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# AN RE/RM APPROACH TO THE DESIGN AND MANUFACTURE OF REMOVABLE PARTIAL DENTURES WITH A BIOCOMPATIBILITY ANALYSIS OF THE F75 Co-Cr SLM ALLOY

## RE/RM-PRIBLIŽEK, NAČRTOVANJE IN IZDELAVA SNEMLJIVIH DELOV ZOBOVJA Z ANALIZO BIOKOMPATIBILNOSTI ZLITINE F75 Co-Cr SLM

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The implementation of computer-aided technologies and systems has paved the way towards a significant advancement of the conventional modelling and manufacture of dental replacements. In this research the focus is on approach that combines reverse engineering as a modelling technique and rapid manufacture, i.e., selective laser melting, as the manufacturing technology, with special emphases on material selection in the fabrication of removable partial dentures. The paper presents the results of a biocompatibility analysis of the F75 Co-Cr dental alloy using the MTT eluate test.

Keywords: reverse engineering, rapid manufacturing, selective laser melting, removable partial dentures, biocompatibility, Co-Cr alloys, MTT eluate test

Uporaba računalniških tehnologij in sistemov je odprla pot do pomembnega napredka konvencionalnega modeliranja in izdelave snemljivih zobnih nadomestkov. Težišče te raziskave je približek, ki kombinira obratno inženirstvo kot tehniko modeliranja in hitre izdelave z laserskim taljenjem kot izdelavno tehnologijo s posebnim poudarkom na izbiri materiala za snemljive dele zobovja. V članku so predstavljeni rezultati analize biokompatibilnosti zobne zlitine F75 Co-Cr z MTT-preizkusom eluiranja.

Ključne besede: obratno inženirstvo, hitra izdelava, selektivno lasersko taljenje, snemljivi deli zobovja, biokompatibilnost, Co-Cr zlitina, MTT-preizkus eluiranja

## 1 INTRODUCTION

Dental prosthetics (also known as prosthodontics) has always maintained close relationships with engineering disciplines, relying mostly on production engineering. The rapid development of computer-aided (CA) technologies, which completely transformed production engineering, also left an indelible mark on dental prosthetics. Striving towards its primary goal – *primum non nocere* (in English, 'Above all, do not harm!'), the area of dental prosthetics has introduced numerous novel technologies and methods that allow the manufacture of precision, custom-made, optimal dental replacements. During the past decade, efforts have been concentrated towards an advancement of the modelling and manufacture of dental replacements by introducing modern CA systems, state-of-the-art materials and machining technologies, as opposed to the traditional way of manual manufacture, which is prone to numerous subjective errors.[1] Amongst modern CA systems that have found broad application in this area, the most widely used are 3D-digitization systems, CAD and reverse engineering (RE), CAE, CAM, rapid manufacture (RM) (or additive manufacture that become the adopted term in the sector) and rapid prototyping (RP). The development and implementation of such technologies and systems have paved the way towards a significant advancement in conventional modelling, manufacture and the inspection of dental replacements.[1–6]

Different dental substrates may have special requirements related to their modelling and manufacture. Removable partial dentures (RPDs) represent a special type of denture, designed for partially edentulous patients who cannot have a fixed partial denture, i.e., a bridge. This type of prosthesis is referred to as removable, as patients can remove and reinsert it when required without professional help. Traditionally, RPD frameworks are manufactured through the so-called lost-wax technique, where a wax pattern burns out in a preheating unit followed by an immediate casting of the melted alloy. Though in use for decades, this technique is sensitive and prone to human-induced errors.[1,6]

In this research the focus is on an approach that combines RE as a modelling technique, and RM i.e., selective laser melting (SLM), as the manufacturing technology, with a special emphasis on material selection in the fabrication of RPD frameworks.

