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The effect of adsorbed fibronectin and osteopontin on macrophage adhesion and morphology on hydrophilic and hydrophobic model surfaces

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ABSTRACT

Macrophages play a crucial role in the host response to biomaterials. Here we investigated the effect of adsorbed fibronectin (FN) and osteopontin (OPN), two important proteins for tissue repair, on macrophage adhesion and morphology. Since cell–biomaterial interactions are modulated via proteins adsorbed onto biomaterial surfaces, FN and OPN were adsorbed on model self-assembled monolayers (SAMs) of alkanethiols on gold with different functional terminal groups (CH_3, OH and tetra(ethylene–glycol)). The initial interaction of inflammatory cells with a biomaterial is crucial for the ensuing phases of an inflammatory reaction. For this reason short-term cultures of primary human macrophages were performed. To account for the competitive adsorption of other proteins serum was added to the culture medium and the effect compared with serum-free medium cultures. In the presence of serum hydrophilic surfaces increased macrophage adhesion. In particular, FN induced a higher cell density, while OPN tended to decrease it. In serum-free medium cell adhesion was greater on hydrophobic surfaces, except for OPN-coated SAMs. Importantly, FN no longer enhanced macrophage adhesion, while OPN maintained its inhibitory effect. Cell polarization studies indicated that macrophage morphology variations induced by surface chemistry are overcome by pre-adsorbed OPN. Taken together our results show that in the presence of serum macrophage adhesion is promoted by FN hydrophilic surfaces, but impaired on OPN-coated surfaces. The effects of inhibited macrophage adhesion on macrophage fusion, and its relevance to the initial stages of the inflammatory response to biomaterials are discussed.

1. Introduction

Implantation of a biomaterial into the body immediately triggers an inflammatory response. Macrophages are amongst the first cells arriving at the injured site, but, like other cells, they do not directly interact with the biomaterial surface. It is widely accepted that cell–biomaterial interactions are mediated by blood proteins that rapidly adsorb to the device upon implantation [1]. However, biomaterial surface properties, including surface chemistry, charge, topography, and hydrophobicity, have been shown to influence protein adsorption and consequently adhesion and activation of many cell types, including macrophages [2–6]. Based on this knowledge, novel biomaterial surfaces have been designed for tissue engineering and regenerative medicine, which may involve modification of the surfaces so that they elicit specific cellular responses. In this context, monocyte/macrophage responses, such as cell adhesion, morphology, foreign body giant cell (FBGC) formation and apoptosis, have been investigated after contact with several substrates [7–10], as well as in the presence of different molecules and proteins adsorbed to those surfaces [11–15]. IgG pre-adsorption was shown to enhance the adhesion of macrophages and FBGC formation, while von Willebrand factor had an inhibitory effect on long-term macrophage adhesion [13,14]. On the other hand, complement protein C3 is critical in mediating the initial adhesion of macrophages on medical grade poly(ether urethane urea) [16]. Surfaces grafted with tripeptide RGD sequences also supported higher macrophage densities than those grafted with other peptides [16]. In general, protein–biomaterial interactions mediating macrophage adhesion are complex and not fully understood.

Fibronectin (FN) is a high molecular weight (450 kDa) dimeric glycoprotein found in the extracellular matrix (ECM). It is the major adhesion-promoting protein of interstitial tissues [17,18]. In addition to its influence on cell attachment and spreading, FN plays an important role in tissue repair [17–19]. Cells adhere to FN mainly through integrins, which recognize the Arg–Gly–Asp (RGD) amino acid sequence [20]. RGD-independent cell binding

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