Journal of Interdisciplinary Medicine

Volume 8 // Issue 2 // June 2023

Contents

Journal of Interdisciplinary Medicine

Editorial Board

INDEXING

The Journal of Interdisciplinary Medicine is indexed via De Gruyter Open in the following international databases:

- Baidu Scholar
- Celdes
- CNKI Scholar (China National Knowledge Infrastructure)
- CNPIEC
- EBSCO Discovery Service
- Google Scholar
- J-Gate
- Naviga (Softweco)
- Primo Central (ExLibris)
- ReadCube
- Summon (Serials Solutions/ProQuest)
- TDOne (TDNet)
- WorldCat (OCLC)

JIM Journal of Interdisciplinary Medicine

Aims and scope

The Journal of Interdisciplinary Medicine aims to publish top quality papers related to any fields of medicine that present an interdisciplinary dimension.

The journal will mainly focus on recent advances in the field of diagnosis and treatment of the most common situations encountered in the clinical or research practice. Interdisciplinary approaches will be extremely welcomed, presenting new advances in the approach of different pathologies from the perspective of various clinical fields.

The Journal of Interdisciplinary Medicine will publish high-quality basic and clinical research related to interdisciplinary medical fields, in a common approach that will integrate the clinical studies with the pre-clinical work dedicated to the discovery of new mechanisms involved in the development and progression of a large spectrum of diseases.

The journal will try to provide the entire medical community with the perspective of the regional specifics of Central and Eastern European countries. The journal will primarily focus on publishing original research papers, but also other types of materials (such as review articles, case reports, state-of-the-art papers, comments to editor, etc) will be extremely welcomed.

sciendo

Journal of Interdisciplinary Medicine

ORIGINAL RESEARCH

CARDIOVASCULAR IMAGING // 3D PRINTING // MEDICAL TECHNOLOGY

Accuracy of 3D-Printed Models of Aortic Valves – a Comparative Analysis Between Planimetric and Photogrammetric Measurements

Daniel Cernica[1], Diana Opincariu[1], Monica Chițu[1,2,3], István Kovács[1,2,3], Theodora Benedek[1,2,3], Imre Benedek[1,2,3]

[1] "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania
[2] Clinic of Cardiology, Emergency Clinical County Hospital, Târgu Mureș, Romania
[3] Center of Advanced Research in Multimodal Cardiac Imaging, Cardio Med, Târgu Mureș, Romania

CORRESPONDENCE

Diana Opincariu
Str. Gheorghe Marinescu nr. 50
540139 Târgu Mureș, Romania
Tel: +40 265 215 551
Email: diana.opincariu@yahoo.ro

ARTICLE HISTORY

Received: April 15, 2023
Accepted: May 27, 2023

Daniel Cernica • Str. Gheorghe Marinescu nr. 38, 540139 Târgu Mureș, Romania. Tel: +40 265 215 551, Email: daniel.cernica@gmail.com

Monica Chițu • Str. Gheorghe Marinescu nr. 50, 540139 Târgu Mureș, Romania. Tel: +40 372 653 100, Email: iuliachitu@yahoo.com

István Kovács • Str. Gheorghe Marinescu nr. 50, 540139 Târgu Mureș, Romania. Tel: +40 372 653 100, Email: kov_istvan@yahoo.com

Theodora Benedek • Str. Gheorghe Marinescu nr. 50, 540139 Târgu Mureș, Romania. Tel: +40 372 653 100, Email: theodora.benedek@gmail.com

Imre Benedek • Str. Gheorghe Marinescu nr. 50, 540139 Târgu Mureș, Romania. Tel: +40 372 653 100, Email: imrebenedek@yahoo.com

ABSTRACT

Background: 3D printing has changed the paradigm of personalized medicine. Similarly to fingerprints, there are no two identical hearts; consequently, in cardiology, diagnosis and treatment, either medical, interventional or surgical, must be individualized according to the specific problem of a particular patient. The **aim** of this proof-of-concept study was to analyze two measurement methods, the planimetric and the photogrammetric method, in the process of creating a 3D-printed model from cardiac computed tomography angiography images and to evaluate the accuracy of an aortic valve anatomical model. **Material and methods:** Cardiac computed tomography images, obtained from 20 patients with severe aortic stenosis, underwent stereolithographic reconstruction using 3D Slicer to create digital 3D models of the aortic valves. Serial measurements of six key elements of the aortic valvular apparatus were measured on the 3D model and compared to the measurements taken on the 2D computed tomography images. **Results:** The differences between the two measurement methods were sub-millimetric in case of the left ventricular outflow tract and the sinotubular junction, and 1.386 mm for the left sinus of Valsalva ($p = 0.0412$), 0.3476 mm for the right sinus of Valsalva ($p = 0.1874$), and 0.6905 mm for the non-coronary Valsalva sinuses ($p = 0.1353$). Sinus heights were also similar, with a difference of 0.0119 mm ($p = 0.6521$). **Conclusion:** In this study, the results of digital photogrammetry were superimposable to those of computed tomography scan measurements. The accuracy of each 3D-printed model depends on geometric complexity, the level of training of the personnel, and on the resources of each 3D printing laboratory.

Keywords: validation, 3D printing, aortic valve, additive manufacturing, anatomical models

INTRODUCTION

Similarly to fingerprints, there are no two identical hearts. Consequently, diagnosis and treatment — either medical, interventional, or surgical — should be individualized according to the specific problem of a particular patient. Cardiovascular imaging has gone through an impressive technological progress in recent years, 3D-printed cardiac imaging currently being the cornerstone of the modern management of patients with complex cardiovascular pathology. 3D printing has opened up new perspectives and opportunities in interventional and surgical fields.[1] The applicability of 3D printing is extensive and includes congenital heart disease, mitral and aortic valvulopathies (e.g., transcatheter aortic valve implantation, TAVI), valvular prosthetics (closure of paraprosthetic leaks), and structural heart disease (closure devices for ventricular defects or closure of the left atrial appendage).[2] The preprocedural work-up could benefit substantially from 3D printing, especially by simulating the implantation of devices of different sizes. The ongoing French observational register FFPP-Print has highlighted its benefits by demonstrating shorter operating times, a significant reduction in the number of prostheses used for each patient, and the ability to assess the risk of complications. Apart from complex procedures, 3D printing can find its utility in anticipating rare but serious complications that can occur during transcatheter interventions such as TAVI. Studies clearly show that morbidity and mortality rates are lower than with conventional surgery for high- or intermediate-risk patients, but this risk should be analyzed individually for each patient.[3]

In this proof-of-concept study, we aimed to investigate whether 3D models created from cardiac computed tomography (CT) images could have comparable dimensions to the native valve apparatus and to study the feasibility of using 3D-printed aortic valves in simulations of transcatheter implantation.

MATERIAL AND METHODS

Study participants

This was a cross-sectional observational study that included 20 patients with severe aortic stenosis who underwent preprocedural planning for TAVI. The study was conducted from January 2020 to December 2021. Exclusion criteria included the presence of a high degree of aortic valve calcifications, irregular heart rate or inability to achieve a heart rate < 65 bpm, or any other conditions that may have interfered with image acquisition. Clinical, echocardiographic, and laboratory examinations, as well as contrast-enhanced cardiac CT and computed angio-tomography of the lower limbs were performed for all patients. For cardiac CT image acquisition, a retrospective ECG-gated scanning protocol was used, with intravenous high-concentration iodine contrast agent, at a heart rate of <65 bpm. Patients with increased heart rate were administered beta blockers or ivabradine before scanning. All CT scans were performed using SOMATOM Definition 128-slice CT equipment (Siemens Healthcare, Germany). Patients with inadequate valvular and vascular anatomy for TAVI, as well as sub-optimal quality of CT images were excluded. All study procedures were performed according to good clinical practice (GCP) guidelines and the Declaration of Helsinki, and were approved by the ethics committee of the institution where the study was conducted. All study participants signed an informed consent prior to enrollment.

Stereolithographic model generation– from CT scan to digital model

As part of an ongoing research project at the "George Emil Palade" University of Medicine, Pharmacy, Science and Technology of Târgu Mureș, patient-specific digital models were generated to determine the feasibility of 3D-printed aortic valve models for preprocedural planning.

Image post-processing was performed for all individual CT images to obtain the stereolithographic reconstruction of the models used for 3D printing. The Digital Imaging and Communications in Medicine (DICOM) datasets that were analyzed had a slice thickness of 0.60 mm using a medium smooth kernel. All image reconstructions were performed by a single analyst using 3D Slicer, an open-source offline image computing platform for image analysis and scientific visualization.

The first step in the stereolithographic processing of 3D models consisted in integrating the imaging data into 3D Slicer and delineating the regions of interest (ROIs) on anatomical structures (Figure 1). The second step consisted in the so-called 'segmentation', based on the intensity of the studied structures, measured in Hounsfield units (HU). For example, for cardiac structures, a range of 230 HU to 15 HU was used. The third step consisted in rapid prototyping, achieved by placing spatial markers at the level of fluid structures called 'blood pool', which enabled us to differentiate the contrast agent from muscle or fibrous structures by subtraction based on Boolean algorithms (Figure 2). Segmentation time was approximately 60–80 min per model, as described previously.[4] The fourth step consisted in transforming DICOM files into .STL (Standard Tessel-

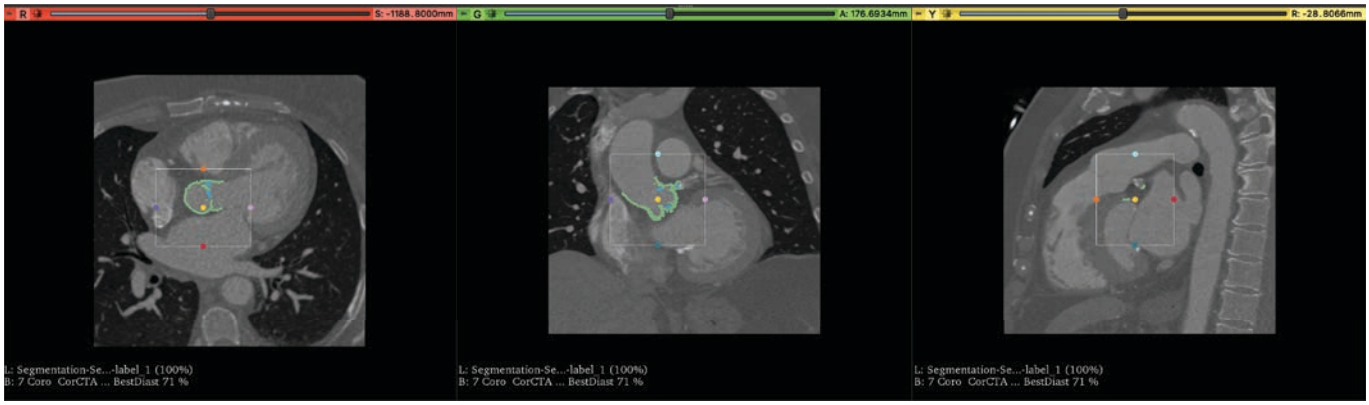

FIGURE 1. Segmentation of ROIs at the level of the aortic valve (green)

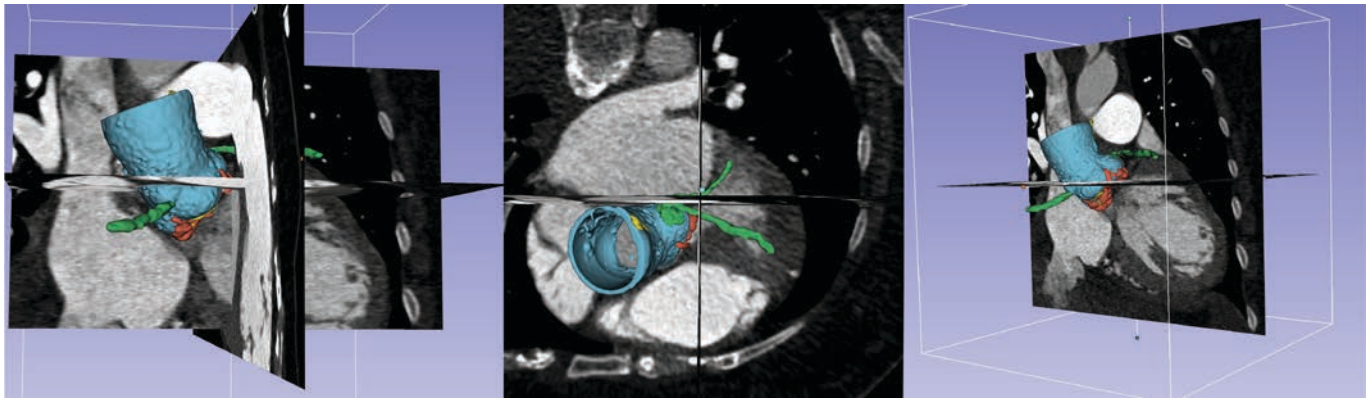

FIGURE 2. Segmentation – selection of "blood pool" at the level of the aortic valve (green)

lation Language) files used in 3D printing. The 3D digital dataset was converted into a virtual 3D model (Figure 3), and automatic or manual cropping functions ('crop mask') were applied. The 3D model obtained after the segmentation process consisted of a structure of triangular facets with semi-finished surface.