The virtual design of dental restorations today almost always requires the application of RE modelling. RE, a modelling technique widely used in different engineering fields, has been increasingly applied in the field of prosthodontics during the past several years, mainly because of the rapid development of dental 3D digitization systems and the corresponding modelling software[2,3]. RM is no exception and its results in the field of prosthodontics significantly depend on RE modelling. Realizing the benefits of RE and RM, recently there have been several research works related to the possible use of these technologies in the design and manufacturing of RPD frameworks.[1,3,7–9]

SLM, an RM technique, is based on a layer-wise material addition that allows the generation of complex 3D parts by selectively melting successive layers of metal alloy powder on top of each other. As presented by Eyers and Dotchev in[5] and Vandenbroucke and Kruth in,[10] SLM is very suitable for dental and medical applications, due to the complex geometry of the produced parts. A pilot study presented by Williams et al.[9] showed that an RPD framework produced by SLM techniques was comparable to conventional frameworks in terms of accuracy, quality of fit and function. However, this conclusion is based on a single study and much work needs to be completed before a final conclusion can be reached.

A very important issue in prosthodontics is the material used, i.e., the alloy.[10–13] This issue is even more important in RPD framework manufacturing as the application is limited to cobalt-chromium (Co-Cr) alloys due to the low rigidity of titanium (Ti) alloys, as described by Aridome et al. in[14], and the unfavourable characteristics of gold (Au) alloys, that are otherwise widely used in the fabrication of other dental restorations. The application of SLM in the manufacturing of RPD, brings some additional material requirements related to mechanical properties[10,14] and biocompatibility.[15,16] Although a dental prosthesis fabricated by SLM showed the potential of SLM as manufacturing technique, there are still very few reports of SLM application in the manufacture of RPDs from Co-Cr alloys. Though Co-Cr alloys have been used in dentistry for years, little is known about the influence of the SLM process on the alloys' biocompatibility and mechanical behaviour.

This paper, with regards to the above discussion, focuses on the applicability and possible benefits of the application of RE and RM techniques in the design and manufacture of RPD frameworks. Moreover, special attention in this research has focused on the material properties related to SLM application in the manufacture of RPD frameworks. The paper also presents the results of an analysis of biocompatibility conducted with an MTT eluate test of the F75 Co-Cr dental alloy.

## 2 RE/RM IN DESIGN AND MANUFACTURING OF RPD FRAMEWORKS

The RE and RM combination has been recognized as fully compatible and very effective. Potential advantages include: a decrease of manufacturing time, an inherent repeatability, and the achievement of high quality through eliminating operator variations that are usually connected with the conventional (manual) design and manufacture of an RPD (**Figure 1**).

The application of RM, i.e., SLM in an RPD framework fabrication implicates the workflow presented in **Figure 2**. This workflow clearly presents three main phases:

1. RE modelling (of the patient's cast),
2. virtual design of the RPD framework,
3. RM of RPD framework.

The first two phases are frequently unified and denominated in references as the CAD phase, while the third is often described as the CAM phase.[1,3,5,7–10]

The RE phase starts with the 3D digitization of the patient's cast. This usually includes acquiring a dental

