Sterilization of an Electronic Medical Device

Joana C. Reis[a1], Sandra Cabo-Verde[b], Fernando Capela-Silva[c], C. Frias[d] José Potes [a], Rita Melo[b], Inês Nunes[b], Helena Marcos[b], Telma Silva, M. Luisa Botelho[b2]

[a]*Veterinary Medicine Department, University of Évora*
Pólo da Mitra, Ap. 94,7002- 554 Évora, Portugal

[1]jmfcr@uevora.pt
[2]mlb@itn.pt

[b]*Radiation Technologies: Processes and Products Group*
Unit of Physics and Accelerators, Nuclear and Technological Institute
Estrada Nacional 10, 2686-953 Sacavém, Portugal
[c]*Biology Department, University of Évora*
Pólo da Mitra, Ap. 94,7002- 554 Évora, Portugal
[d]*Mechanical Engineering and Industrial Management Department*
Faculty of Engineering, University of Porto (FEUP),
Rua Roberto Frias s/n, 4200-465 Porto, Portugal

Abstract— **Radiosterilization was applied to a medical device with a "programmable memory" to allow *in vivo* implantation. Irradiation on a Cobalt-60 facility at 25 kGy at a dose rate of 2kGy/h corrupted the memory. Therefore an alternative sterilization method using UV was developed and validated based on ISO 11737-1 and ISO 14937. These procedures may be useful and effective for research purposes when only a small number of items might be involved but applicability at an industrial scale is unlikely.**

Keywords— **Programmable memories, ionizing radiation, ultraviolet radiation.**

I. INTRODUCTION

The question of how to effectively sterilize biomedical devices for research, either for in vitro or in vivo use, is of outermost importance. A sterile medical device is the one free from all viable microorganisms. The choice of the sterilization method is dependent on the initial bioburden, on the device characteristics, on environmental and safety considerations [1], [2].

Many materials do not withstand dry heat or autoclave sterilization. High energy irradiation procedures and ethylene oxide may alter physical properties and induce phenomena like aging, degradation, free radical generation, shrinking, changes in hydration capacity and thermal properties [3],[4]. Some of the effects of high-energy sterilization methods on materials may be beneficial for tailoring its properties, but this is not always the case [4]-[6]. Irradiation affects polymers heavily, namely biodegradable polymers, but also exerts its effects on programmable memories and flash memory cells, like those integrating some biomedical devices [7], [8].

Ethylene oxide is associated with environmental pollution and endurance of toxic residues in the polymeric materials [9], [10].

Other sterilization methods are increasingly being applied such as low temperature plasma sterilization. Gas plasma methods allow sterilization at low temperature and low moisture, in a vacuum chamber [11]. Plasmas produce reactive fluxes of ions, atoms and ultraviolet (UV) photons from a given precursor gas that interact with molecules on the surface, including microorganisms. The plasma based inactivation of harmful biological systems is, however, not yet widely used, because method validation is hindered by the limited knowledge about the interaction mechanisms at the interface between plasma and the biological system and the still limited availability of these sterilization systems; plasma-based methods may also induce chemical etching of polymers [12].

Ultraviolet irradiation is a ready available and cost-effective method of expedite surface sterilization and, depending on the bioburden, it is possible to eliminate microorganisms without affecting the material properties. Changes in materials are dependent on dose/rate and time of irradiation [13]. Ultraviolet radiation has poor penetrating power, thus exerting its effects on surface, a clear disadvantage when material geometry is complex.

The analyzed devices were composed by a 16-bit processor, which corrupted its memory when submitted, in a Co-60 source, to 25 kGy at a dose rate of 2 kGy/h and a series of six sensors. On the other hand, it was not possible to sterilize by moist or dry heat since the sensors depolarize at temperatures equal or above 60ºC. The objective of this study was to develop an alternative sterilization method, which ensured absence of toxic residues. The methodology of its development and validation is discussed.

II. Materials and Methods

A. The Devices

The devices aimed to sterilize were composed of a 16-bit processor, powered by lithium battery and encapsulated in polymethylmetacrilate (PMMA) and a set of six sensors/actuators composed of polyvinylidene fluoride (PVDF) and silver electrodes, coated by dip-coating as described by Frias, Reis et al. [14].

