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A pilot study on the use of 3D printers in veterinary medicine

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Abstract

Additive manufacturing (AM) is the process of joining materials to create layer-by-layer three-dimensional objects using a 3D printer from a digital model. The great advantage of Additive Manufacturing is to allow a freer design than traditional processes. The development of additive manufacturing processes has permitted to optimize the production of the customized product through the modeling of the geometry and the knowledge of the morphometric parameters of the body structures. 3D printing has revolutionized the field of Regenerative Medicine because, starting from computerized tomography (CT) images and using traditional materials such as plastic and metals, it can provide customized prostheses for each patient, which adapt perfectly to the needs of the subject and act as structures support. 3D printing allows you to print three-dimensional porous scaffolds with a precise shape and chemical composition suitable for medical and veterinary use. Some of these scaffolds are biodegradable and appear to be ideal for bone tissue engineering. In fact, they are able to simulate extracellular matrix properties that allow mechanical support, favoring mechanical interactions and providing a model for cellular attachment and in vivo stimulation of bone tissue formation.

Key words: bone lesions, dog, prosthesis, regenerative medicine, 3D printing.

Introduction

Additive manufacturing is the process of joining materials to manufacture three-dimensional models (3D) in an additive layer by layer without the need for a mold (11).

Today, additive manufacturing is applied in many sectors, for example in the industries aerospace, electronics, automotive, medical and veterinary medicine (7,15). Recently, AM has become very important because it creates patient-specific biomedical devices in fields such as biotechnology and healthcare. New implants and prostheses that were previously impossible to produce with traditional methods are rapidly becoming available with the progress of AM (2).

In general, AM technology starts from the modeling of 3D objects by computer-aided design (CAD) to the fabrication of structures through a layer-by-layer 3D printing process. In the modeling phase, some software applications allow to produce, modify and optimize the structures designed to improve the final products (20).

Additive manufacturing, respect to traditional methods, makes use of 3D printers, starting from a digital model allows greater freedom in design, with the possibility of creating also very complex and customized geometries (15,20,1), allowing the manufacture of biomedical constructs with a high precision and ensuring faster production and lower production costs (11). Furthermore, 3D printed products require almost only the amount of material needed for the specific object, so there is less waste of material and less need for disposal (9,6).

In addition, AM provides more design control of complex scaffolds, making them customizable and reproducible. A successful implant must be able to restore bone function and promote regeneration of bone in the damaged site (4).

In 1986 Charles Hull devised the first technique of AM: stereolithography (SLA), but later other techniques were developed such as Fused Deposition Modeling (FDM), direct energy laser deposition (DED), Selective Laser Sintering (SLS), production of laminated objects (LOM) and 3D bioprinting (12, 20).

This latter technique allows the fabrication of biological scaffolds. In fact, in 3D bioprinting, layer-by-layer organized positioning of biological materials, biochemical and living cells, a regular control of the positioning of functional components, is used to create 3D structures with mechanical and biological functions. Starting from a computer design, we can print and transplant the tissue or organ that the patient needs. It is a very complex approach and so far, the bioprinting has been reached only of some tissues such as for example skin, bones, cartilage, and cardiac, hepatic and renal tissue (18). In these technologies, different materials can be used, such as steel or different types of titanium, depending of the kind of lesions and its localization. All materials used for medical applications must satisfy three requirements: biocompatibility, mechanical properties, because the designed implants must morphologically mimic and support the formation of bone tissue structure, and, in some case, biodegradability, in such cases when the devices must be resorbable (20, 8, 19). Biocompatibility is the positive interaction between a biocompatible material and living cells or tissues bringing to wound healing, tissue reconstruction and integration (20). For example, cells must adhere, function normally and migrate on the surface (5). This characteristic must be quantified to decrease the patient's risk and implant failure, since metals, in biological systems, can produce corrosion and therefore cause physiological damage (8, 19). The scaffold must have mechanical properties coherent with the anatomical site where it is to be implanted. The implanted scaffold must have sufficient mechanical integrity for proper operation from the time of implantation until the remodeling process (13). Titanium is the most widely used metal biomaterial because it has the desired mechanical properties (20). Finally, biodegradability is also called bio-absorbance. In this case, the scaffold must be biodegradable so as to allow the cells to produce own extracellular matrix (13).