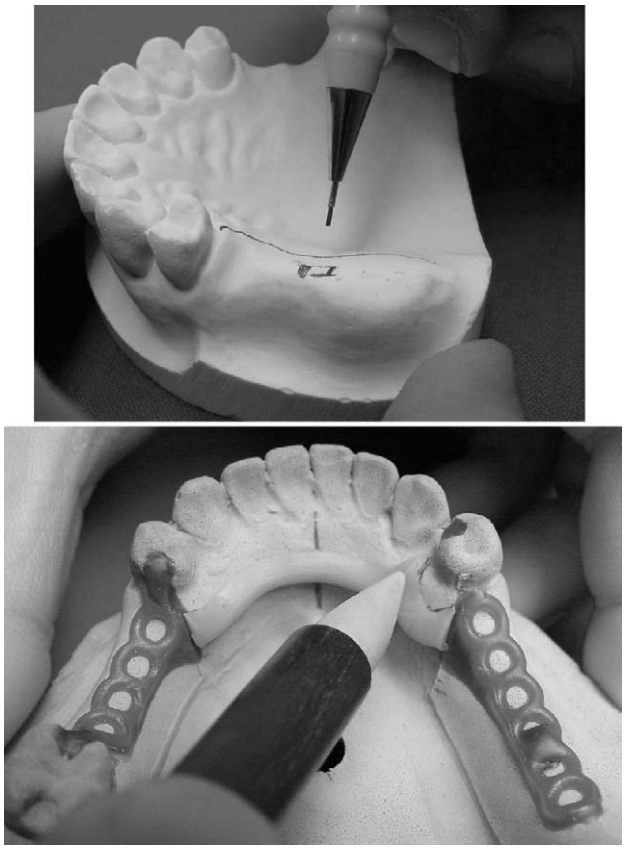

**Figure 1:** Conventional (manual) design of RPD
**Slika 1:** Konvencionalno (ročno) načrtovanje RPD

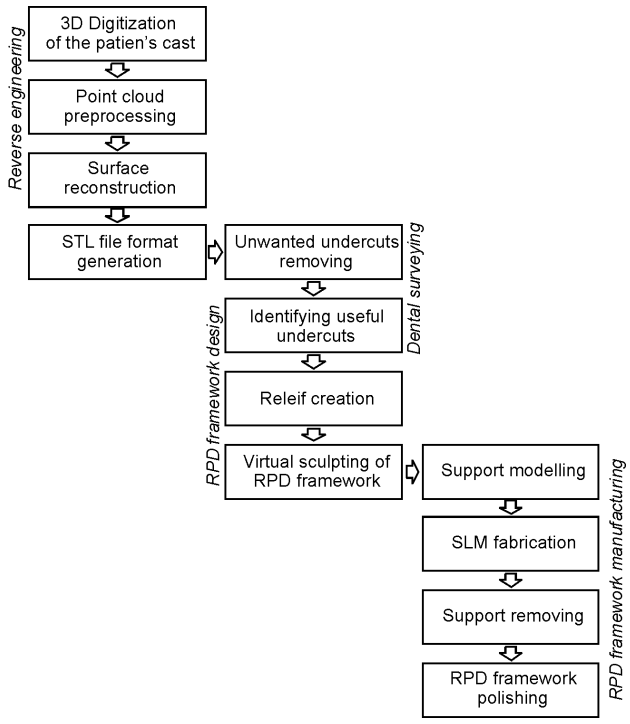


**Figure 2:** The typical workflow of an RE/RM approach in RPD framework fabrication
**Slika 2:** Značilen tok RE/RM približka v okviru RPD-izdelave

impression and extra oral scanning of a gypsum model produced from the impression (**Figure 1**).[1,6] However, the process could replace the need for an impression by the application of intra-oral scanning[2] or CT.[6,17,18] The point cloud obtained almost always needs to be pre-processed in order to insure a high-quality surface reconstruction, i.e., a credible CAD model. Regarding the applied 3D digitization technique/system, the pre-processing step can include different processes, such as noise filtering, data reduction, segmentation of the point cloud parts or assembling.[19] The obtained surface model (the reference model or the "buck") is usually exported to an STL file format based on triangular polygons, which is a suitable format for virtual dental surveying and virtual sculpting environments.[8]

The initial step of the RPD framework-design phase is the virtual dental surveying that is needed to identify areas of undercut present on the CAD model of the patient's teeth and soft tissues (**Figure 3a**). Unwanted undercuts have to be removed in order to ensure an unobstructed withdrawal of the RPD from the patient's orifice.[1,8] However, there are some useful undercuts that need to be retained and their identification and measurement are important as they serve as secure holders of flexible clasps that provide reliable retention.[1] The next step is the modelling of reliefs – the parts of a model that prevent the RPD framework from resting on the surfaces of soft tissues (**Figure 3b**).[1] After the reliefs have been added, virtual sculpting of the RPD framework elements (1-occlusal rest, 2-polymeric retention frame, 3-lingual bar, 4-acrylic line, 5-non-active clasp, 6-guide plate) may begin (**Figure 3c**).