Although the model processed in STL format could have been printed at this stage, anatomical models generally require an additional stage. Optimization of the model can be achieved using computer-aided design (CAD) in programs such as Autodesk Meshmixer (Autodesk Inc., San Rafael, CA, USA). This step involved remeshing, a process that optimizes the model's geometry and density, and tessellation, a process that optimizes the triangular facets of the mosaic that make up the digital 3D model (Figure 4). Post-processing time using CAD software was approximately 60 min per model.

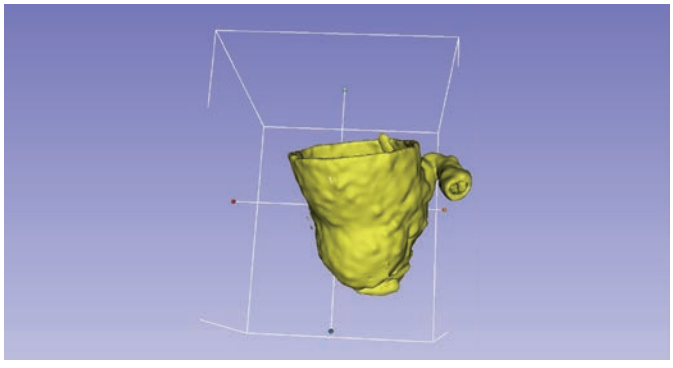

FIGURE 3. Digital 3D model of the aortic valve, ascending aorta and coronary arteries, lateral view

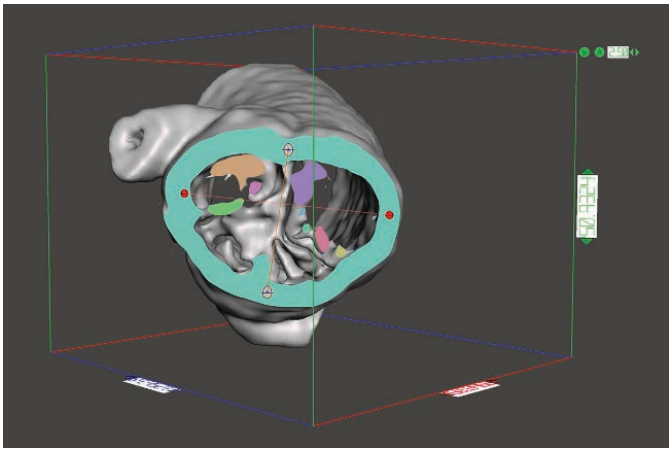

FIGURE 3. Digital 3D model of the aortic valve, ascending aorta and coronary arteries, lateral view

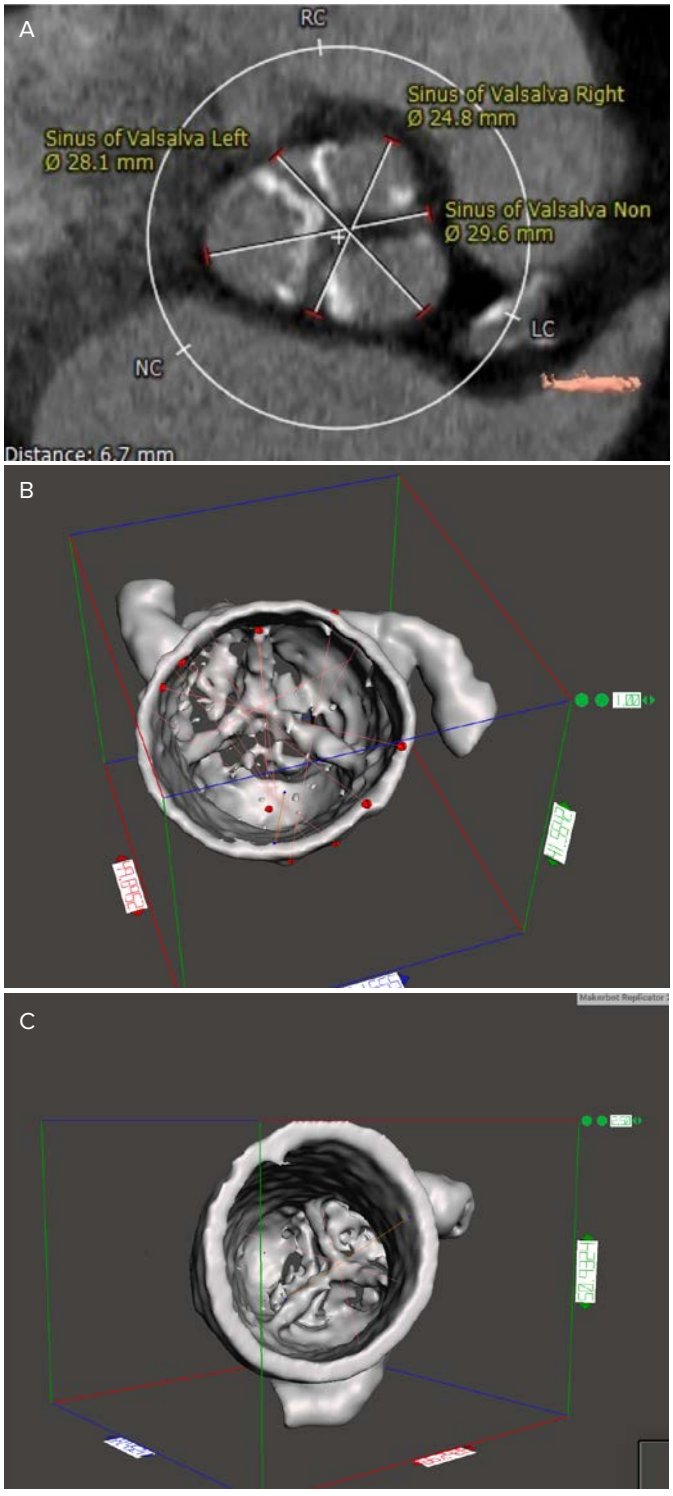

FIGURE 5. Measurement methods: planimetric (**A**) and photogrammetric (**B**, **C**)

In the final step, a digital snapshot of the CT scans, along with an embedded report of the measurements, was exported to Autodesk Meshmixer and overlaid on the STL file for reference to ensure that the STL file features were measured at the same location as the digital measurements. We identified six elements of the aortic valvular apparatus that were accessible to be measured and obtained a total of 13 linear measurements. The following parameters were measured on both the multiplanar dataset CT model and the STL digital model of the aortic valve complex:

- the origin of the left coronary artery (LCA) and the right coronary artery (RCA);
- right, left and noncoronary sinuses of Valsalva;
- the diameter of the aortic annulus;
- the left ventricle outflow tract (LVOT);
- the ascending aorta;

Landmark measurements on digital models

After finalizing image post-processing and the 3D-printed models, we conducted serial measurements of several parameters used for TAVI preprocedural planning. As an initial step, we performed one set of measurements on the 2D cardiac CT dataset using direct planimetry and multiplanar reconstruction in Syngo.via software (Siemens Healthcare, Germany), during mid-systole, at approximately 240–330 ms (Figure 5).

To ensure accuracy, all anatomic analyses were performed using the center line of the lumen as reference. Then, the effective measurement was calculated as the arithmetic mean of minimum and maximum values.

We measured key elements in the STL file in a 3D AutoCAD program and used vertex-to-vertex analysis to compare them to digital measurements of 3D aortic valve models (photogrammetric method).

Statistical analysis

Statistical analysis of the recorded data was performed using GraphPad Prism version 8.4.3 (GraphPad Software, San Diego, CA, USA).

After testing for normality using the D'Agostino–Pearson algorithm, quantitative data were expressed as mean ± standard deviation. Paired data were compared using Student's t test or the Wilcoxon matched-pairs signed rank test, and differences between means were calculated using the Bland–Altman method, with 95% confidence intervals.

Intra-class correlation was analyzed by calculating the Pearson correlation coefficient for normally distributed data and the Spearman coefficient for non-normal distribution. A p value of <0.05 was considered statistically significant.

TABLE 1. General clinical characteristics of the study population

Parameter	Value
Age, years	72.43 ± 4.96
Males, n (%)	10 (52%)
Smoking, n (%)	8 (40%)
Arterial hypertension, n (%)	15 (75%)
Obesity, n (%)	11 (55 %)
Dislipidemia, n (%)	5 (25%)
Diabetes mellitus, n (%)	6 (30 %)
Chronic kidney disease, n (%)	9 (45 %)
Coronary artery disease, n (%)	17 (85%)
Left ventricular systolic dysfunction, n (%)	5 (25%)
Bicuspid valve stenosis, n (%)	2 (10 %)
Degenerative aortic stenosis, n (%)	18 (90 %)
Aortic regurgitation, n (%)	15 (75 %)

RESULTS

The general characteristics of the study population are listed in Table 1. Mean age was 72.43 ± 4.96 years, the majority of the patients were men, and the most frequent cause of aortic stenosis was degenerative aortic stenosis, followed by congenital bicuspid valve with degenerative features.

The measurement of digital and 3D-printed models

For the evaluation of the valvular and perivalvular apparatus, anatomical structures were analyzed separately. The comparative analysis of planimetric and photogrammetry measurements, as well as the differences between measurement values provided by the two methods are listed in Table 2. The difference regarding the height of the coronary ostium was 0.0238 mm for the LCA (p = 0.9022) and 0.2095 mm for the RCA (p = 0.4349). The measurements of the aortic annulus showed a difference between minimum values of 1.0000 mm (p = 0.1033) and between maximum values of 0.5700 mm (p = 0.3315). Although the differences between planimetric vs. photogrammetric diameters of the sinuses of Valsalva were larger for the left sinus of Valsalva (1.386 mm, p = 0.0412), they were not significant for the right sinus of Valsalva (0.3476 mm, p = 0.1874) and the noncoronary sinus of Valsalva (0.6905 mm, p = 0.1353). Sinus heights were similar with the two methods, with a difference of 0.0119 mm (p = 0.6521). Ascending aorta measurements were also similar, with a difference between minimum values of –0.0523 mm (p = 0.7854) and between maximum values of 0.2571 mm (p = 0.3373). Measurements of the sinotubular junction were comparable, with a difference between minimum values of 0.0285 mm (p = 0.9564) and between maximum values of 0.3714 mm (p = 0.3273). LVOT assessment showed no differences between the two methods, with a difference between minimum values of 0.7238 mm (p = 0.1381) and between maximum values of 0.3714 mm (p = 0.3315).