In AM techniques, 3D printing is used as it can provide functional prosthetic implants created specifically for the patient and allows the creation of personified and compatible tissues and organs for patients who need them (18). In recent years, 3D printing was used to manufacture prosthetic limbs in both humans and animals, improving the health of both species (17, 5).

Our aim is to try using 3D printing in veterinary medicine to create customized 3D metal prostheses through the use of Computerized Tomographic imaging. We have developed a three-dimensional prosthetic model adaptable to canine pelvic bones through to the collaboration between the Department of Veterinary Medicine and the Department of Engineering of the University of Perugia.

Material and methods

Collection of images

The pelvic computed tomography (CT) scan of a five years old German shepherd dog suffering for a condition of lumbo-sacral degenerative stenosis and suspected for degenerative hip joint disease was used for the developing of a theorical, complex shape, model for a personalized prosthesis. A volumetric 3D reconstruction of the CT scan series furnished a pelvic model, on which to subsequently perform the design of the acetabular prosthetic model.

Computed Tomography is integrated with software that reconstructs images, modifies them and allows to obtain a 3D reconstruction of the examined body structures.

The parameters that were used in the CT execution were: the slice thickness of 2 mm, the beam energy of 140 KV and 150 mAv, the reconstruction diameter of 250 mm and the convolution matrix of 40.

The file, with CT medical scans, was saved in DICOM format (Digital Imaging and Communications in Medicine).

Obtaining of the 3d model

The pelvic images obtained from the Tomography have been inserted within MIMICS (Materialise Interactive Medical Image Control System), software from which to derive the 3D model of the highlighted anatomical part and which guarantees results faithful to the real condition of the patient. It is necessary to choose the orientation, preferably trying to keep that one used during the execution of the CT, after that the software proceeds with slicing, segmenting the image. Therefore, from the 3D model, selecting the part of bone of interest, the model to be printed was obtained (Fig. 1).

Print the canine pelvic model using the FDM printer

The Stratasys Fortus 400 printer using Fused Deposition Modeling (FDM) technology was used to make the model of the pelvic canine bone.

Inside the software that communicates with the machine, the file has been inserted in .stl format and the parameters have been set, so it is necessary to choose the best orientation of the piece on the work surface. The system inside has a code that chooses the orientation based on the feature to be privileged. For this reason, the piece was realized by orienting it in the Y direction, to have a mechanical resistance at the maximum bending. After choosing the orientation in the MIMICS software that communicates with the printer, the data relating to the chosen material were inserted, which in our case was ABS (acrylonitrile-butadiene-styrene) and therefore the thickness of the layer was decided. During slicing the useful path to the nozzle is created to create the final object and support. The thickness of the layer chosen to obtain the model was 0.25 mm because it was not necessary to have a high level of the thickness. Performed the slicing, for the realization of each layer, the nozzle moves according to precise x and