The virtual sculpting stage is based on software tools enabling analogous work to that used in physical sculpting. This is enabled through a haptic interface that incorporates positioning in 3D space and allows rotation and translation in all axes, transferring hand movements into the virtual environment (**Figure 4**). Moreover, haptic systems enable the operator to feel contact with the object that is the subject of the modelling. Besides this usage of the haptic arm in a freehand manner, the process of virtual sculpting also allows the application of standard CAD parametric features based on sizes, shapes, relations and positions.[1,8]

Once the CAD model of the RPD framework is obtained, it can be passed to the SLM after its preparation in appropriate software. This preparation primarily involves the creation of an adequate support (**Figure 5**) that acts as a firm base for the RPD framework to be built onto and which also conducts heat away during the sintering processes. As the supports need to be removed after the solidification of the part, it is important to avoid placing them on the fitting surface of the RPD.[8]

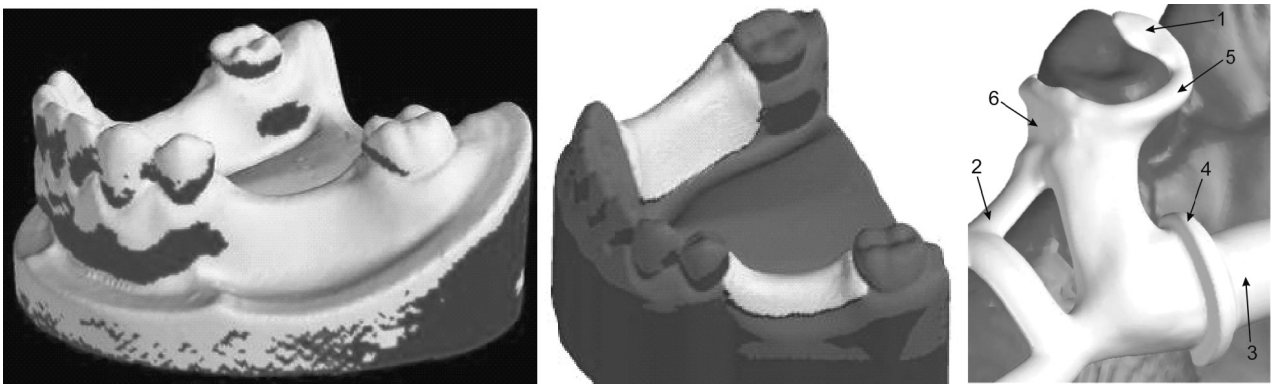


**Figure 3:** RPD framework design phase – the main steps: a) Identification of undercuts, b) Relief modelling, c) RPD framework elements
**Slika 3:** Okvir faze RPD-načrtovanja – glavni koraki: a) identifikacija spodnjih prerezov, b) modeliranje reliefa, c) elementi okvirja RPD

