVER-CONTAINING SOLUTIONS ON DENTIN BONDING PERFORMANCE: 2-YR EVALUATION

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Aim: to assess the effects of ammonia- and water-based 2-step silver-containing solutions on microtensile bond strength (μTBS), chemo-morphological characterization (SEM/EDS), and matrix-metalloproteinases (MMPs) activity of a universal adhesive to sound dentin after 2 years of artificial aging.
Methods: sixty sound human molars were cut to expose mid-coronal dentin and assigned to 6 groups, based on dentin pre-treatment and Zipbond Universal (ZB, SDI) application mode (n = 10): 1) ZB in the self-etch mode (control SE group, ZBSE); 2) Riva Star (SDI) and ZBSE; G3) Riva Star Aqua (SDI) and ZSE; G4) ZB in the etch-and-rinse mode (control ER group, ZBER); G5) Riva Star and ZBER; G6) Riva Star Aqua and ZER. The μTBS test, SEM/EDS, and in situ zymography analyses were conducted at baseline (T0) and after 2 years (T2) in artificial saliva. Data were statistically analyzed ($p < 0.05$).
Results: experimental groups exhibited a significantly lower μTBS than controls (Control>Riva Star = Riva Star Aqua; $p > 0.05$). Aging reduced bond values in all the groups ($p < 0.05$). ER provided higher bond strength than SE ($p < 0.05$). No silver ions were detected in fractured sticks treated with Riva Star and Riva Star Aqua (SEM/EDS). Riva Star significantly reduced MMPs activity ($p < 0.05$) compared to control and Riva Star Aqua ($p < 0.05$).
Conclusions: bonding performance and endogenous enzymatic activity were affected by the 2-step silver-containing solutions tested. The MMPs activity was product-dependent, with Riva Star showing long term inhibitory effects.

STRUCTURAL AND CHROMATIC CHANGES OF TEETH FOLLOWING ORTHODONTIC DEBONDING

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Aim: the objective of this study is to explore the effects of fixed orthodontic appliances on enamel structure by assessing microfractures, surface roughness, and alterations in color.
Methods: this review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines. A systematic search of online databases was conducted using the keywords 'enamel' AND 'orthodontic debonding'. Eligibility criteria included both *in vivo* and *ex vivo* clinical trials conducted on human teeth.
Results: a total of 14 relevant papers were analyzed. Various instruments and techniques were utilized across different studies to assess surface roughness, color change, and surface fractures.
Conclusions: the findings of this study suggest that ceramic brackets may lead to an increase in enamel fractures, particularly during bracket removal. Surface roughness of enamel exhibits variability depending on the adhesive substance and polishing methods used post-removal. Fixed orthodontic appliances could induce changes in enamel color, which may be alleviated by the use of nano-hydroxyapatite or specific polishing techniques. Further research is necessary to identify effective strategies for managing these color changes and improving the overall outcomes of fixed orthodontic treatment.

DEVELOPMENT OF A CUSTOM SOFTWARE FOR DESIGNING AND PRINTING PERSONALIZED RESORBABLE BIOMATERIAL MESH FOR ADVANCED JAW ATROPHIES

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Aim: this study aims to design custom resorbable biomaterial mesh to be used in advanced atrophies using newly developed software, and to print these mesh with a 3D BIOX CellInk printer as an alternative to the titanium mesh traditionally used for Guided Bone Regeneration (GBR).

Methods: seventeen patients with mandibular or maxillary atrophies were selected for this study. Of these, 11 cone beam CT scans were used. Six scans were excluded due to the following reasons: scattering, presence of prostheses over the defect, presence of a cyst-like lesion within the defect, and the inability to import the files into the software. The defects were categorized into horizontal and vertical types. The designs generated by the custom software were then converted into .stl format and further refined using MeshMixer.

Results: custom resorbable biomaterial mesh were successfully designed. However, further development is required, particularly in the following areas: software to optimize the removal of unnecessary parts of the CT scans and a feature to control mesh thickness.

Conclusions: this new approach could eliminate the need for a second surgery to remove the titanium mesh used in traditional GBR procedures. Additionally, alternative materials, such as hydroxyapatite, could be explored for GBR scaffolds.

Further software enhancements and in vivo experiments are necessary to assess the feasibility of this method as a future treatment for maxillary atrophies.

CUSTOMIZED 3D-PRINTED BONE GRAFTS FOR ALVEOLAR RIDGE PRESERVATION: PRELIMINARY HISTOLOGICAL FINDINGS FROM A 3D-PRINTED SYNTHETIC CUSTOMIZED BONE PILOT STUDY

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Aim: this pilot study investigates the clinical application and preliminary outcomes of a 3D-printed synthetic customized bone composed of 30% β-TCP and 70% HA, for socket-preservation.

Methods: a 50-year-old female, with no relevant medical history, presented with pain in the first quadrant. Clinical and radiographic assessments led to an extensive tooth decay diagnosis with an unfavorable prognosis. The authors opted for socket preservation rather than immediate implantology. To evaluate the potential of the 3D-customized bone graft in alveolar preservation, a patient-specific bone graft was designed using a Cone-Beam Computed Tomography (CBCT) and printed with Direct Ink Writing 3DP Technique. The procedure involved an atraumatic, flapless tooth extraction, followed by direct placement of the customized graft. Creos Xenoprotect and keratinized tissue were employed to promote soft tissue healing. At five months post-procedure, a bone biopsy was performed at the preserved site using a trephine. A NobelActive TiUltra® implant was then placed using Dynamic Navigation.