The reliability of 3D models – intraclass correlation coefficients

The intraclass correlation coefficients were statistically significant for all measurements, indicating good reliability of 3D CT measurements (Table 3). The highest correlation

TABLE 2. Comparison of planimetric and photogrammetric measurements

Landmark	Planimetric (mean ± SD)	Photogrammetric (mean ± SD)	Difference between means	p value
LCA height, mm	14.71 ± 3.543	14.69 ± 3.676	0.0238	0.9022
RCA height, mm	17.50 ± 3.665	17.29 ± 3.694	0.2095	0.4349
Left sinus of Valsalva diameter, mm	32.93 ± 4.289	31.55 ± 4.822	1.3860	0.0412
Right sinus of Valsalva diameter, mm	30.11 ± 4.320	29.76 ± 4.266	0.3476	0.1874
Noncoronary sinus of Valsalva diameter, mm	32.77 ± 3.827	32.08 ± 3.231	0.6905	0.1353
Ascending aorta min diameter, mm	34.02 ± 3.414	33.97 ± 3.532	0.0524	0.7854
Ascending aorta max diameter, mm	35.52 ± 3.301	35.26 ± 3.663	0.2571	0.3373
Sinotubular junction min diameter, mm	29.24 ± 4.792	29.21 ± 5.251	0.0286	0.9564
Sinotubular junction max diameter, mm	30.21 ± 4.170	29.84 ± 4.096	0.3714	0.3273
Aortic annulus min diameter, mm	21.38 ± 3.329	22.38 ± 4.577	1.0000	0.1033
Aortic annulus max diameter, mm	27.49 ± 3.450	28.07 ± 3.984	0.5762	0.3315
LVOT min diameter, mm	21.32 ± 4.962	22.04 ± 5.516	0.7238	0.1381
LVOT max diameter, mm	28.83 ± 3.647	29.20 ± 4.232	0.3714	0.4093
Sinus of Valsalva height, mm	24.55 ± 4.979	24.43 ± 4.519	0.1190	0.6521

min, minimum; max, maximum

TABLE 3. The intraclass correlation between measurements obtained from the two methods

Landmark	Correlation coefficient (r)	95% confidence interval	p value
LCA height, mm	0.9712	−0.4230 to 0.3753	0.9022
RCA height, mm	0.9464	−0.7580 to 0.3390	0.4349
Left sinus of Valsalva diameter, mm	0.8022	−2.710 to −0.06124	0.0412
Right sinus of Valsalva diameter, mm	0.9631	−0.8788 to 0.1835	0.1874
Noncoronary sinus of Valsalva diameter, mm	0.8472	−1.616 to 0.2350	0.1353
Ascending aorta min diameter, mm	0.9692	−0.4483 to 0.3436	0.7854
Ascending aorta max diameter, mm	0.9460	−0.8027 to 0.2884	0.3373
Sinotubular junction min diameter, mm	0.8932	−1.105 to 1.047	0.9564
Sinotubular junction max diameter, mm	0.9161	−1.143 to 0.4001	0.3273
Aortic annulus min diameter, mm	0.8146	−0.2222 to 2.222	0.1033
Aortic annulus max diameter, mm	0.7544	−0.6314 to 1.784	0.3315
LVOT min diameter, mm	0.9214	−0.2535 to 1.701	0.1381
LVOT max diameter, mm	0.8789	−0.5479 to 1.291	0.4093
Sinus of Valsalva height, mm	0.9731	−0.6616 to 0.4235	0.6521

min, minimum; max, maximum

coefficient was obtained for the height of the sinus of Valsalva (Figure 6).

DISCUSSION

This study was a proof of concept that aimed to compare the planimetric and the photogrammetric measurement method in the process of creating a 3D-printed model from cardiac CT angiography images. Another aim of the study was to verify the accuracy of each step of this process as part of quality assurance and to discuss the challenges encountered with each method. The main findings of the study are that 3D-printed models provide a feasible, non-invasive method to assist the 3D visualization of patient-specific aortic root anatomy, and that 3D modeling represents a new opportunity to plan in situ device placement during transcatheter aortic valve replacement. Furthermore, 3D modeling may complement traditional methods

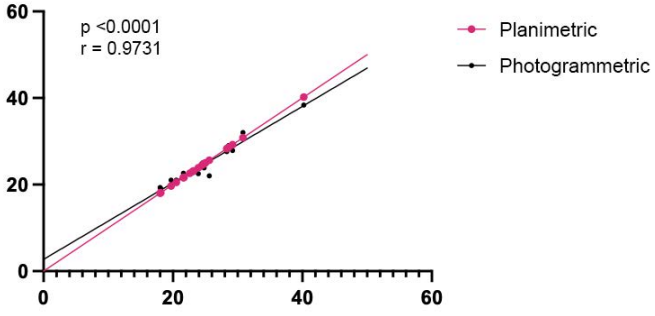

FIGURE 6. Correlation between the two measurement methods for the height of the sinus of Valsalva

used to predict and potentially avoid complications such as pulmonary artery rupture.[5]

One potential source of error is related to DICOM image registration from cardiac CT, as landmark points can vary due to observer interpretation or scan quality. Improvements in image acquisition and observers specialized in cardiac radiology can mitigate this barrier. Our study has demonstrated that the processing steps can be done with meaningful levels of accuracy. The results showed high correlation coefficients and infra-millimetric differences between the planimetric and photogrammetric method, which were not significant statistically. The results also revealed that the 3D printing of aortic valve models can be precise but with limitations related to inter-observer variability,[6] which can lead to clinically important variations in geometry and dimensions. Potential sources of errors should be flagged to increase precision during the process of generating cardiovascular models. In our study, an important source of error was segmentation, as we encountered issues in delineating structures of interest and in using ROI tools such as dynamic grow mask or crop tools. However, further research, and particularly the use of AI, may improve semi-automatic functions and lead to the development of algorithms that delineate ROI more precisely. Other potential error sources were related to STL file processing and critical non-linear measurements on the 3D digital model. Digital analysis can mitigate these barriers using point cloud and mesh techniques, or colorimetric 3D maps when 3D-printed models are used.[7]

Another important limitation of our study is the fact that the entire process has been conducted by a single clinician.

In future studies, at least two observers should perform the image post-processing to identify key inter-observer differences. Furthermore, the number of patients included in the study was low. Increasing their number can decrease the risk of errors and improve the development of solutions regarding the workflow of 3D printing aortic valves.

In a study conducted by Fourie *et al*. on 3D models created from CT imagery, the authors observed important differences in the dimensions of 3D-printed models between clinicians and emphasized the importance of inter-observer errors and of semi-manual processing during the entire workflow.[8] Furthermore, each stage in the process of 3D-printing a cardiovascular model is susceptible to errors. Similar results were obtained in a study conducted by Santana *et al*., who observed discrepancies in the interpretation and subsequent analysis of medical images by different observers.[9] Considering the continuous exposure to sources of error, it is mandatory to judiciously monitor potential errors generated by semi-automated processing. Cross-check points should be inserted in the intermediate stage, overlaying the generated digital model to original DICOM data presented in the current study. Future research is needed to determine the accuracy of 3D-printed aortic templates.[10]

CONCLUSIONS

In cardiovascular valve diseases, the precision of 3D-printed models is an important element that can contribute to increasing quality of procedural simulations and predicting potential complications. Various methods were studied for assessing the accuracy of 3D printing, each with strengths and weaknesses. The selected method of assessment may require a 3D-printing team with expertise in radiological field and in processing and post-processing of additive manufacture field.

CONFLICT OF INTEREST

Nothing to declare.

FUNDING

This work was supported by the "George Emil Palade" University of Medicine, Pharmacy, Science and Technology of Târgu Mureș Research Grant number 510/1/17.01.2022, and the Doctoral School of "George Emil Palade" University of Medicine, Pharmacy, Science and Technology of Târgu Mureș.

REFERENCES

1. Vukicevic M, Mosadegh B, Min JK, Little SH. Cardiac 3D Printing and its Future Directions. *JACC Cardiovasc Imaging*. 2017;10:171–184.
2. Levin D, Mackensen GB, Reisman M, McCabe JM, Dvir D, Ripley B. 3D Printing Applications for Transcatheter Aortic Valve Replacement. *Curr Cardiol Rep*. 2020;22:23.
3. Jiménez Restrepo A, Chahal D, Gupta A, et al. Roadmap to Success: 3D Printing in Pre-Procedural Planning. *J Am Coll Cardiol Case Rep*. 2020;2:358–360.
4. Cernica D, Benedek I, Polexa S, Tolescu C, Benedek T. 3D Printing-A Cutting Edge Technology for Treating Post-Infarction Patients. *Life (Basel)*. 2021;11:910.
5. Shirakawa T, Yoshitatsu M, Koyama Y, Mizoguchi H, Toda K, Sawa Y. 3D-printed aortic stenosis model with fragile and crushable calcifications for off-the-job training and surgical simulation. *Multimed Man Cardiothorac Surg*. 2018;2018.
6. Odeh M, Levin D, Inziello J, et al. Methods for verification of 3D printed anatomic model accuracy using cardiac models as an example. *3D Print Med*. 2019;5:6.
7. Nulty A. A comparison of trueness and precision of 12 3D printers used in dentistry. *BDJ Open*. 2022;8:14.
8. Fourie Z, Damstra J, Schepers RH, Gerrits PO, Ren Y. Segmentation process significantly influences the accuracy of 3D surface models derived from cone beam computed tomography. *Eur J Radiol*. 2012;81:e524–530.
9. Santana RR, Lozada J, Kleinman A, Al-Ardah A, Herford A, Chen JW. Accuracy of cone beam computerized tomography and a three-dimensional stereolithographic model in identifying the anterior loop of the mental nerve: a study on cadavers. *J Oral Implantol*. 2012;38:668–676.
10. Rynio P, Wojtuń M, Wójcik Ł, Kawa M, Falkowski A, Gutowski P, Kazimierczak A. The accuracy and reliability of 3D printed aortic templates: a comprehensive three-dimensional analysis. *Quant Imaging Med Surg*. 2022;12:1385–1396.

sciendo

Journal of Interdisciplinary Medicine

ORIGINAL RESEARCH

ORTHOPEDICS // BIOMATERIALS

Biomechanical Uniaxial Analysis of Porcine Tendon in the CellScale BioTester® 5000

Alexandru Fofiu[1,2], Emil M. Arbănaşi[3,4,5,6], Robert G. Tripon[7], Shuko Suzuki[8], Traian V. Chirilă[2,8,9,10,11], Tiberiu Bățagă[12]

[1] Department of Orthopedics-Traumatology, Emergency County Hospital Bistrița, Bistrița- Năsăud, Romania
[2] Faculty of Medicine, "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania
[3] Doctoral School of Medicine and Pharmacy, "George Emil Palade" University of Medicine, Pharmacy, Sciences and Technology, Târgu Mureș, Romania
[4] Department of Vascular Surgery, "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania
[5] Clinic of Vascular Surgery, Mureş County Emergency Hospital, Târgu Mureș, Romania
[6] Center for Advanced Medical and Pharmaceutical Research (CCAMF), "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania
[7] Department of Ophthalmology, "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania
[8] Queensland Eye Institute, South Brisbane, Queensland, Australia
[9] School of Chemistry and Physics, Queensland University of Technology, Brisbane, Queensland, Australia
[10] Australian Institute of Bioengineering and Nanotechnology (AIBN), University of Queensland, St Lucia, Queensland, Australia
[11] School of Molecular Science, University of Western Australia, Crawley, Western Australia, Australia
[12] Department of Orthopedics-Traumatology, "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania

CORRESPONDENCE

Traian V. Chirilă
Queensland Eye Institute
140 Melbourne Street, South Brisbane
Qld. 4101, Australia
Tel: +61 7 3239 5000
Email: traian.chirila@qei.org.au

ARTICLE HISTORY

Received: June 20, 2023
Accepted: June 27, 2023

Alexandru Fofiu • Str. Gheorghe Marinescu nr. 50, 540136 Târgu Mureș, Romania. Tel: +40 746 022 992, Email: alexandrufofiu@gmail.com

Emil Marian Arbănaşi • Str. Gheorghe Marinescu nr. 50, 540136 Târgu Mureș, Romania. Tel: +40 758 530 111, Email: emil.arbanasi@umfst.ro

Robert G. Tripon • Str. Gheorghe Marinescu nr. 50, 540136 Târgu Mureș, Romania. Tel: +40 749 951 988, Email: robert.tripon@umfst.ro

Shuko Suzuki • Queensland Eye Institute, 140 Melbourne Street, South Brisbane, Qld. 4101, Australia. Tel: +61 7 3239 5000, Email: shuko.suzuki@qei.org.au

Tiberiu Bățagă • Str. Gheorghe Marinescu nr. 50, 540136 Targu Mures, Romania. Tel: +40 745 607 046, Email: tiberiu.bataga@umfst.ro

ABSTRACT

Background: The study was aimed to evaluate whether a mechanical biaxial tester can be used in a uniaxial mode to evaluate the mechanical properties of tendons. **Materials and methods:** The study was carried out on specimens of porcine superficial digital flexor tendon (n = 9). The mechanical properties (elastic modulus, and stress at 15% strain) were measured two times consecutively in the uniaxial mode with the BioTester® 5000 (CellScale) equipment. **Results:** Values of 0.313 ± 0.096 MPa for the elastic (Young's) modulus and of 0.702 ± 0.174 MPa for the stress (at 15% strain) were measured, indicating that the porcine superficial digital flexor tendon is not a strong tendon. **Conclusions:** When suitable specimens cannot be obtained for a biaxial evaluation, tendons can be evaluated mechanically in the BioTester® 5000 employing the uniaxial mode.

Keywords: tendons, mechanical properties, porcine SDFT, uniaxial testing

INTRODUCTION

Historically, the biomechanics of ligaments and tendons was a field of experimental physiology, which, before the 1970s, has been associated with only minor developments in spite of genuine interest from orthopedic surgeons. Today the situation is very much different, as illustrated in the number of available publications, although the outputs were estimated with rather great variability. For instance, according to such an estimation,1 over 4,000 papers have been published on the topic within the 2000–2010 decade. A PubMed search in March 2023 using "biomechanics of tendons and ligaments" as a search term resulted in 3,680 publications since 1963, while a recent search on Google Scholar using the same term and time range provided around 19,000 entries (a figure likely to be seriously affected by repetitions). Regardless, only a small number of these publications have included actual numerical data for the relevant mechanical characteristics. A variety of mechanical parameters have been reported including yield or ultimate tensile stress (strength) (henceforth, UTS), yield or ultimate load, yield or ultimate elongation (strain), elastic (or Young's) modulus (henceforth, YM), stiffness, and toughness. They were evaluated mostly in the tensile mode, but also in the compressive or shear modes. An excellent summary and analysis of the measured ex vivo biomechanical tensile properties in human and animal tendons and ligands, reported between 1976 and 2015, has placed the recorded values within the 2–230 MPa range for UTS; 1.3–3,000 MPa for YM; and 2–1,100 N/mm for stiffness.2 By any standard, these are exceedingly broad data distributions, arising from causes such as different origins of tendons, different harvesting and conditioning procedures of tendons prior to testing, sample slipping and distortion during testing, stress concentrations occurring in the tissue, and a great diversity of the evaluation instrumentation and techniques. Slippage of tendon samples from the clamps during measurements is still the major factor in preventing reliable and reproducible results to be generated by the testing machines, and a large assortment of clamping and mounting systems have been proposed including frozen clamps ('cryojaws'), or even cyanoacrylate glues.

The earliest significant mechanical evaluation of human tendons was carried out by Guillaume Wertheim,3 who presented his results in 1846 to the French Academy of Sciences. He measured the YMs and UTSs not only for tendons but also for bones, nerves, blood vessels, and muscles. Wertheim was an outstanding experimentalist, and the values published by his team for certain tissues could still be seen in reference texts 100 years after his premature death. For the mechanical characteristics of human tendons, Wertheim measured values for YM (then termed as 'coefficient d'élasticité') between 1.26 and 1.97 GPa and for UTS ('cohésion') between 41 and 102 MPa in five human plantaris tendons and one flexor longus tendon harvested post mortem. In 1936, in an introduction to his own report,4 Cronkite discussed Wertheim's results and also cited from indirect sources the work with human tendons of other early investigators including Valentin (1847), who measured values of UTS between 15 and 22 MPa for palmaris longus and plantaris tendons; Rauber (1876), who measured an UTS of 68 MPa in an unspecified tendon; and Triepel (1902), who measured an average value of 44 MPa for UTS in a plantaris tendon specimen harvested during surgery. In a major study,4 Cronkite himself carried out tensile evaluation on 294 human tendons harvested post mortem, using a mechanical tester for solid materials with a custom-made clamping system. The average UTS of the tendons in different cadavers varied from 60 to 124 MPa, and a large variation of up to 200% between the minimum and maximum values in the same body. He concluded that "it is obviously futile to establish a norm for tensile strength for tendons in general", a conclusion still valid today. Cronkite also noted that the strength of fresh specimens did not differ substantially from that of embalmed (fixed) tendons. Further work carried out during the 1960s was reviewed in studies by Harris et al.5,6 at Tulane University, in which they also reported findings on human tendons, using a custom-made optical-mechanical tester and a more advanced statistical processing of the data points obtained from 30 embalmed plantaris tendons5 and 54 unembalmed specimens removed from amputated lower limbs6. In the first study,5 the values measured for UTS were between 73 and 147 MPa (average 98 MPa), and for YM were ~1.24 GPa for wet specimens and ~2.76 GPa for dried specimens. In their second study,6 the average UTS measured for extensor tendons was 92.3 MPa, while for flexor tendons was 75.5 MPa. However, the YM was variable, and no clear pattern could be defined upon increasing stress. LaBan was the first investigator to associate the tendon's tensile strength to its collagen fibrillar organization.7 Using a custom-made tensile device combined with a microscope, he found that in a canine calcaneal tendon, no tearing of collagen fibers could be noted at a stress below 6 MPa. Within this range, the tendon displayed viscoelastic properties rather similar to those of certain synthetic polymers.

Since then, an impressive variety of measuring techniques and devices have been used to evaluate the mechanical properties of tendons. Over the last few decades, the uniaxial machines (e.g., Instron® or Zwick/Roell testers) became popular and are still used extensively. In the uni-

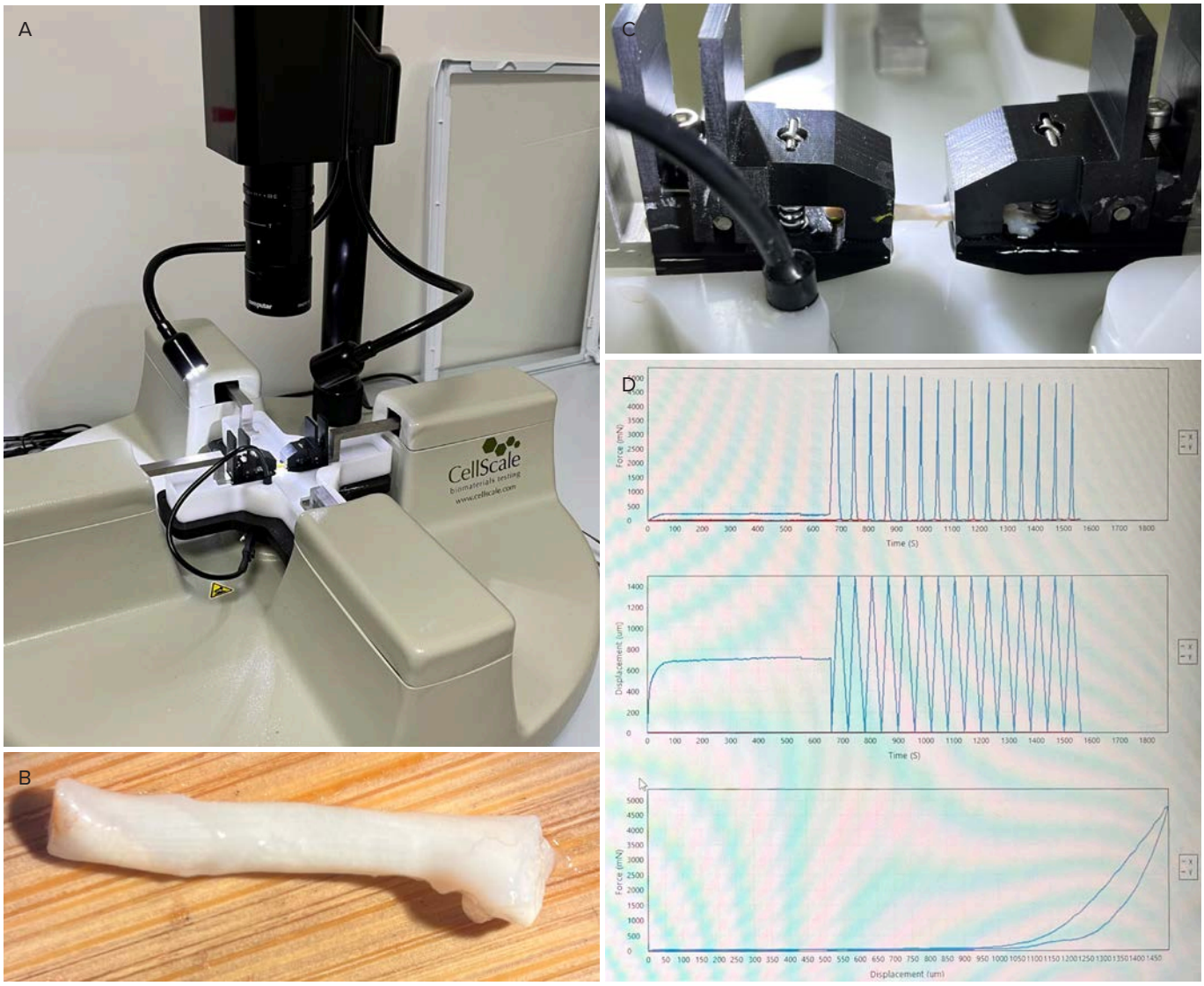

FIGURE 1. **A** – The BioTester® 5000 instrument; **B** – An excised segment of the porcine superficial digital flexor tendon; **C** – A tendon specimen mounted in the tester by clamping; **D** – Graphical output from the tester's software corresponding to the main evaluation stages

axial test, the sample is subjected to a force along one direction only until failure occurs due to stretching (or other type of force). In recent years, the biaxial testing started to be employed in many laboratories. In the biaxial test, the sample is stretched along two distinct perpendicular directions. This method is recommended for anisotropic materials such as soft biological tissues; however, it is difficult sometimes to find tendons that can be excised and fashioned into square plane slabs as required by the cruciform clamping system of the testing instrument. In such cases, the biaxial tester could be used in the uniaxial mode, and an example of such versatile machine is the BioTester® 5000 manufactured by the CellScale company (Waterloo, ON, Canada) (Figure 1A).