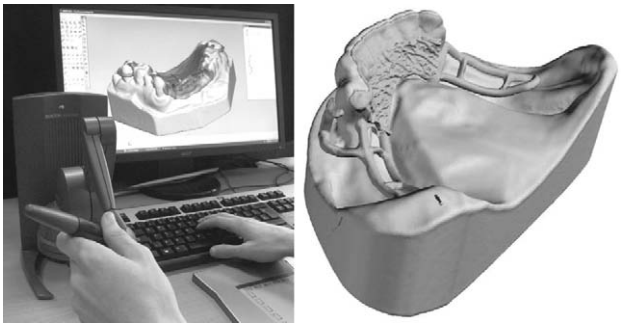

**Figure 4:** Virtual design of RPD frameworks
**Slika 4:** Virtualno načrtovanje podlag RPD

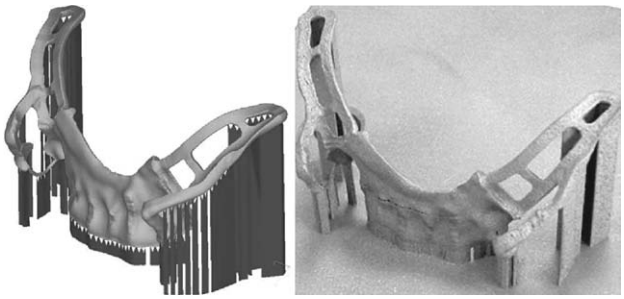

**Figure 5:** Support in SLM manufacturing of RPD frameworks
**Slika 5:** Podlaga pri SLM-izdelavi RPD-podlage

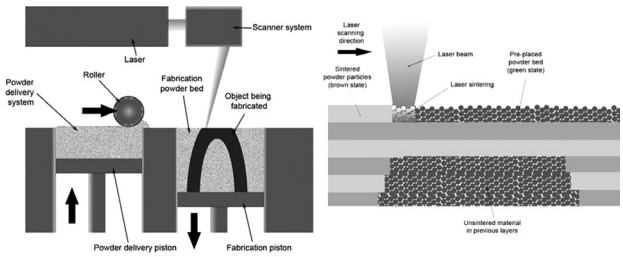


**Figure 6:** Principle of the SLM process
**Slika 6:** Princip SLM-procesa

During the SLM process, powdered material is spread by a hopper and wiper mechanism. To accommodate a new layer of the material, the build platform has to move down by one layer thickness. Subsequently, the powder is deposited incrementally on top of each solidified layer, and the process is repeated (**Figure 6**).

The manufactured RPD framework needs to be finished and polished and this is performed using traditional dental laboratory procedures.[8,9] Finally, the finished RPD framework has to be evaluated on the patient's cast. This is performed by a prosthodontic expert, who will assess the quality of fit according to recommended practice.[8]

## 3 BIOCOMPATIBILITY TESTING OF THE SLM Co-Cr ALLOY F75 (BY MTT ELUATE TEST)

One of very important issues that need to be investigated is the biocompatibility of the specific Co-Cr alloy used for SLM, since – to the best of authors' knowledge – there are no known published conclusions about biocompatibility. Though the basic chemical elements in alloys used for SLM (F75) and conventional investment casting, also known as the lost-wax technique (Remanium 380+) generally match, they differ by a small percentage due to the specific requirements needed by the process (**Table 1**). However, it has been shown that a modification in composition and pre-treatment can influence the cytotoxicity of an alloy on a large scale.[20,21]

The previous discussion motivated the authors to start research related to the biocompatibility testing of the specific Co-Cr alloy used for SLM.

In biocompatibility evaluations of alloys, cell culture studies are the usual starting point as they enable an investigation of the toxicity in a simplified system that minimizes the effect of confounding variables.[21] Thus, within this study a murine fibroblast cell line (L929) was used in accordance with the requirements of the ISO standard 7405 (ISO 2008).[15] The cytotoxicity determination of the Co–Cr alloy used for the fabrication of an SLM RPD framework was based on the MTT eluate test method.

### *3.1 Sample preparation*

The investigation included the fabrication of two groups of test samples – the first from the SLM and the second from conventional investment casting (**Figure 7**).

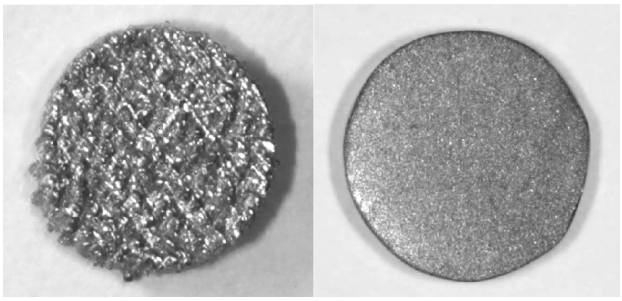

**Figure 7:** Test samples obtained by SLM (left) and by vacuum casting (right)
**Slika 7:** Preizkušanci SLM (levo) in vakuumskega taljenja (desno)