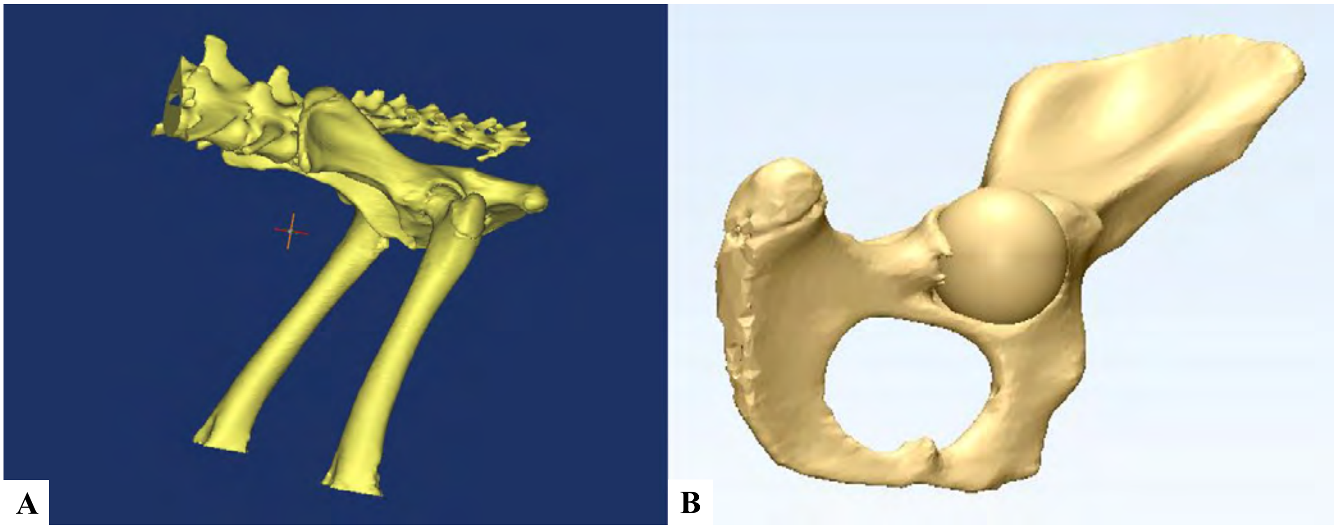

Figure 1. Three-dimensional reconstruction of the studied bone segment.
A. 3D model obtained from Mimics; **B.** Canine pelvis of the subject under examination.

y coordinates, describes the external path, the contours and then fills the internal parts. By lowering the plate it is possible to produce the object to printed layer by layer.

Design of the prosthetic component

The pelvic bone model was used to manually draw the shape of the prosthesis, to be fixed at the site of interest. An acetabular prosthetic plate was created, which wraps around the iliac bone, and the holes around the acetabulum in which to insert specific bone screws for fixation. For the realization of the customized prosthesis, the CAD 3 Matic software was used, marketed by Materialize-Magics. The three-dimensional geometry of the bone, obtained with MIMICS, was transferred to this software where a prosthesis perfectly adapted to the iliac bone was obtained and designed. As a prosthetic printing material, stainless steel was used. It has been decided to create two spherical components separately, which then have been united. The external sphere in contact with the spongy bone is trabeculated, while the internal one is full so as to resist the loads to which the prosthesis is stressed. Trabeculation, once the prosthesis is implanted in the body, can promote osseointegration. Trabeculation is also necessary to reduce the modulus of elasticity but it is also necessary to have a further non-trabeculated portion, which acts as a support for the prosthesis and which allows it to withstand the loads to which the prosthesis will be subjected. In the external part of the acetabulum, in which the holes for the fixation were designed, a dense material reinforcement was inserted, for a better fixing to the iliac bone and to the prosthetic resistance. Within the Materialize-Magics software, trabeculation was realized through the standard program structures. A hexagonal trabeculation was chosen because it guarantees better stability and offers an optimal density to favor osseointegration. The file of the iliac bone and the first prosthetic version were imported into Insight and then processed. Based on the orientation of the iliac bone, the planned acetabular prosthesis was oriented horizontally (Fig. 2). After slicing, printing was done with the prefixed material.

Figure 2. 3D graphic representation of the prosthesis. **A.** & **B.** Isometric view of the prosthetic component;
C. Positioning of the prosthesis during the process of slicing that lead to the finished product.

