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OLLSCOIL NA hÉIREANN
GAILLIMH

NATIONAL UNIVERSITY OF IRELAND
GALWAY

SPRING EXAMINATIONS 2001

M.Sc. in BIOMEDICAL SCIENCE

BM 501: Introduction to Biomedical Science

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Time allowed : **ONE** hour.

Answer one question.

Q.1 The Human Genome project has recently announced the full sequence of the human genome. Two separate groups were involved in elucidating the full sequence. Describe the reasons behind the project to elucidate the genome, and possible consequences of this achievement. Mention, in your answer, the significance of two separate groups performing the sequencing.

Q.2 Ethical scientific practices are important not only in that they ensure scientists think about the consequences of their actions, but also to protect against fraudulent practices. Discuss these issues. Where do our ideas on ethics come from?

Q.3 Discuss the current state of biomedical research in Ireland. Suggest the strengths and weaknesses, and what areas might be important in the future.

Q.4 Briefly discuss any two of the topics discussed in the weekly Thursday seminar series.

Q.5 Read the introduction and methods from a paper for journal submission, and then answer the questions (a) – (d) at the end of the text.

Introduction

Calcium phosphate coatings (CPC) are widely used to enhance the ability of metal implant surfaces to facilitate bone bonding [1,2]. These coatings can accelerate bone-implant bonding and improve the final results of surgery. Over the past several years a new method, laser ablation of hydroxyapatite (HA) targets has been developed for CPC deposition [3-10]. The technique of pulsed laser deposition (PLD) has some advantages over conventional thermal and

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plasma spraying methods. Coatings deposited by PLD are dense, thin (~ 1μm), highly adhesive, and can be deposited without elevating the temperature of the substrate above room temperature. CPC characteristics can be precisely controlled by various deposition parameters, such as laser fluence at the target, target density and composition, residual pressure in the vacuum chamber and distance between target and substrate [6].

In this paper we present the experimental results of the effect of HA target density on physical properties of, and cell responses to, CPC's deposited on Ti substrates using a KrF laser at room temperature.

Materials and Methods

Coatings

Coatings (approximately 1-3 μm in thickness) were deposited on Ti-foil by laser ablation of HA targets in a vacuum chamber at room temperature and 2 Pa residual air pressure. KrF laser (EMG203, Lambda Physik) was used at a pulse repetition rate of 10 Hz at a wavelength (λ) of 248 nm and pulse duration of 30 ns. The focused laser beam was scanned across the target surface to minimise the negative effect of crater formation. Laser fluence on targets was varied in the range 3 - 9 J cm^{-2}. The process was performed in a vacuum chamber at 2 Pa pressure and at RT.

Coating characterisation.

The coatings were characterised using adhesion scratch tests, scanning electron microscopy (SEM) and energy dispersive X-ray microanalysis (EDX). SEM and EDX were performed using a Jeol scanning electron microscope. The evaluation of the HA biological properties were made on the basis of the study of HA erosion kinetics in phosphate buffered saline (PBS) solution and measurements of osteoblast growth.

Cell Culture experiments

The cells used were primary derived human osteoblasts (HOBs), obtained from the femoral head of trabecular bone. The cells were maintained in Dulbecco's Modified Eagles' Media (DMEM) (Gibco Life Technologies, Paisley, Scotland) supplemented with 50 IU ml^{-1} penicillin, 0.05 mg ml^{-1} streptomycin, 2 mM glutamine, 20 mM HEPES buffer, 1% non-essential amino acids (all Gibco BRL), 10% foetal calf serum (FCS) and 50 mg l^{-1} ascorbic acid (BDH, Poole UK). Cultured cells were taken from a subconfluent flask, and 2 x10^4 cells per well were added to each sample. The plate was incubated at 37 ^{o}C and 5% CO_2 for 4 hours. The samples were then removed from the wells and carefully washed in phosphate buffered saline (PBS). After 48 hours the samples were washed in PBS, placed into fresh, sterile wells and a 1 in 10 dilution of alamar Blue dye (Serotec, UK) in Hank's Balanced Salt Solution (GIBCO BRL, Paisley, Scotland) added. The plates were incubated for 1 hour before the dye was removed, placed into fresh wells, and measured using a Cytofluor fluorimeter at 530 nm excitation wavelength and 590 nm emission wavelength.

(a). Describe the controls that should be included in this experiment. What would such controls show?

(b). What other information should be included in the methods? How would this influence the experiment?

(c). What practical steps are generally taken to ensure that experiments have been performed correctly and no bad or fraudulent practice has occurred?

(d). 4 Targets, A, B and C were used, A being linked with higher cell activity, and having the highest initial density. Surface roughness of all the produced films was measured, but did not suggest any affect on cell activity. Write an abstract for this study (no more than 200 words) and suggest a title.