Results: postoperative recovery was uneventful. Radiographic follow-ups at 3, 5, and 20 weeks confirmed stable alveolar bone preservation. Histological analysis revealed well-integrated mineralized tissue with minimal stroma.

Conclusions: histological findings suggest that the bone graft supports bone regeneration while maintaining alveolar ridge morphology. These results, from an ongoing study, indicate promising osteoconductive properties, but further cases and extended follow-up are needed to confirm it.

3D RECONSTRUCTION VS REALITY: MANUAL OR AI FOR ACCURATE TOOTH SEGMENTATION?

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Aim: the aim of this study was to evaluate the accuracy of teeth segmentation process from CBCT scan.
Methods: undamaged extracted teeth were collected from patients who had pre-operative CBCT for clinical purposes. Each extracted tooth underwent a CBCT and a surface scan using a lab scanner. Segmentation was performed on both preoperative and post-extraction CBCT using different methods: Automatic Segmentation (AS) with 3D Slicer, Manual Segmentation (MS) with 3D Slicer and ITK. The segmented models were superimposed on the STL of the scanned tooth (gold standard) to generate six colormaps. Volume and surface area were measured; linear discrepancies were assessed between overlaid models.
Results: this pilot study included 20 teeth. According to qualitative analysis, the root area was found to be the least accurate across all segmentation methods. The AS from the preoperative CBCT demonstrated the closest match to the scanned tooth, with a mean linear difference of 0.19. In surface area, MS with ITK from the tooth's CBCT and AS from the preoperative CBCT showed the highest accuracy (3% and 5% differences, respectively). For total volume, AS from the preoperative CBCT was closer to the scan, with an 8% underestimation. In all analyses, AS from the extracted tooth's CBCT exhibited lower reliability.
Conclusions: automatic dental segmentation from preoperative patient CBCT is a valuable aid in clinical dental practice. Although it is a valid method for generating STL models, the level of accuracy is still low in critical but crucial areas, such as the root apex.

OPTIMIZING DENTAL MATERIAL SELECTION WITH AI: A PRACTICAL APPROACH TO FORMULATING EFFECTIVE PROMPTS

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Aim: Artificial Intelligence (AI) is enhancing diagnostic accuracy, treatment planning and the selection of dental materials. The aim of this study is to evaluate the effectiveness of a guide based on structured prompts in supporting clinicians in the use of AI to make more accurate and informed decisions.
Methods: our study involved 90 dentists aged between 32 and 65 years, assessing their use of and attitude toward AI in the selection of clinical materials. We developed a guide to help formulate effective AI prompts (e.g., "describe", "compare", "specify"). Participants tested the guide using prompts created in accordance with its instructions and subsequently rated its effectiveness on a 1-3 Numerical Rating Scale (NRS). Statistical analysis was performed using Fleiss' kappa.
Results: among the tested group (n = 90), 44% (n = 49) of the dentists considered the guide effective, 25.2% (n = 28) found it only slightly effective, and 11.7% (n = 13) observed no improvement in AI use. The Fleiss' kappa value of -0.0112 indicates poor agreement among the dentists.
Conclusions: the guide may serve as a helpful tool to support better clinical decisions with the aid of AI. Despite a Fleiss' kappa of -0.0112, the guide provides a practical starting point for integrating AI tools into everyday workflows. However, challenges remain, including variability in database updates, not fully representative sample size, reliance on technological devices and individual clinicians' working habits.

EVALUATION OF TWO OPTICAL SCANNING SYSTEM FOR CLEFT LIP AND PALATE ANALYSIS

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Aim: laser scanning and stereophotogrammetry are advanced techniques used to create digital reconstructions and measure the palate in patients with Cleft Lip and Palate (CLP). This study evaluates the reliability of data from both systems by assessing the repeatability and reproducibility of measurements.
Methods: 32 children with unilateral CLP were analyzed using laser scanning and stereophotogrammetry, generating 96 digital models from three different scans. The Vectra 3D Analysis Module (VAM) was used to analyze both the volume and area of the cleft. For area measurements, an automated protocol was applied, while semi-automatic and fully automatic protocols were used for volume. Three methods assessed intra- and inter-operator repeatability, and inter-instrument reproducibility: Technical Error of Measurement (TEM), relative TEM, and the Bland-Altman method.
Results: volume measurements showed lower consistency than area measurements. Protocol 1 for volume quantification was unreliable, with intra-operator and inter-operator errors of 19.1% and 18.8%, respectively. Reproducibility across systems was insufficient, with significant bias. Protocol 2 for volume showed poor reproducibility (<46%) but acceptable bias. In contrast, area measurements showed high reproducibility (87%) and low bias.
Conclusions: area measurements showed high consistency, while volume measurements depended on the protocol used. Protocol 1 had significant errors, while Protocol 2 performed better but still lacked full reproducibility.