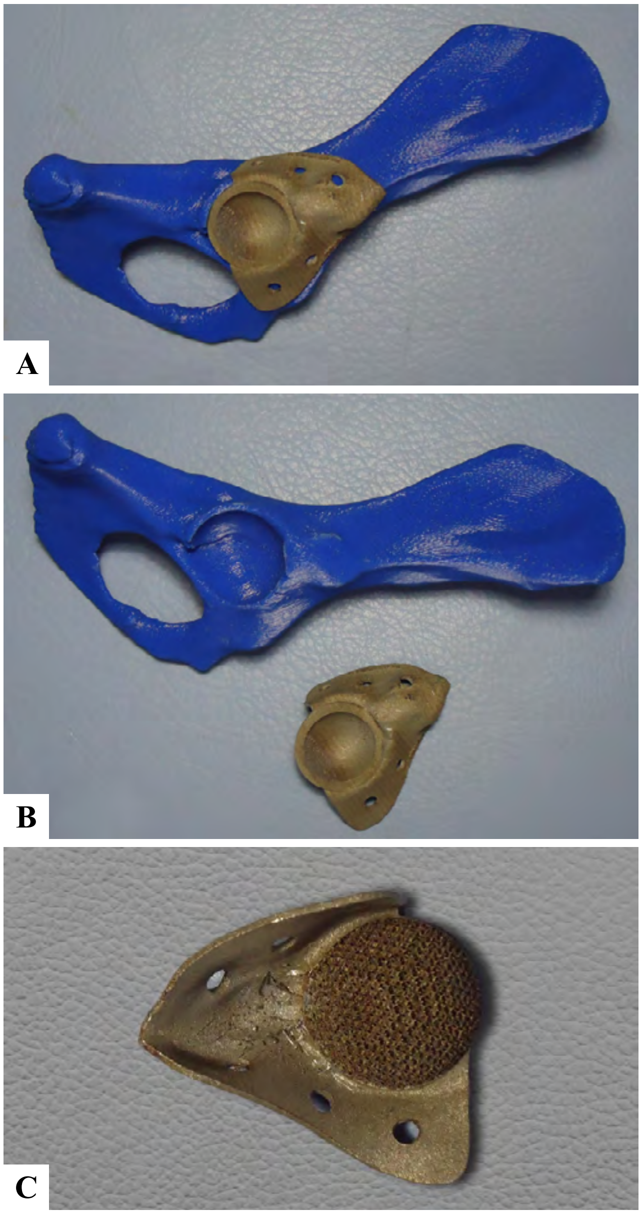

Figure 3. Final construction of the bone segment and acetabular prosthesis. **A.** Prosthesis perfectly adapted to the pelvic model; **B.** Prosthesis and pelvic model; **C.** Acetabular prosthesis made of stainless steel using DMLS technology.

Prosthesis printing by direct metal laser sintering

The machinery that has been used and which uses DMLS (Direct Metal Laser Sintering) technology is "MYSINT 100", designed and manufactured by Sisma Group. It uses the fusion of metals by a laser source of 200 W, building the prosthesis layer by layer.

The prosthesis made, fits perfectly to the plastic model in ABS, which represents the pelvic bone of our patient. The stainless-steel prosthesis has a dust diameter of 20 microns, the part that is located in corresponding at the acetabulum is made of a trabecular structure to promote bone integration and measures 2.5 mm, while the plate that surrounds it measures 1.00 mm. The prosthesis made with 3D printing is personalized for the patient and can be perfectly adapted to the part of the bone to be replaced, moreover, the trabecular geometry is slight, with cellular rooting properties and it has the ability to favor osseointegration (Fig.3).

Prosthetic evaluation of mechanical resistance and stress

In the next phase of the study the simulation of the prosthesis behavior and the interaction it has with the portion of bone on which it is fixed was done. An evaluation of torsion, compression and elongation of the prosthesis was made and the distribution of the areas was also evaluated of greatest stress using "thermoelasticity stress measurement technique". Through a thermal imaging camera, the temperature variations were recorded in order to highlight the mechanical resistance of the prosthesis and the dynamic amplitude of the temperature fluctuation, caused by a dynamic loading of the prosthesis, to measure its stress field. The tests carried out allowed us to identify the areas of maximum stress, and to make a quantitative estimate of the present tension concentrations. Delta-Therm, a full-field, non-contact imaging system used to characterize the behavior of stressed materials and structures was used. Subsequently, an excellent visual correlation of the stresses caused on the prosthesis was obtained from the images. Through the results obtained, the areas most sensitive to stress were evaluated and for this reason reinforcements were created in the areas concerned (red in Fig. 4). Then, through the DMLS printer, a new more resistant prosthesis was made in too sensitive areas.

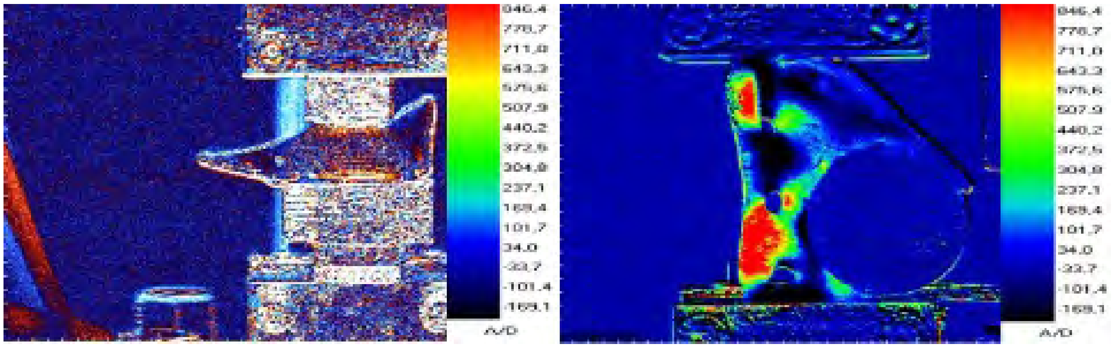

Figure 4. Stress analysis. In red the areas of prosthesis that undergo maximum stress.