In the present study, we have evaluated specimens of the porcine superficial digital flexor tendon (SDFT), which are elliptical in cross-section and could not be fashioned into square slabs with a width allowing the cruciform clamping normally required for biaxial testing.

MATERIALS AND METHODS

Materials

The study was carried out on nine individual segments of the SDFT retrieved post mortem from three pigs (species Sus domesticus) of the breed White Large, 10 months old, procured from a local authorized slaughterhouse (Agro-

Ardeal S.R.L., Orheiu Bistriţei, Bistriţa-Năsăud). The animals were sacrificed for commercial purposes, and the tendons would have been discarded if not used in this study. Only the tendons from the frontal legs of the animals have been harvested, having an average length of 19.39 ± 1.49 mm and an elliptical circumference with an average diameter of 2.95 ± 0.32 mm (Figure 1B). After harvest, the tendons were stored at –20°C. Phosphate buffered saline (PBS) was supplied by Lonza (Verviers, Belgium).

Uniaxial analysis in the CellScale BioTester® 5000

The BioTester® 5000 (CellScale) included four actuators (only two actuators being used for uniaxial testing), load cells, systems of rakes with tines and hooks (BioRakes®) for specimen mounting, clamp sample mounting systems, and user interface software for simple or multi-modal testing with real-time feedback. The 23-N load cell was used in our experiments.

After harvesting, the tendon segments were cut with a scalpel into specimens of approximately 13 mm in length, which were stored in PBS at room temperature prior to measurements. The thickness of each specimen was measured in triplicate with a caliper by the same person and the values averaged for further data processing.

The specimens were clamped along the longitudinal axis between two opposite arms of the instrument. A working distance of 10 mm was set as the initial distance between the two arms. The specimens were inserted manually between the clamps (Figure 1C), preferably by the same person to minimize possible bias related to the act of insertion.

To ensure that the samples were tensioned, the evaluation started with a tensile preloading for 60 s until a force of 230 mN was attained, followed by a 10-min hold period at this force. After this preconditioning period, 15 cycles were initiated, each consisting of a stretch period (where the maximum tension was set at 15% of the initial length of the specimen), a deformation of 1% per second, and a relaxation period, as illustrated in a typical graphical output of the BioTester (Figure 1D). The series of nine specimens was evaluated two times in the tester.

By employing the LabJoy 2.0 software (CellScale, Waterloo, ON, Canada), the raw data were generated in an Excel file, ready to be processed for calculating strength and YM for each specimen.

Statistical analysis

The data were plotted as mean values ± standard deviation. For the statistical comparison of values, GraphPad® Prism software (version 6.0) was used, with application of the Wilcoxon matched-pair rank test for continuous data (for n = 9).

RESULTS

The set of nine tendon specimens was evaluated in the BioTester two times consecutively with an aim of assessing stability of the measuring technique and reproducibility of results. The results were presented as bar graphs in Figure 2. Neither the YM (Figure 2A), nor the strength (at 15% strain) (Figure 2B) sets of values showed any significant difference between the two measurements, as reflected in the high p values.

DISCUSSION

Most of what has been reported so far on the mechanical properties of porcine tendons was summarized in a recent review.8 Following the information provided in this source, the values reported for porcine digitorum profundum flexor and extensor tendons included ~0.8 to ~1.7 GPa for YM, and ~40 to ~90 MPa for UTS, while for the porcine Achilles tendon values of 248 to 409 MPa for YM and 42 to 76 MPa for UTS have been also reported.

Many investigators expressed their results in units of maximum load (N) or stiffness (N/mm). For instance, Domnick et al.9 compared porcine flexor digitorum profundus tendons with human cadaveric semitendinosus tendons in a Zwick/Roell uniaxial tester using frozen clamps to prevent slippage. There were no significant differences between the two sets regarding stiffness (porcine ~211 N/mm, human ~208 N/mm), but the maximum load was higher for porcine tendons (~1.8 kN) as compared with human (~1.4 kN). It was concluded that the use of porcine flexor tendons as grafts in human surgery is justified mechanically. To compare these data with our findings, it is necessary to know the dimensions of the samples in order to render the reported parameters into units of YM and stress.9 There is only one study published on porcine SDFT, reporting maximum loads of ~230 N and stiffness of ~56 N/mm.10 Again, these results cannot be compared with our results due to lack of dimensional details for the specimens.

When it was possible to compare our results for YM and stress with other reported values (less than 0.4 MPa for YM, and less than 0.9 MPa for stress, see Figure 2), it could be seen that the former were much lower. Nonetheless, this can be explained by the fact that the existing literature data refer to the porcine flexor digitorum profundum (deep) tendon or to the extensor tendons, which are

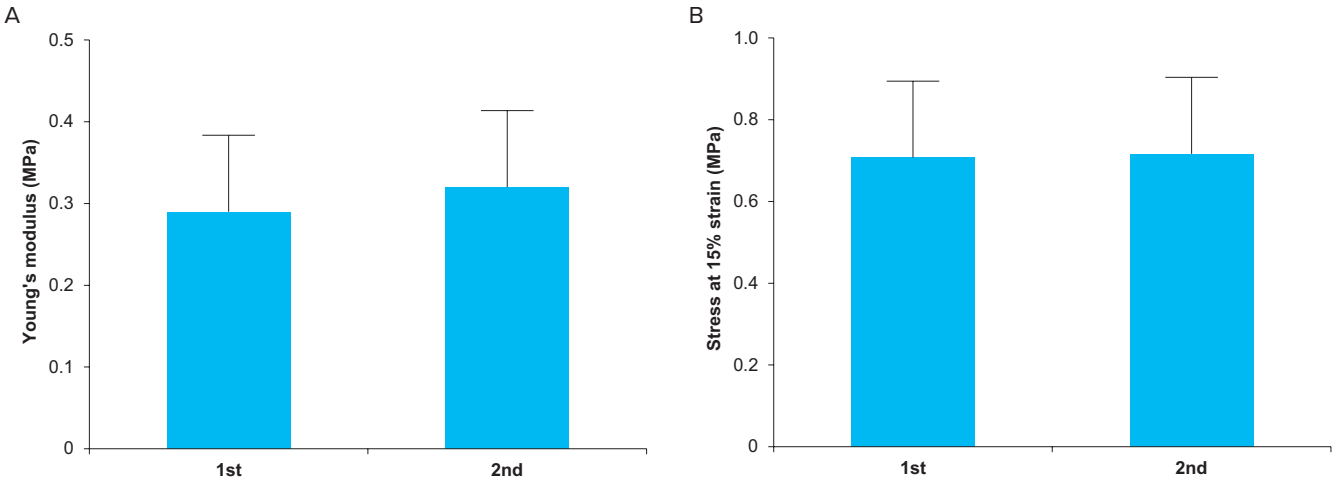

FIGURE 2. The results of two consecutive measurements of the 9-specimen set for the YM (**A**) and for strength at 15% strain (**B**)

much stronger mechanically than the superficialis tendon that was used in our study.

CONCLUSION

The porcine SDFT can be conveniently evaluated mechanically in the BioTester® 5000 (CellScale) in the uniaxial mode using an adequate clamping system. The measuring technique is stable and leads to reproducible results.

ACKNOWLEDGEMENT

The authors acknowledge the ongoing technical assistance of V. Păcurar and Dr. B. Cordos.

CONFLICT OF INTEREST

The authors declare no potential conflicts of interest or any financial interests that are relevant to the content, authorship, or publication of this article.

FUNDING

This research project is part of the doctoral activity of one of the authors (A.F.), which is supported by funds from the Clinical Emergency County Hospital Bistriţa. The project did not receive any other specific grant from funding agencies in the public or commercial sector.

DATA AVAILABILITY STATEMENT

The authors confirm that all relevant data are included in the published paper. Additional information can be provided upon reasonable request.

REFERENCES

1. Jung H-J, Fisher MB, Woo SLY. Role of biomechanics in the understanding of normal, injured, and healing ligaments and tendons. *Sports Med Arthrosc Rehabil Ther Technol*. 2009;1:9.
2. Buschmann J, Meier Bürgisser G. Biomechanics of Tendons and Ligaments. Tissue Reconstruction and Regeneration. Duxford, UK: Woodhead Publishing/Elsevier Ltd; 2017, p. 31–61.
3. Wertheim MG. Mémoire sur l'élasticité et la cohésion des principaux tissus du corps human. *Ann Chim Phys [Ser 3]*. 1847;21:385–414.
4. Cronkite AE. The tensile strength of human tendons. *Anat Rec*. 1936;64:173–186.
5. Walker Jr LB, Harris EH, Benedict JV. Stress-strain relationship in human cadaveric plantaris tendon: a preliminary study. *Med Electron Biol Eng*. 1964;2:31–38.
6. Benedict JV, Walker LB, Harris EH. Stress-strain characteristics and tensile strength of unembalmed human tendon. *J Biomech*. 1968;1:53–63.
7. LaBan MM. Collagen tissue: implications of its response to stress in vitro. *Arch Phys Med Rehabil*. 1962;43:461–466.
8. Burgio V, Civera M, Rodriguez Reinoso M, et al. Mechanical properties of animal tendons: a review and comparative study for the identification of the most suitable human tendon surrogates. *Processes*. 2022;10:485.
9. Domnick C, Wieskötter B, Raschke MJ, et al. Evaluation of biomechanical properties: are porcine flexor tendons and bovine extensor tendons eligible surrogates for human tendons in in vitro studies? *Arch Orthop Trauma Surg*. 2016;136:1465–1471.
10. Funchal LFZ, Astur DC, Pizzolatti ALA, Grimaldi AP, Jimenez AE, Moré ADO, de Mello Roesler CD, Cohen M. Tendon grafts with preserved muscle demonstrate similar biomechanical properties to tendon grafts stripped of muscular attachments: a biomechanical evaluation in a porcine model. *J Exp Orthop*. 2021;8:57.

sciendo

Journal of Interdisciplinary Medicine

ORIGINAL RESEARCH

DENTAL TECHNOLOGY // BIOCHEMISTRY // PHARMACY

Denture Base Polymer Biodegradation: In Vitro Study

Alessandra-Aniela Cerghedi[1], Zita Fazakas[2], Melinda Székely[1], Carmen Biriș[1], Cristina Molnar-Varlam[1]

[1] Faculty of Dental Medicine, "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania
[2] Faculty of Pharmacy, "George Emil Palade" University of Medicine, Pharmacy, Science and Technology, Târgu Mureș, Romania

CORRESPONDENCE

Alessandra-Aniela Cerghedi
Str. Gheorghe Marinescu nr. 38
540139 Târgu Mureș, Romania
Tel: +40 265 215 551
Email: cerghedianiela@yahoo.com

ARTICLE HISTORY

Received: May 3, 2023
Accepted: May 27, 2023

Zita Fazakas • Str. Gheorghe Marinescu nr. 38, 540139 Târgu Mureș, Romania. Tel: +40 265 215 551, Email: fazzita@yahoo.com

Melinda Székely • Str. Gheorghe Marinescu nr. 38, 540139 Târgu Mureș, Romania. Tel: +40 265 215 551, Email: mszekely_2000@yahoo.com

Carmen Biriș • Str. Gheorghe Marinescu nr. 38, 540139 Târgu Mureș, Romania. Tel: +40 265 215 551, Email: biriscarmen74@yahoo.com

Cristina Molnar-Varlam • Str. Gheorghe Marinescu nr. 38, 540139 Târgu Mureș, Romania. Tel: +40 265 215 551, Email: molnar.stanca@gmail.com

ABSTRACT

Introduction: Acrylic resins are the most frequently used materials for the bases of prostheses, and are also used in the re-optimization and the repair of prostheses. The **aim** of our study was to investigate whether direct contact with the resin causes decomposition of methyl methacrylate and formation of ketone bodies in the oral cavity. **Material and methods:** The in vitro study included 12 samples of autopolymerized polymethyl methacrylate and 8 samples of thermopolymerized polymethyl methacrylate. Some of the resin samples were intentionally prepared incorrectly, with modified powder to liquid ratios or thermal regime, to be able to compare them with samples prepared according to the manufacturer's instructions. Some of the samples were immersed in gastric juice or alcohol for 48 h at 37° C in a thermostatic bath to simulate the environment and temperature of the oral cavity, while others were kept at room temperature. The Legal reaction was used to identify the presence of ketone bodies in the solutions. **Results:** The samples that were prepared incorrectly and were kept at 37° C were the most affected. The presence of ketone bodies was demonstrated by the formation of a precipitate on the bottom of the test tube and the color change of the solution. Samples that were kept at room temperature were less affected, both from a spectrophotometric and biochemical point of view. **Conclusion:** The quality of polymethyl methacrylate can be improved by respecting the manufacturer's instructions and work protocols, and by avoiding substances that are considered aggressive, such as gastric juice, alcohol, and local factors in the oral cavity.