**Table 1:** Composition of the Remanium GM 380+ and Sandvik Osprey F75 alloys in mass fractions, *w*/%
**Tabela 1:** Sestave zlitin Remanium GM 380+ in Sandvik Osprey F75 v masnih deležih, *w*/%

| Ingredients, *w*/% | Co | Cr | Mo | Si | Mn | N | C | Fe | Ni |
|---|---|---|---|---|---|---|---|---|---|
| Remanium GM 380+ | 64.6 | 29 | 4.5 | <1 | <1 | <1 | <1 | / | / |
| Sandvik Osprey F75 | Balance | 27–30 | 5–7 | <1 | <1 | / | <0.35 | <0.75 | <0.5 |


**Figure 8:** SLM system – Realiser MTT-Group, UK
**Slika 8:** SLM-sistem Realiser MTT-grupe, VB

The first group of samples was manufactured by the SLM system *Realiser MTT-Group* (**Figure 8**) and the software was *Magics* 9.5, *Materialise NV.* The Co-Cr layer thickness was 0.075 mm, the laser's maximum scan speed was 300 mm/s, and the beam diameter was 0.150–0.200 mm. The F75 Co-Cr alloy (Sandvik Osprey Ltd., UK) used in this study is a spherical powder with a maximum particle size of 0.045 mm (particle size range 0.005–0.045, mean size approx. 0.030 mm). After the SLM process was completed and specimens' supports were removed, the discs were finished by polishing according to the usual dental laboratory procedure.

The second group of samples – four disc specimens with a radius of 5 mm and thickness of 1 mm – were fabricated from a non-precious Co-Cr alloy *Remanium GM380+* (Dentaurum, Ispringen, Germany) containing no Ni, Be or Fe, and widely used for RPD framework casting. The discs were obtained from wax patterns, invested in *Rema dynamic* (Dentaurum, Ispringen, Germany) investment, and vacuum casted in a *Nautilus CC* (Bego, Bremen, Germany) system (**Figure 9**). After casting, the discs were divested and blasted with 100-µm aluminium oxide particles, and then polished with silicon carbide papers in the sequence 320, 400, 600, 1200, 1500 and 2000. The final polishing was performed using oxide pastes.


**Figure 9:** Nautilus CC system for vacuum casting
**Slika 9:** Nautilusov sistem za vakuumsko taljenje

### *3.2 MTT eluate testing*

The test performed was the MTT (tetrazolium colorimetric assay) eluate test, widely used for the quantitative evaluation of cell proliferation and survival.[16,20] The assay depends on the cleavage of tetrazolium salt 3-[4,5–dimethylthiazol–2–yl]-2,5-diphenyltetrazoliumbromide (MTT) to purple formazan crystals by mitochondrial dehydrogenases in viable cells.[21] The assay detects living but not dead cells and the rate of MTT reduction to formazan products. It is also dependent on the degree of cell activation. Therefore, the assay is suitable for measuring cytotoxicity, proliferation and activation.[16]

Eluates of both the CM and SLM disc samples were prepared. The samples were extracted with 10 mL of Dulbecco's modified Eagle's medium (DMEM) without serum for 48 h. The extraction was performed in an atmosphere of 5 % $CO_2$ and 95 % air at 37 °C. All the extracts were filtered for sterilization and used as a culture medium for L-929 cells. MTT assays with L-929 cells treated with different eluates were completed after 48 h of incubation. The experiment was repeated twice (thus eight CM and eight SLM samples were tested in two independent experiments).

The cells (L929) were cultured in Petri dishes containing eluates of the CM or SLM alloy discs (**Figure 10**). They were incubated for (3, 5, 7 and 9) d at 37 °C in 95 % air and 5 % $CO_2$. The control samples contained a regular culture medium. After the incubation, the cells were detached using enzymatic digestion and counted in a counting chamber using trypan blue. Briefly, $5 \times 10^3$ cells were seeded to a 96-well plate and cultured for 48 h at 37 °C in 95 % air and 5 % $CO_2$.

