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The use of adenosine triphosphate bioluminescence for assessing the cleanliness of additive-manufacturing materials used in medical applications

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ABSTRACT

Additive Manufacturing (AM) is widely gaining popularity as an alternative manufacturing technique for complex and customised parts. AM materials are used for various medical applications in both metal and polymer options. Adenosine Triphosphate (ATP) bioluminescence technology is a rapid, user-friendly method of quantifying surface cleanliness and was used in this study to gather quantitative data on levels of contamination on AM materials at three different stage processes: post build, post cleaning and post sterilization. The surface cleanliness of eleven AM materials, three metals and eight polymers, was tested. ATP bioluminescence provided the sensitivity to evaluate different material surface characteristics, and specifically the impact of surface finishing techniques on overall cleanliness.

1. Introduction

There is a clear synergy between the benefits of additive manufacturing (AM) technology and the requirements of patient-specific medical devices. AM parts are best suited to high-value applications that require rapid fabrication of complex geometry. Some of the most challenging medical applications demand bespoke anatomical features to be accurately replicated and delivered in a compressed timescale to meet the needs of trauma surgery. As the field of AM continues to expand then the list of AM-based medical devices is equally likely to grow.

A classification of medical applications of AM by Tuomi et al. [1] divides these applications into five areas: (1) medical models; (2) external aids; (3) surgical guides; (4) surgical implants and (5) biomanufacturing. The range of applications covers the relatively simple task of providing insight to the surgeon/patient (medical models) through to biologically-active tissue implants (biomanufacturing). The area of surgical guides covers patient-specific custom-designed drilling, cutting and repositioning devices, and this area provides an ideal fit with AM technology. Typical guides used in maxillofacial and orthopaedic applications are hand-held (small build volumes), incorporate patient-specific features that engage appropriate internal anatomical structures and can be easily cleaned and sterilised [2,3].

Surgical guides have been fabricated by AM in a range of polymers and metals [2,3]. Recent research within the field of maxillofacial surgery [4] has evaluated the use of AM surgical guides by a range of surgeons. The results show that surgical teams are keen to engage with AM technology but they have a number of pre-conceived perceptions as to the types of materials that are appropriate. It may be that material choice (specifically metal versus polymer) is strongly influenced by experience of previous conventional manufacturing processes, and there is little quantitative data to guide the clinical team for new AM applications. Three areas have emerged that need more empirical evidence to guide surgical decisions in the use of AM materials for surgical guides: geometrical accuracy, surface roughness and cleanliness/sterility. Patient safety is the primary consideration when implementing any new medical intervention, therefore quantifying the cleanliness/sterility of AM materials is the main focus of this research paper.

AM technology and material vendors are continuing to develop a wide range of materials that have the potential for medical applications. For invasive surgical devices and implants, there are a

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series of ISO 10993 standards for the biological evaluation of medical devices that are in permanent (or prolonged) contact with the patient. In these cases criteria on biocompatibility and toxicity take precedent over other material issues. For medical devices that are single-use, disposable items that have limited contact with biological tissue (as in the case of surgical guides) there is a wider choice of potential materials. A typical surgical guide will arrive at the operating theatre within a sterile package, and labelled for a specific patient. The whole medical intervention could last hours but the AM material may only be in contact with the patient for a matter of minutes. In this scenario there are no clear guidelines or specifications to help define cleanliness and sterility.

The whole AM process, in terms of build orientation, cellular elements, removal of support structures and post-processing, provides a number of opportunities to introduce potential contamination into a medical device that could provide a hazard for the end user. Many AM manufacturing processes have fully-prescribed methods for post processing, but there are significant opportunities to detrimentally impact part cleanliness, especially when dealing with complex anatomical-based structures that include small voids that are difficult to fully access with fluids and cleaning implements. Techniques that enable contamination levels to be quantified during the various clinical delivery stages (post-build, post-cleaning and post-sterilisation) of AM medical parts is therefore highly desirable.

Adenosine triphosphate (ATP) bioluminescence technology is a rapid, user-friendly method of quantifying surface cleanliness that has been employed to evaluate contamination of a wide range of instruments and surfaces. Recent studies have used ATP to assess invasive medical devices [5]. hospital surfaces [6] and environmental hygiene monitoring [7]. The bioluminescence test utilises the light-producing reaction between ATP, luciferin and luciferase to measure the amount of ATP present on a surface. ATP is the basic source of energy for all animal and microbial cells; its presence on a surface provides an estimate of all viable and non-viable organic residues, including microbiological contamination. The use of ATP bioluminescence tests is growing within healthcare, pharmaceuticals and food science industries. The ATP technology has two key advantages over traditional microbiological testing. Firstly, the technique provides results within minutes (as opposed to days) and effectively gives a real-time evaluation of surface cleanliness. Secondly, the test apparatus is highly portable and does not need specialist training or dedicated controlled facilities. ATP testing is therefore a very practical technique that can be adopted by non-specialists. The source of ATP can be anything that the sample comes into contact with, for example the way it is handled or where the sample was stored. The ATP method cannot identify the exact source of the contamination.