Keywords: bases of prostheses, unstable compounds, thermostatic bath, Legal reaction

INTRODUCTION

Polymethyl methacrylate (PMM) is a synthetic polymer derived from methyl methacrylate, a monomer that can be obtained through several methods, most frequently from acetone cyanohydrin, produced by the precipitation of acetone and hydrogen cyanide. These dental materials are produced through a free radical polymerization reaction activated by chemicals, heat, or light. N,N-dimethyl-p-toluidine is a chemical activator used for autopolymerizing materials. In the case of heat-polymerizing materials, heat can be provided by a hot water bath or microwave energy, whereas light-polymerizing materials use visible light as an activator. Acrylic-based resins are used in various applications in dental prac-

tice, such as denture bases, orthodontic appliances, and temporary crowns.[1]

Denture liners improve the fit of denture bases, increasing the retention and stability of removable prostheses. There are different types of denture liners, including hard reline resins and soft lining materials. Soft lining materials can be divided into two categories.[2–5] The first includes materials in which the liquid is composed of a monomer, such as methyl methacrylate, ethyl methacrylate, or butyl methacrylate, as well as plasticizers, such as phthalates, citrates, or sebacates, which increase the flexibility, transparency, durability, and longevity of the material. The second group consists of tissue conditioners, in which the liquid also contains a plasticizer and a mixture of ethyl alcohol.[6,7]

One of the most important issues related to the clinical use of acrylic resins is biodegradation, defined as a change in their chemical, physical, and mechanical properties due to local factors in the oral cavity.[8,9] The oral cavity is a complex environment in which dental materials are exposed to both endogenous and exogenous substances. The complex interactions that take place in this environment result in a general phenomenon of biodegradation that affects the materials present in the oral cavity, including PMM. These changes permanently affect the properties of the material and may endanger its functionality.[10] The aim of our study was to investigate the biodegradation of resins in the presence of aggressive substances.

MATERIAL AND METHODS

The study was divided into two parts: 1) the production of PMM samples; 2) the chemical analysis of the samples made.

For the first part of the study, we needed autopolymerized PMM (PMMA) (Figure 1) and thermopolymerized PMM (PMMT) (Figure 2), as well as a conformer in the form of a cone trunk, plaster, packing table, press, and water bath. The study group included 20 acrylic resin samples divided into two subgroups: PMMA (n = 12) and PMMT (n = 8). Four PMMA and four PMMT samples were prepared using a modified powder to liquid ratio and thermal regime to compare them with those that were prepared according to the manufacturer's instructions.

FIGURE 1. Preparation of PMM trunk cones

FIGURE 2. The immersion of samples in alcohol and gastric juice

FIGURE 3. The Legal reaction and the presence of ketone bodies

FIGURE 4. The formation of the precipitate at the bottom of the test tube

Considering the wide use of PMM in dentistry, in the second part of the study we analyzed whether different compounds are released from the resin or it retains its chemical integrity after being immersed in different substances considered to be aggressive. We chose an endogenous substance, gastric juice, which has a pH between 1 and 2.5, bitter taste, and no or slightly opalescent color, and an exogenous substance, ethyl alcohol, with a pH of around 4, no color and pungent smell. The gastric juice used in the study had a pH of 1.54, and the alcohol had a pH of 4.07. For both PMMA and PMMT, half of the samples were kept at room temperature (24 °C) for 48h, and the other half were placed 48h in a thermostatic bath o simulate the temperature in the oral cavity (37 °C).

We also performed several biochemical determinations. To identify the presence of ketone bodies, we used the Legal reaction, adding 2 drops of 10% sodium hydroxide, 3 drops of 10% sodium nitroprusside solution, and 1 mL of glacial acetic acid to the solutions in which the samples were immersed (Figures 3 and 4). The precipitate at the bottom of the test tubes and the color change that occurred after adding the reagents demonstrated the presence of ketone bodies in the solution, the intensity of the color being directly proportional to the amount of ketone bodies. Each solution containing the samples underwent a spectrophotometric analysis at a wavelength of 300 nm to measure the amount of ketone bodies (Figure 5).

FIGURE 5. Spectrophotometric analysis

RESULTS

The results show that both the PMMA and the PMMT samples kept at 24 °C had insignificant, almost non-existent changes. The samples that were incubated at 37 °C have changed both in terms of pH and spectrophotometrically, demonstrating that with the increase of temperature, decomposition of PMM and formation of ketone bodies takes place. At the same time, the micro-environment of the oral cavity had a demonstrable effect on the samples. Significant changes occurred in samples that were not prepared according to protocol, were immersed in gastric juice or alcohol, and were kept at 37 °C. We found statistically significant differences between the minimum pH values of PMMA samples prepared correctly (pH 2.34) and those prepared incorrectly (pH 4.35), when immersed in alcohol (Table 1). The differences in the pH values of PMMT samples prepared correctly and incorrectly were negligible both in gastric juice and alcohol, with an average difference of <0.5 (Tables 2, 3 and 4). Some of the samples

TABLE 1. Modifications observed in PMMA samples after immersion in alcohol and gastric juice

Medium	Temperature	Sample no.	Prepared according to the manufacturer's instructions	Mean pH	pH change	Ketone bodies
Alcohol (initial pH 4.07)	37 °C	3	Yes	2.346	↓	0.516 λ
		4	Yes	3.270	↓	0.514 λ
		6	No	3.700	↓	0.514 λ
	24 °C	3	Yes	3.583	↓	0.503 λ
		4	Yes	4.400	↑	0.507 λ
		6	No	4.356	↑	0.516 λ
Gastric juice (initial pH 1.57)	37 °C	1	Yes	3.976	↑	0.518 λ
		2	Yes	4.293	↑	0.523 λ
		5	No	4.186	↑	0.516 λ
	24 °C	1	Yes	3.630	↑	0.508 λ
		2	Yes	3.813	↑	0.515 λ
		5	No	3.846	↑	0.507 λ

TABLE 2. Modifications observed in PMMT samples after immersion in alcohol and gastric juice

Medium	Temperature	Sample no.	Prepared according to the manufacturer's instructions	Mean pH	pH change	Ketone bodies
Alcohol (initial pH 4.07)	37 °C	2	Yes	3.803	↓	0.476 λ
		4	No	4.513	↑	0.515 λ
	24 °C	2	Yes	3.573	↓	0.520 λ
		4	No	4.100	↑	0.503 λ
Gastric juice (initial pH 1.57)	37 °C	1	Yes	1.180	↓	0.525 λ
		3	No	1.273	↓	0.518 λ
	24 °C	1	Yes	1.290	↓	0.525 λ
		3	No	1.110	↓	0.505 λ

prepared incorrectly, with excess of monomer, could not analyzed due to technical issues (Table 1).

The spectrophotometric analysis revealed that the samples that contained the highest amount of ketone bodies were those that were intentionally prepared incorrectly, as well as those that were incubated in the thermostatic bath, demonstrating that PMM decomposition occurs with the increase of temperature and non-compliance with the manufacturer's instructions regarding the optimal ratio of ingredients. The maximum value obtained was 0.523 λ, and the minimum value was 0.476 λ. The amount of ketone bodies was very similar to pure acetone.

One-sample *t*-tests comparing mean pH values obtained after immersing the samples in gastric juice and alcohol yielded statistically significant differences ($p \leq 0.001$) for the majority of samples (Tables 3 and 4).

DISCUSSION

One of the most important consequences of biodegradation is the release of unbound monomers and additives that can be toxic to the oral cavity. In terms of material stability, biodegradation can cause significant changes in the physical and mechanical properties of a material, which can lead to catastrophic failure. Several studies have highlighted the release of compounds from different types of resins,[7] dental prostheses,[11–13] and repairing materials,[4] as well as orthodontic appliances, restorative materials, and tissue conditioners[14–17] with different chemical compositions. However, few clinical studies have focused on compound release from acrylic materials. A study evaluated the residual monomer concentration resulting from self-polymerizing resins. One of the consequences of the

TABLE 3. One-sample *t*-test comparing mean pH values of samples immersed in alcohol (pH 4.07)

Resin type	Temperature	Sample no.	pH (mean ± SD)	Difference between initial pH and pH at 48 h	p value
PMMA	37 °C	3	2.346 ± 0.484	−1.723	0.025
		4	3.270 ± 0.095	−0.800	0.005
		6	3.700 ± 0.088	−0.370	0.019
	24 °C	3	3.583 ± 0.344	−0.486	0.134*
		4	4.400 ± 0.125	0.330	0.045
		6	4.356 ± 0.032	0.286	0.004
PMMT	37 °C	2	3.803 ± 0.065	−0.266	0.019
		4	4.513 ± 0.049	0.443	0.004
	24 °C	2	3.573 ± 0.051	−0.496	0.004
		4	4.100 ± 0.017	0.030	0.095

TABLE 4. One-sample *t*-test comparing mean pH values of samples immersed in gastric juice (pH 1.57)

Resin type	Temperature	Sample no.	pH (mean ± SD)	Difference between initial pH and pH at 48 h	p value
PMMA	37 °C	1	3.976 ± 0.015	2.406	0.000
		2	4.293 ± 0.025	2.723	0.000
		5	4.186 ± 0.021	2.616	0.000
	24 °C	1	3.630 ± 0.17	2.243	0.001
		2	3.813 ± 0.049	2.276	0.001
		5	3.846 ± 0.037	−0.360	0.013
PMMT	37 °C	1	1.180 ± 0.000	−0.296	0.008
		3	1.273 ± 0.045	2.060	0.001
	24 °C	1	1.290 ± 0.072	−0.453	0.001
		3	1.110 ± 0.000	−0.460	0.008

biodegradation of materials, highlighted by several studies, is the elution of unbound components such as methyl polymethacrylate, benzoyl peroxide,[18] and phthalate esters. According to studies, most of the residual monomer is released in the first 24 h after immersion, after which the release rate decreases.[17,19–21] Some of these studies reported changes in pH as a result of biodegradation. Low pH increases the concentration of monomers, which affects the properties of acrylic resins used for denture bases.[19–21]

Ketone bodies are an important sign of chemical degradation of the prosthesis. Acrylic resins become porous as a consequence of degradation, and the risk of fracturing due to porosity increases considerably.[22] With time, poor hygiene of the prosthesis will affect the stability of the color of acrylic resins, and due to microporosity, a permanent microbial infiltrate will appear, which will lead to stomatitis and inflammation accompanied by discomfort.[23]

The toxicity of methyl methacrylate can be attributed to free radicals released during the polymerization reaction, due to oxidative stress; moreover, residual methyl methacrylate can also exhibit toxicity.[18] In a study in which glutathione was used to analyze the effect of methyl methacrylate regarding the expression of oxidizing enzymes, cell cultures have highlighted the toxicity of residual methyl methacrylate from resin-based materials, causing genotoxicity and changes in cellular cytokine factor expression.[17]

CONCLUSION

To improve the chemical stability of resins, it is mandatory to follow the protocol provided by the manufacturer and to instruct the patient on how to maintain and properly use his dental prosthesis. The quality of PMMA and PMMT is conditioned by respecting the work protocol and by local factors related to the oral cavity, digestive factors (gastric juice), and food-related factors (alcohol).