After the incubation period, 20 µL of MTT solution were added to each well and incubated for another 3 h. The purple formazan product was dissolved in 100 µL of

**Figure 10:** MTT eluate testing
**Slika 10:** MTT preizkus eluiranja

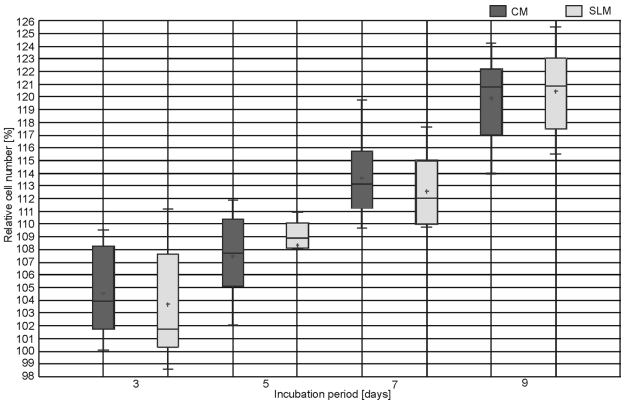


**Figure 11:** MTT test results for CM and SLM alloy after different incubation periods – graphical review of the confidence intervals
**Slika 11:** Rezultati MTT-preizkusov CM- in SLM-zlitine po različnih dobah inkubacije – grafična predstavitev intervalov zanesljivosti

0.04-M hydrochloric acid in isopropanol. The reduced MTT was then measured spectrophotometrically in a dual-beam, microtiter plate reader *Multiscan MCC/340* at 540 nm with a 690-nm reference. The optical density values of the experimental groups were divided by the control and expressed as a percentage of the control.

### *3.3 Statistical results of the MTT eluate testing*

A statistical analysis was carried out using the Statgraphics Centurion program. The data were evaluated statistically using the Student's t-test and a value of $p < 0.05$ was considered to be statistically significant. The results of the MTT eluate testing of the disc samples that were taken after an extraction period of (3, 5, 7 and 9) d are listed in **Table 2**, and graphically presented in **Figure 11**.

Of particular interest was the confidence interval evaluation for the difference between the means. Since all four intervals contain the value 0, there is no statistically significant difference between the means of the CM and SLM samples at the 95 % confidence level. Furthermore, a t-test was used for testing a specific hypothesis about the difference between the means of the populations from which the two samples come. In this case, the test was constructed to determine whether the difference between the two means equals 0.0 (null hypothesis: mean1 = mean2) versus the alternative hypothesis that the difference does not equal 0.0 (mean1 ≠ mean2). Since the computed P-values are not less than 0.05 in all four cases, the null hypothesis cannot be rejected.

## 4 DISCUSSION

Publications investigating the corrosion of dental alloys give information firstly about the release of potentially harmful ions from the dental device. Cell-culture tests give an insight into whether the released ions in a cell-culture medium imitating the oral environment could have a negative effect on the biological system. The results of the MTT eluate testing with the SLM samples do not show significant cellular damage potential. Statistical analyses carried out showed that the alloys did not release harmful material that could cause acute effects against L929 cells under the given experimental conditions. Furthermore, the MTT test showed no permanent damage to the cell function. The viability was much higher than 50 % after all the extraction periods for both the CM and SLM alloy. Replication during an extended contact period with

**Table 2:** Results of statistical analysis of MTT eluate testing of CM and SLM disc samples
**Tabela 2:** Rezultati statistične analize MTT-preizkusa eluiranja CM- in SLM-vzorcev