B. Bioburden Assessment

The method for bioburden determination was based on the ISO 11737-1 guidelines. Briefly, samples devices were subjected to serial washes using physiological solution with 0.1% Tween and homogeneized by a stomacher equipment. The enumeration of the microorganisms was carried out by direct plating and membrane filtration (nitrocellulose membrane, 0.45 µm) onto Tryptone Soya Agar (Oxoid, UK) petri dishes. The incubation conditions were 30ºC during 14 days. This procedure was validated by repetitive sampling.

The inactivation procedures used to establish an efficient device sterilization method were a combination of physical (UV radiation) and chemical agents (e.g. 70% ethanol; 10% hypochlorite solution; 10% hydrogen peroxide). The inactivation assays were performed using artificial contamination of the samples (spiked samples) using suspensions of devices microbial population of approximately 10^6/sample CFU.

The microbial growth evaluation after exposition to potential inactivation procedures was carried out using the validated bioburden determination method or by immersion of samples devices into Tryptone Soy Broth (TSB) and monitorization of culture medium turbidity during incubation at 30ºC during 21 days.

The microorganisms isolated before and after exposition to inactivation procedures were morphologically characterized by bacteriological conventional techniques.

III. Results

The estimated average devices bioburden was 10^2 CFU/sample and the most frequent types of microorganisms isolated were gram-negative rods (43%) and gram-positive rods (29%).

In a first approach, spiked devices were placed on a sterile surface, 30 cm away from a UV source (UV C germicidal lamp, 15W, 95% of the radiation emitted is around the wavelength of 254 nm), and samples position was exchanged at half-time, allowing exposition to radiation of the two larger surfaces of the devices. After 2 hour of exposition to UV a five decimal log reduction was achieved and a bioburden of one CFU/sample was obtained after 72h of UV. The morphological characterization of the survivor's microorganisms indicated the persistence of grarm –positive spore forming rods and gram-negative rods (the same morphological types isolated from non treated devices).Based on the obtained results the treatment with UV alone was not effective in eliminating the microorganisms present in the spiked samples. A combined decontamination treatment with 70% ethanol and then 6 h exposition to UV was also tested, nevertheless it was verified the survival of a single type of microorganisms classified as gram-negative oxidase negative rods.

In an attempt to have a synergetic effect on the sterilization of the devices, mechanical (e.g. washing), chemical (e.g. bleach, hydrogen peroxide) and physical (e.g. UV), inactivation agents were united in one methodology. Therefore a group of devices (n=3) was subjected to the following treatment: five minute wash in water (21ºC) in a automatic washing machine (Miele Professional G7883), followed by immersion in sterile 10% sodium hypochlorite for 20 minutes, without agitation, washing in sterile water, and immersion in 10% hydrogen peroxide for 30 minutes, without agitation, followed by rinse in sterile water and drying under laminar flux, before 2 hour of exposure to UV (in two different positions). The devices subjected to this treatment, presented no microbial growth after 21 days of culture in TSB. The proposed sterilization treatment was applied to devices that were efficiently implanted in sheep. Post-operative clinical data and post-mortem histological study excluded the presence of infection.

IV. Discussion

The developed and validated treatment was effective, for the experimental conditions, by its combination of procedures; the action of washing, chemicals and UV radiation act synergistically. The initial washing procedure aimed lowering of initial bioburden by mechanical reduction. Spore forming bacilli are known to be resistant to UV radiation, and to have variable levels of resistance to killing by hypochlorite and hydrogen peroxide. Spores are more resistant than growing cells to UV radiation at 254 nm, mainly due to DNA repair mechanisms and DNA protection by α/β-type small acid-soluble spore core proteins (SASP) [15]. Resistance to hydrogen peroxide seems to depend on an intact spore coat, SASP and maintenance of the spore core water content [16], [17]. Hypochlorite kills spores by membrane damage, impairing germination [18] and known resistance mechanisms are related to SASP and repair mechanisms [19]. The treatment with hypochlorite previous to hydrogen peroxide probably induced enough coat and inner membrane damage to potentiate hydrogen peroxide action and facilitate UV action.

Spore pre-treatment with oxidizing agents is known to increase susceptibility to agents that induce DNA damage [20].

V. Conclusions

The present study exemplifies how substitute sterilization protocols may be developed when more conventional and proved sterilization methods cannot be used, either due to technical issues related to the items to be sterilized, or unavailability of other means. Alternative methods to sterilize may be developed, especially for research purposes when only a small number of items might be involved but they are seldom adaptable to large-scale production lines.

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