CONFLICT OF INTEREST

Nothing to declare.

REFERENCES

1. Zafar MS. Prosthodontics Applications of Polymethyl Methacrylate (PMMA): An Update. *Polymers (Basel)*. 2020;12:2299.
2. Sadamori S, Siswomihardjo W, Kameda K, Saito A, Hamada T. Dimensional changes of relined denture bases with heat cured, microwave-activated, auto polymerizing, and visible light-cured resins. A laboratory study. *Aust Dent J*. 1995;40:322–326.
3. Azzarri MJ, Cortizoa MS, Alessandrini JL. Effect of the curing conditions on the properties of an acrylic denture base resin microwave-polymerized. *J Dent*. 2003;31:463–468.
4. León BLT, Del Bel Cury AA, Rodrigues Garcia RCM. Loss of residual monomer from resilient lining materials processed by different methods. *Rev Odonto Ciênc*. 2008;23:215–219.
5. Bayraktar G, Guvener B, Burial C, Uresin Y. Influence of polymerization method, curing process, and length of time of storage in water on the residual methyl methacrylate content in dental acrylic resins. *J Biomed Mater Res B: Appl Biomater*. 2006;76:340–355.
6. Urban VM, Machado AL, Vergani CE, et al. Effect of water-bath post-polymerization on the mechanical properties, degree of conversion, and leaching of residual compounds of hard chairside reline resins. *Dent Mater*. 2009;25:662–671.
7. Celebi N, Yuzugullu B, Canay S, Yucel U. Effect of polymerization methods on the residual monomer level of acrylic resin denture base polymers. *Polym Adv Technol*. 2008;19:201–206.
8. Braden M. Some aspects of the chemistry and physics of dental resins. *Adv Dent Res*. 1988;2:93–97.
9. Sawtell RM, Downes S, Patel MP, Clarke RL, Braden M. Heterocyclic methacrylates for clinical applications-further studies of water sorption. *J Mater Sci Mater Med*. 1997;8:667–674.
10. Gonçalves TS, Menezes LM, Silva LEA. The residual monomer of auto-polymerized acrylic resin according to different manipulation and polishing methods. *Angle Orthod*. 2008;78:722–727.
11. Hassan M, Asghar M, Din SU, Zafar MS. Thermoset Polymethacrylate-Based Materials for Dental Applications. Amsterdam: Elsevier, 2019; p. 273–308.
12. Nejatian T, Pezeshki S, Yaqin Syed AU. Acrylic denture base materials. In: Khurshid Z, Najeeb S, Zafar MS, Sefat F, eds. Advanced Dental Biomaterials. Cambridge: Woodhead Publishing, 2019; p. 79–104.
13. Filho RR, Paula LV, Costa VC, Seraidarian PI. Avaliação de monômero residual em resinas acrílicas de uso ortodôntico e protético: análise por espectroscopia. *R Dental Press Orthodon Ortop Facial*. 2007;12:96–104.
14. Munksgaard EC. Plasticizers in denture soft-lining materials: leaching and biodegradation. *Eur J Oral Sci*. 2005;113:166–169.
15. Gutierrez-Villarreal MH, Rodríguez-Velazquez J. The effect of citrate esters as plasticizers on the thermal and mechanical properties of poly(methyl methacrylate). *J Appl Polym Sci*. 2007;105:2370–2375.
16. Bitencourt SB, Kanda RY, de Freitas Jorge C, et al. Long-term stainability of interim prosthetic materials in acidic/staining solutions. *J Esthet Restor Dent*. 2019;1–8.
17. Rokaya D, Srimaneepong V, Sapkota J, Qin J, Siraleartmukul K, Siriwongrungson V. Polymeric Materials and Films in Dentistry: An overview. *Journal of Advanced Research*. 2018;14:25–34.
18. Al-Thobity AM. The impact of polymerization technique and glass-fiber reinforcement on the flexural properties of denture base resin material. *Eur J Dent*. 2020;14:92–99.
19. Díez-Pascual AM. PMMA-Based Nanocomposites for Odontology Applications: A State-of-the-Art. *Int J Mol Sci*. 2022;23:10288.
20. Xu X, He L, Zhu B, Li J, Li J. Advances in polymeric materials for dental applications. *Polym Chem*. 2017;8:807–823.
21. Alqurashi H, Khurshid Z, Azeem Ul Yaqin S, et al. M. Polyetherketoneketone (PEKK): An emerging biomaterial for oral implants and dental prostheses. *J Adv Res*. 2020;28:87–95.
22. Mylonas P, Milward P, McAndrew R. Denture cleanliness and hygiene: an overview. *Br Dent J*. 2022;233:20–26.
23. Razia ZA, Faheema KD. Prevalence of Denture-Related Stomatitis in Edentulous Patients at a Tertiary Dental Teaching Hospital. *Front Oral Health*. 2021;2:772679.

JIM Journal of Interdisciplinary Medicine

About JIM

EDITORIAL PROCESS

All manuscripts submitted to the Journal of Interdisciplinary Medicine (JIM) will be first subject to a technical review, including quality check of all the files submitted, including tables, figures and references. Plagiarism check will be performed prior to referring the manuscript for review, in order to identify any possible fraud or scientific misconduct.

After technical review and anti-plagiarism assessment, the articles will be referred for review following a double-blinded review procedure. Reviewers can be suggested by the authors, however selection of the reviewers will be made by the editors, according to their expertise in the field of the article. The identity of the reviewers will not be disclosed to the authors, as well as the identity of the authors will not be disclosed to the reviewers.

The possible editorial decisions following the review procedure are: accepted, minor revisions required, major revisions required or rejected.

The editorial decision will be communicated to the authors as soon as the review process has been finalized. In case of revisions, the revised article will be sent to the reviewers, who will decide on a new recommendation for revision, acceptance or rejection. The estimated time from the submission to first decision is approximately 4 weeks, and from the final revision to acceptance approximately 2 weeks.

Prior to publication, all corresponding authors will receive a proof of their article in order to confirm the accuracy of the text or suggest modifications.

PUBLICATION ETHICS AND PUBLICATION MALPRACTICE STATEMENT

The Journal of Interdisciplinary Medicine adheres to the COPE principles of transparency and best practice in scholarly publishing. The Journal ensures an equal treatment for all articles by the Editor, Editorial team and journal reviewers, and has strict rules for confidentiality, disclosures, conflict of interest and authorship. At the same time, the Journal has strict regulations against publication fraud and plagiarism and well defined procedures to be taken if a publication fraud is suspected.

Conflict of interest

All participants in the peer-review and publication process — not only authors but also peer reviewers, editors, and editorial board members of journals — must consider their conflicts of interest when fulfilling their roles in the process of article review and publication and must disclose all relationships that could be viewed as potential conflicts of interest.

A conflict of interest exists when professional judgment concerning a primary interest (such as patients' welfare or the validity of research) may be influenced by a secondary interest (such as financial gain). Perceptions of conflict of interest are as important as actual conflicts of interest.

All manuscripts must acknowledge any possible conflict of interest related to the manuscript. If there is no conflict of interest in relation to the work performed or to the preparation of the manuscript, the authors should state that there are no conflict oif interest in relation to the manuscript. All the authors should also acknowledge any kind of material support, financial support or funding grants related to the work described in the manuscript.

Reviewers will be asked at the time they are asked to critique a manuscript if they have conflicts of interest that could complicate their review. Reviewers must disclose to editors any conflicts of interest that could bias their opinions of the manuscript, and should recuse themselves from reviewing specific manuscripts if the potential for bias exists. Reviewers must not use knowledge of the work they're reviewing before its publication to further their own interests.

Editors and Journal Staff Editors who make final decisions about manuscripts will recuse themselves from editorial decisions if they have conflicts of interest or relationships that pose potential conflicts related to articles under consideration. Editorial staff will not use information gained through working with manuscripts for private gain.

In cases where the Managing Editor has any conflict of interest in connection with a manuscript, the entire work related to the review process of that manuscript will be undertaken by the Editor-in-Chief. In cases where the

Editor-in-Chief has any conflict of interest in relation to a manuscript, the entire work related to the review process of that manuscript will be undertaken by the Managing Editor. In cases where both the Managing Editor and the Editor-in-Chief have any conflict of interest in relation to a manuscript, the entire work related to the review process of that manuscript will be undertaken by another member of the editorial board.

Submissions from members of the editorial board, editors and employees of the journal will be handled by the Editor-in-Chief, who will allocate the manuscripts for review to independent and blinded reviewers. Submissions from members of the owner institution will be assigned for review to members of the editorial board or external reviewers, taking into consideration the necessity to avoid any potential conflict of interest in the process of reviewer allocation.

Editorial manuscripts sent by members of the editorial board, following an invitation by the Editor-in-Chief, will undergo a review process in the editorial office.

Confidentiality

Editors of JIM will not share information regarding the manuscripts submitted to JIM to any other than the authors and the reviewers.At the time of reviewer allocation, reviewers will be instructed to keep the manuscripts and associated material strictly confidential. Reviewers should not publicly discuss author`s work and must not retain any manuscript for their personal use.

In case of manuscript rejection, the full content of the manuscript will be deleted from the editorial content of the Journal. In case of manuscript acceptance and publication, the Journal will keep copied of all the manuscript-related materials for at least three years.

The identity of the reviewers will not be revealed to authors, under no circumstances.

Human and animal rights

The authors should make sure that all the experiments on humans or animals are in accordance with the guiding principles described in the Declaration of Helsinki. Animal experiments should comply with the institutional and national guidelines or regulations for laboratory animals. Informed consent should be obtained from all the subjects participating in any experiment or clinical study and all the clinical studies should obtain the approval from the ethics committee of the institutions where the study is carried out, prior to initiation of experiments or studies.

When reporting research involving human data, authors should indicate whether the procedures followed have been assessed by the responsible review committee (institutional and national), or if no formal ethics committee is available, were in accordance with the Helsinki Declaration as revised in 2013 (www.wma.net/en/30publica tions/10policies/b3/index.html). If doubt exists whether the research was conducted in accordance with the Helsinki Declaration, the authors must explain the rationale for their approach and demonstrate that the institutional review body explicitly approved the doubtful aspects of the study.

When reporting experiments on animals, authors should indicate whether institutional and national standards for the care and use of laboratory animals were followed. Further guidance on animal research ethics is available from the International Association of Veterinary Editors' Consensus Author Guidelines on Animal Ethics and Welfare (http://veteditors.org/ethicsconsensusguidelines.html).

Protection of research participants

In order to respect the patient's right to privacy, no information related to patients' identification data, such as names, images or hospital identification codes should be included in the manuscript, unless there is a clear written approval obtained from the patient for this. This signed approval should be sent to the editorial office along with the manuscript.