| | Period of cell incubation in d | *3* | | *5* | | *7* | | *9* | |
|---|---|---|---|---|---|---|---|---|---|
| Descriptive statistics | Technology | CM | SLM | CM | SLM | CM | SLM | CM | SLM |
| | Count | 8 | 8 | 8 | 8 | 8 | 8 | 8 | 8 |
| | Average % of relative cell No. | 104.655 | 103.639 | 107.604 | 108.364 | 113.73 | 112.694 | 119.778 | 120.525 |
| | Standard deviation | 3.62557 | 4.56325 | 3.43234 | 2.94701 | 3.34183 | 2.92113 | 3.45744 | 3.58149 |
| | Coeff. of variation,% | 3.4643 | 4.40303 | 3.18979 | 2.71955 | 2.93839 | 2.5921 | 2.88655 | 2.97158 |
| | Minimum | 100.05 | 98.64 | 102.04 | 101.54 | 109.78 | 109.78 | 114.01 | 115.61 |
| | Maximum | 109.45 | 111.23 | 111.97 | 111.02 | 119.74 | 117.64 | 124.31 | 125.64 |
| Comparison of means | 95 % CIM* | 104.655 +/– 3.03106 | 103.639 +/– 3.81498 | 107.604 +/– 2.86951 | 108.364 +/– 2.46377 | 113.73 +/– 2.79385 | 112.694 +/– 2.44213 | 119.778 +/– 2.8905 | 120.525 +/– 2.99421 |
| | 95 % CIDM** | 1.01625 +/– 4.41952 | | –0.76 +/– 3.43048 | | 1.03625 +/– 3.36576 | | –0.7475 +/– 3.77485 | |
| | t | 0.493186 | | –0.475165 | | 0.660339 | | –0.424715 | |
| | P-value | 0.629527 | | 0.641997 | | 0.519754 | | 0.6775 | |

* CIM – Confidence Interval for Means, ** CIDM – Confidence Interval for the difference between the Means assuming equal variances

potential toxic substances, however, showed good biocompatible properties of the chosen SLM alloy. Additionally, the negative effect decreased with time for both the examined substances. Therefore, both alloys can be rated as non-cytotoxic.

It has, however, to be noted that SLM, as a complex thermo-physical process, produces a variation in the final product depending on several factors, such as the material, laser, scan and parameters of the environment used.[10] Changeable variables include: laser power, layer thickness, scan speed and hatch spacing. With the current settings, as can be seen from the study, the final product complies with the required biocompatibility standards, showing no potentially harmful effect. However, those values can be adjusted accordingly, optimizing some aspects that can have a negative effect on the materials' properties, such as porosity. For example, for a low energy input, successive scan tracks may not be fully molten, leaving large pores along the scan lines, as seen in the mentioned study. If so, a combination other parameters might change the surface properties, which might also result in changes in the ion release and therefore require separate biocompatibility studies. This study has, however, showed that the initial screening gave positive results and the F75 SLM alloy can be subjected to further tests.

The study concerning the ion release from the cast and SLM samples, presented in,[10] revealed the more favourable behaviour of the SLM specimens. The main ion detected was cobalt, since the corrosion of the alloy is determined by the main component, and the passivating effect of chromium. The SLM test specimens showed lower emissions than the cast specimens, probably because the laser-melted material is more homogeneous, contains fewer pores and has a finer microstructure. This also highlights the importance of the finishing procedure, which still has to be conducted manually.

Another physical factor that might influence the biocompatibility 'in vivo' is the surface roughness[21]. The changes in this parameter can be explained by the so-called stair effect, inherent to the layer-wise production of SLM. In the oral environment, this might increase plaque retention, leading to the formation of acidic micro-fields that might change the metallic ion release unfavourably. This effect can be reduced by decreasing the layer thickness or by increasing the sloping angle[10].

## 5 CONCLUSION

This paper shows that a complete RE/RM procedure for RPD framework fabrication should bring significant advantages both to practitioners and patients. Special attention was focused on the biocompatibility analysis of the dental alloys used with the SLM. Moreover, the paper presents a biocompatibility evaluation of the F75 SLM dental alloy using the MTT eluate test. On the basis of the obtained results, within the limitations of the study, it can be concluded that the RE/RM procedure showed a promising potential in RPD framework fabrication, as well as that the F75 alloy used for SLM manufacturing showed positive initial results regarding its biocompatibility. However, further studies, including in vivo tests and tests of mechanical properties, have to be conducted before the final release of the alloy for mass production.

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