In the context of medical applications, a measure of residual organic matter is an indicator of surface cleanliness, but also quantifies the potential for surface reservoirs to harbour bacteria, fungi and viruses. Therefore ATP bioluminescence may be employed to give a dual estimate of: (1) the cleanliness of a surface at a fixed point in time; (2) the likelihood that a surface is susceptible to microbiological contamination over a longer period of time.

To date, the use of ATP bioluminescence to measure the cleanliness of AM materials intended for medical use has not been reported. The aim of this paper is to demonstrate that ATP bioluminescence testing is an appropriate technique for quantifying the cleanliness of a range of polymeric and metallic AM samples. It is hoped that the results can be used to highlight which AM materials (and associated surface modifications) have the greatest potential to be used in single-use, disposable medical applications, specifically materials that maintain levels of surface cleanliness that are appropriate for patient-specific surgical guides.

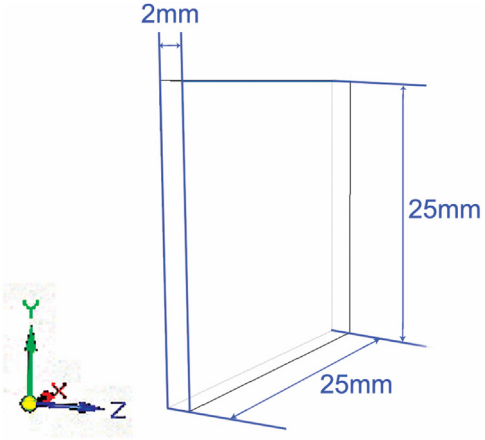

Fig. 1. Dimensions of the material samples—25 mm × 25 mm × 2 mm.

2. Materials and methods

The aim of this study was to evaluate the ATP bioluminescence test in terms of its application to a range of representative AM materials to quantify their surface cleanliness. In this context, material properties are of more concern than geometrical features. The test sample geometry was therefore kept relatively simple, and is shown in Fig. 1. The two 25 × 25 mm square areas were the surfaces of interest for cleanliness/sterility testing, and the majority of samples were fabricated with the (x, y) plane as the up-facing surface. The surface area of the samples needed to be a minimum of 10 × 10 mm to order gain a good enough reading,

Eleven AM materials were chosen to provide a representative sample of polymers and metals that have been employed in a range of medical applications. Details of the AM materials used in this research study are provided in Table 1. Each material category had 12 test samples manufactured. The three metals are all manufactured using Laser Melting (LM) technology, with one of the cobalt chrome set of samples having additional electropolishing finishing. The eight polymer categories can be divided into: three Stereolithography (SLA, 3D-Systems, USA) resins; three polyjet (Objet, Statasys Ltd., Israel) materials; and two Selective Laser Sintering (SLS, EOS GmBH, Germany) materials.

The ATP bioluminescence test employed in this study was the 3 M Clean-Trace system (www.3M.com/infectionprevention). The procedure starts by taking the test swab and applying it to the surfaces to be evaluated. The swab is gently rotated as it is swept across the test area. The swab is then immediately placed in a cylindrical vial, which brings it into contact with the enzyme solution (luciferin–luciferase) and the enzyme reacts with any ATP residue on the swab. The cylindrical vial is then placed in a hand-held 3 M luminometer, and the light generated from the bioluminescence reaction is captured, and the measurement is expressed in Relative Light Units (RLUs). The greater the level of ATP present on the swab, the higher the RLU reading produced. The test can be performed in less than 30 s, providing a real-time indication of the cleanliness of the surface tested. The swab and enzyme solution are disposed of after each test reading.

The 3 M instrument manufacturers recommend a pass/fail threshold of 250 RLUs to indicate part cleanliness [8]. In addition, a literature review by Amodio and Dino [6], covering the period 1990–2012, has shown that the <250 RLUs threshold is the most widely used benchmark for indicating clinical surface cleanliness. In addition a recent Danish standard DS 2451-10 has been monitoring hospital cleanliness with standardised ATP measurements using a hygiene 5 level, the cleanest of the levels, which is set at 250 RLU's [9].