Identifying information, including names, initials, or hospital numbers, should not be published in written descriptions, photographs, or pedigrees unless the information is essential for scientific purposes and the patient (or parent or guardian) gives written informed consent for publication. Informed consent for this purpose requires that an identifiable patient be shown the manuscript to be published. When informed consent has been obtained, it should be indicated in the published article.

Scientific misconduct

Scientific misconduct includes but is not necessarily limited to data fabrication; data falsification including deceptive manipulation of images; and plagiarism. All manuscript submitted to JIM will be first subject to a plagiarism check, that will be performed prior to referring the manuscript for review, in order to identity any possible fraud or scientific misconduct. The journal will use highly specialized anti-plagiarism softwares and if any suspicion of scientific misconduct is identified, the standard procedure recommended by COPE (Committee on Publication Ethics) will be followed.

Clinical trials

Authors of manuscripts related to clinical trials should register the clinical trial in the official clinical trial related public registries prior to submission to JIM, following the rules stated by the International Committee of Medical Journal Editors. Information related to registration of clinical trials can be found at ClinicalTrials.gov. In case of clinical trials, the trial registration number should be mentioned at the end of the abstract. Whenever a trial registration number is available, the authors should list this number the first time they use the trial acronym.

JIM Journal of Interdisciplinary Medicine

Instructions for authors

MANUSCRIPT SUBMISSION

All manuscripts should be submitted via email to **office@interdisciplinary.ro**.

The submission should include the following attachments:

1. Cover letter: all manuscripts submitted to JIM should be accompanied by a cover letter, signed by the corresponding author on behalf of all co-authors, stating that the reported study and manuscript are original and have not been published elsewhere, and the manuscript has not been submitted "in extenso" to any other journal. All disclosures relating to the preparation of the manuscript should be mentioned in the cover letter. The corresponding author should state clearly whether or not there are any conflicts of interest.

2. License to publish: The Journal of Interdisciplinary Medicine requires authors of original papers to assign copyright of their published contributions to the journal. A model of the License to Publish is available at **www.interdisciplinary.ro**.

Authorship is based on the following 4 criteria:

1. Substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work; AND
2. Drafting the work or revising it critically for important intellectual content; AND
3. Final approval of the version to be published; AND
4. Agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. In addition to being accountable for the parts of the work he or she has done, an author should be able to identify which co-authors are responsible for specific other parts of the work. In addition, authors should have confidence in the integrity of the contributions of their co-authors. All those designated as authors should meet all four criteria for authorship, and all who meet the four criteria should be identified as authors. Those who do not meet all four criteria should be acknowledged.

If authors request removal or addition of an author after manuscript submission or publication, journal editors should seek an explanation and signed statement of agreement for the requested change from all listed authors and from the author to be removed or added.

The corresponding author is the one individual who takes primary responsibility for communication with the journal during the manuscript submission, peer review, and publication process, and typically ensures that all the journal's administrative requirements, such as providing details of authorship, ethics committee approval, clinical trial registration documentation, and gathering conflict of interest forms and statements, are properly completed, although these duties may be delegated to one or more coauthors.

Authors should not submit the same manuscript simultaneously to more than one journal, in the same or different language.

MANUSCRIPT TYPES

The Journal of Interdisciplinary Medicine accepts the following categories of articles:

Original research

Manuscripts should be word processed. The manuscript must contain the title of the article, the authors' names, qualifications and address/es.

Peer Review: all articles undergo initial screening for suitability for the Journal of Interdisciplinary Medicine.

The length of contributions: ideally contributions should be no more than 4,000 words, including tables and figures. Suitable papers are then peer reviewed by two or more referees. Additional specialist advice may be sought if necessary, for example, from a statistician, before a final decision is made by the Editor-in-Chief.

An original research article should include a short **Abstract** of no more than 300 words, using the follow-

ing headings: Background, Aim of the study, Material and Methods, Results and Conclusions.

The manuscript should be structured as follows:

1. Introduction/Background: This introduces the aim of the study and the corresponding research hypothesis/es.

2. Material and methods: This section should describe all experimental details, research methodology, and study groups. The methodology should be detailed enough to allow reproducibility of the experiments. Give full descriptions of all equipment used (type, manufacturer, town, country). Details of statistical analysis should be reported here together with a level of significance [α value]. Authors should provide details of the statistical software package used (name, version, producer, town, country). Abbreviations of standard SI units of measurement should be employed. **Declaration of Helsinki:** The authors should state that their study complied with the Declaration of Helsinki, that the locally appointed ethics committee approved the research protocol and that written informed consent was obtained from the subjects (or their guardians) before the commencement of the study. Where animals are involved, the authors should state that their study complies with their institutional guidelines for the care and use of laboratory animals.

3. Results: This section should present the data arising from the experiments and their statistical significance. Do not discuss these findings in the Result Section.

4. Discussions: This section should contain a detailed analysis and interpretation of the results. Results should not be repeated in the Discussion section.

5. Conclusions: This presents the conclusions deriving from the outcome of the study and their clinical significance if appropriate.

Case reports

Case reports are intended for the presentation of interesting cases of interdisciplinary medicine encountered in clinical practice and should refer to actual and uncommon cases.

The report should have an abstract limited to 200 words, structured in the following manner: Introduction, Case presentation, and Conclusions.

The manuscript should be no more than a maximum of 2000 words, excluding references, figures, and figure legends. It should be structured as Introduction, Case presentation, Discussions, and Conclusions.

A case presentation should have a maximum of four authors, twenty references, and five figures.

Case series

Case series should include an abstract limited to 200 words, structured into Introduction, Case series presentation, and Conclusions.

The manuscript should be no more than 2000 words excluding references, tables, figures and figure legends. Case series should have a maximum of four authors, twenty references, and five figures.

Case report / Image focus

This category is intended to facilitate the publishing of representative images related to any clinical pathology. Accepted images may be published on the cover of the Journal. Images should be submitted as a figure accompanied by a clinical message that contains a description of the case and a detailed explanation of the figure, using a maximum of 300 words. For Case report / Image focus, the number of authors should be limited to four and the number of references to 10.

Reviews

The Journal of Interdisciplinary Medicine publishes review papers in any medical field of interest at an international level. Review articles should include a non-structured abstract of no more than 200 words with a maximum of 6000 words excluding references, tables, and figures.

Clinical update

The Journal of Interdisciplinary Medicine publishes update articles that describe current advances in any clinical field related to interdisciplinary medicine. Articles should include a non-structured abstract of no more than 200 words with a maximum of 4500 words excluding references, tables, and figures.

Letter to the editor

Letters to the editor should address either a recently published article in the Journal of Interdisciplinary Medicine, or a new topic in the field of cardiovascular emergencies.

Concerning a letter, discussing a recently published article, the comments contained in the letter will be forwarded to the authors of the original paper who will be invited to respond. Any response will be published in the same journal issue as the letter to the editor. A letter to the

editor should be no longer than 500 words, 5 references, and three authors. No abstract is required.

Editorial

Editorials should address either a particular topic that is currently of interest in the field of interdisciplinary medicine or to an article which is published in the same issue of the journal. The number of references should not exceed twenty-five in total.

MANUSCRIPT CONTENT

Style and spelling: Authors, whose first language is not English, are requested to have their manuscripts checked carefully, preferably by an English native-speaker, before submission, to expedite the review process.

Manuscript format: The manuscript must be submitted as a Word document and should be presented in the following order:

- Title page.
- Abstract, or a summary of case reports (references should not be included in abstracts or summaries).
- Main text separated under appropriate headings and subheadings using the following hierarchy: BOLD CAPS, bold lower case, Plain text, italics.
- Tables should be in Word format and placed in the main text where the table is first cited. Tables must be cited in the main text in numerical order.
- Acknowledgements, Competing Interests, Funding, and all other required statements.
- Reference list.
- Images must be uploaded as separate files (view further details under the Figures/illustrations section). All images must be cited within the main text in numerical order, and legends should be provided at the end of the manuscript. Appendices should be uploaded using the File Designation "Supplementary File" and cited in the main text.

The contents of your manuscript should be arranged in the following order:

1. **Title page** – should include: (1) the title of the article; (2) the name(s) of authors; (3) the institutional affiliations of the authors; (4) the position, institution, and location of all authors; (5) the telephone number, fax number and e-mail address of the corresponding author; (6) disclosure of grants, contracts and any other form of financial support received for the study.
2. **Abstract** – an abstract prepared in accordance to the type of the manuscript.
3. **Keywords** – between 3 and 6 keywords.
4. **Full text** – All manuscripts should be typed double-spaced, in Times New Roman 12 fonts, using Word format. References, tables and figures should be cited in numerical order, as they appear in the text. The abbreviations should be explained the first time they appear in the text, followed by the abbreviation in brackets.
5. **Acknowledgements** – should indicate clearly any source of funding received for the study, including grants, research contracts or any form of financial support.
6. **References.** References should be cited in numerical order, as they appear in the text, and should be indicated in superscript following the end of the sentence or the end of the part of the phrase they refer to.
7. **Tables** should be typed on separate pages at the end of the manuscript and should be numbered in Arabic numerals in the order of mention in the text. The abbreviations used in the table should be explained in a footnote below the table. Tables should not repeat the text and should be clear enough to be self-explanatory.
8. **Figures** should be prepared in TIF or JPG format, at a resolution of minimum 300 dpi. For figures reproduced or adapted from another source, this should be labeled as "Reproduced with permission from..." or "Adapted with permission from..." and should be accompanied by written permission from both the author and publisher of the original material. Figures should be combined with a legend which clearly describes the illustration.

REFERENCE STYLE

The journal will publish the reference list according to the style of Index Medicus (or spelled out if not listed in Index Medicus). List all the authors in each reference following the format and punctuation indicated below as examples:

Reference to an article

1. Benedek I, Gyongyosi M, Benedek T. A prospective regional registry of ST-elevation myocardial infarction in Central Romania: impact of the Stent for Life Initiative recommendations on patient outcomes. *Am Heart J*. 2013;166:457-465.

Reference to a book

2. Nichols WW, Rourke MF. Aging, High Blood Pressure and Disease in Human. 3rd ed. London/Melbourne: Lea and Febiger; 1990.

Reference to a chapter in a book

3. Nichols WW, O'Rourke MF. Aging, high blood pressure and disease in humans. In: Arnold E, ed. McDonald's Blood Flow in Arteries: Theoretical, Experimental and Clinical Principles. 3rd ed. London/Melbourne/Auckland: Lea and Febiger, 1990; p. 398-420.

Reference to a webpage

4. Panteghini M. Recommendations on use of biochemical markers in acute coronary syndrome: IFCC proposals. eJIFCC 14. http://www.ifcc.org/ejifcc/vol14no2/1402062003014n.htm (28 May 2004)

PUBLICATION FEE

To publish in the Journal of Interdisciplinary Medicine, authors are required to pay a publication fee for an accepted article. The fee is 50 EUR or 250 RON, for all types of articles.

The invoice will be sent to the corresponding author after the decision of acceptance is released, and the manuscript will be sent to the publication system only after the corresponding author pays the publication fee.

There is no fee for manuscript submission or revision.

Waivers for publication fee may be issued by request, following the request send by the corresponding author to office@interdisciplinary.ro.

COMPLAINTS

In cases where the authors wish to file a complaint, please contact the editorial office:

Journal of Interdisciplinary Medicine
Str. 22 Decembrie 1989 nr. 76–78
540124 Târgu Mureș, Romania
E-mail: office@interdisciplinary.ro

Please describe the reason for complaining and specify the address for correspondence. Where the complaint is related to the editorial process, related to a manuscript, please include the name of the manuscript and the date the manuscript was submitted. The Editor-in-Chief, together with the editorial office will analyze the complaint and will answer in maximum three working days.