Results and discussion

Additive manufacturing allows design freedom to create complex structures and shapes using different materials. These structures can be realized quickly compared to traditional techniques (14). The AM method has as input the realization of the 3D model of the object followed by a semi-automatic file conversion process that involves the decomposition of the object into printable layers from 3D prints (3).

Additive production is today at the base of tissue engineering: with 3D printers it is possible to realize real scaffolds on which to implant cells able to proliferate and populate these structures (13, 21).

From this study we created a three-dimensional prosthetic model adaptable to canine pelvic bones.

The prosthesis made with 3D printing adapts perfectly to the part of the bone that needs to be treated because it was built for the specific patient. In addition to this, in our study we simulated the behavior of the prosthesis and interaction with the portion of bone on which it is fixed. The stress distribution on the prosthesis we made was measured with the thermoelasticity non-contact measurement technique, obtaining a spatial distribution of the temperature variations recorded with the thermocamera to study mechanical resistance, especially when it replaces stressed bone parts. This method allowed us to evaluate the torsion, compression and stretching of the prosthesis and to avoid dangerous concentrations of stress. The relative temperature fluctuations obtained in our study were referred to specific reference signals, with respect to the load cycle and the mechanical component. These techniques allow the use of different materials, such as steel or different types of titanium, all biocompatible and resistant, depending on the type of lesions and it's localization (15). In our study to print the prosthesis we used stainless steel, an iron-based alloy that has the mechanical properties of steels but also a marked resistance to corrosion, a feature mainly due to the presence of chromium which acts as a protection through to the action of an invisible layer of oxides, which on the surface of the steel, make it immune to the attacks of external chemical agents (http://acciaioinossidabile.com/). A limitation is that processing materials such as metal is certainly more complicated than processing plastic materials, so it is necessary to understand how to program machinery and how to optimize production by creating an effective work process (2). Through the results obtained, we were able to evaluate the areas most sensitive to stress by creating reinforcements in the areas concerned and using the DMLS printer we created a new more resistant prosthesis in the points that were too sensitive. In the generation of the prosthesis for the treatment of hip dysplasia, additive manufacturing allows not only the personalization of the prosthesis for each patient, but the optimization of the prosthesis structure, as well as the trabeculated part, hypothetically allowing a better osseointegration, a better adaptation to the stiffness or flexibility of the bone with which the prosthesis comes into contact, reducing the shielding of stress. In the medical field additive production can be a great benefit, both in improving the operating work of the surgeon and in guaranteeing greater validity of the prosthesis on the patient. In recent years there have been significant developments in various areas of additive production (2). In the biomedical sector, the interest of 3D printing is growing with the need for personalized medicine (6). In the future, in fact, the medical sector is the one that will benefit most from both the creation of prostheses and the printing of organs and tissues to be implanted to solve the problem of rejection of the patient's organ (16).

Conclusion

In this study, we realized a complex shape personalized prosthesis that represent a starting point for the developing an applicable one that could be efficient for the resolution of bone diseases in animals, particularly in dogs suffering from hip dysplasia. For the design and construction of the prosthesis we have used 3D printing because it allows the obtaining of prosthetic implants specifically for each patient starting from digital data, guaranteeing superior design.

With this method individual, personalized and functional implant are obtained, both from a chemical and biological point of view, which can replace or supplement the normal activity of damaged tissues both in humans and animals.

The results obtained show an interesting distribution of the stress concentration of the prosthesis to conclude that this measurement methodology represents an innovative system for the evaluation of points that are less resistant to physical stress and interface failure.

From this, we believe that, in the future, more sophisticated routine protocols can be identified that allow prompt intervention and rapid production and installation of artificial prosthetic principles.

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