Colloids and Surfaces B: Biointerfaces 155 (2017) 71–82

Contents lists available at ScienceDirect

Colloids and Surfaces B: Biointerfaces

journal homepage: www.elsevier.com/locate/colsurfb

Comparative surface energetic study of Matrigel® and collagen I interactions with endothelial cells

Michael J. Hill[a], Debanjan Sarkar[a,b,*]

[a] Department of Biomedical Engineering, University at Buffalo, The State University of New York, Buffalo, NY, 14260, USA
[b] Department of Chemical and Biological Engineering, University at Buffalo, The State University of New York, Buffalo, NY, 14260, USA

ARTICLE INFO

Article history:
Received 6 February 2017
Received in revised form 28 March 2017
Accepted 3 April 2017
Available online 4 April 2017

Keywords:
Collagen
Matrigel
Endothelial
Surface
Polyurethane
VOGCT

ABSTRACT

Understanding of the surface energetic aspects of the spontaneously deposited proteins on biomaterial surfaces and how this influences cell adhesion and differentiation is an area of regenerative medicine that has not received adequate attention. Current controversies surround the role of the biomaterial substratum surface chemistry, the range of influence of said substratum, and the effects of different surface energy components of the protein interface. Endothelial cells (ECs) are a highly important cell type for regenerative medicine applications, such as tissue engineering, and In-vivo they interact with collagen I based stromal tissue and basement membranes producing different behavioral outcomes. The surface energetic properties of these tissue types and how they control EC behavior is not well known. In this work we studied the surface energetic properties of collagen I and Matrigel® on various previously characterized substratum polyurethanes (PU) via contact angle analysis and examined the subsequent EC network forming characteristics. A combinatorial surface energy approach was utilized that compared Zisman's critical surface tension, Kaelble's numerical method, and van Oss-Good-Chaudhury theory (vOGCT). We found that the unique, rapid network forming characteristics of ECs on Matrigel® could be attributed to the apolar or monopolar basic interfacial characteristics according to Zisman/Kaelble or vOGCT, respectively. We also found a lack of significant substratum influence on EC network forming characteristics for Matrigel® but collagen I showed a distinct influence where more apolar PU substrata tended to produce higher Lewis acid character collagen I interfaces which led to a greater interaction with ECs. Collagen I interfaces on more polar PU substrata lacked Lewis acid character and led to similar EC network characteristics as Matrigel®. We hypothesized that bipolar character of the protein film favored cell-substratum over cell-cell adhesive interactions which resulted in less rapidly forming but more stable networks.

1. Introduction

In vivo, endothelial cells (ECs) contact two distinct classes of extracellular matrix (ECM): their own secreted basement membranes (BM) and stromal tissue. BM is a thin proteinaceous layer consisting mainly of collagen IV and laminin which separates differentiated cell types from stromal tissue under physiologically normal conditions [1]. Under pathological conditions ECs must invade the stromal tissue for angiogenesis to ensue. A possible pathway for this process involves thinning or injury of the BM which allows for ECs to contact stromal tissue consisting mainly of structural proteins collagen I/III (among others), which induces cell proliferation and nascent blood vessel formation [2]. Fig. 1a depicts this series of events schematically. Increased understanding of the physics of this process has implications for not only the fields of tissue formation and stability but also regenerative medicine applications such as tissue engineering (TE) where isolated ECM components are often used as structural supports for ECs and other cell types. Despite the importance of this, the physical cause of these differential contact relations (EC-BM, EC-stromal) and disparate physiological outcomes has not been established. BM molecules appeared more recently in metazoan evolution than the interstitial ECM proteins and their unique physical properties likely played a role in the increasing differentiation of the evolutionary tree [3]. Therefore, increased understanding of the function of BM whether as a mechanical or energetic barrier is desirable, not only for the understanding of angiogenesis, but also the relationship of BM to the differentiated state, in general.

* Corresponding author at: Department of Biomedical Engineering, University at Buffalo, The State University of New York, Buffalo, NY, 14260, USA.
E-mail addresses: debanjans@gmail.com, debanjan@buffalo.edu (D. Sarkar).

http://dx.doi.org/10.1016/j.colsurfb.2017.04.004
0927-7765/

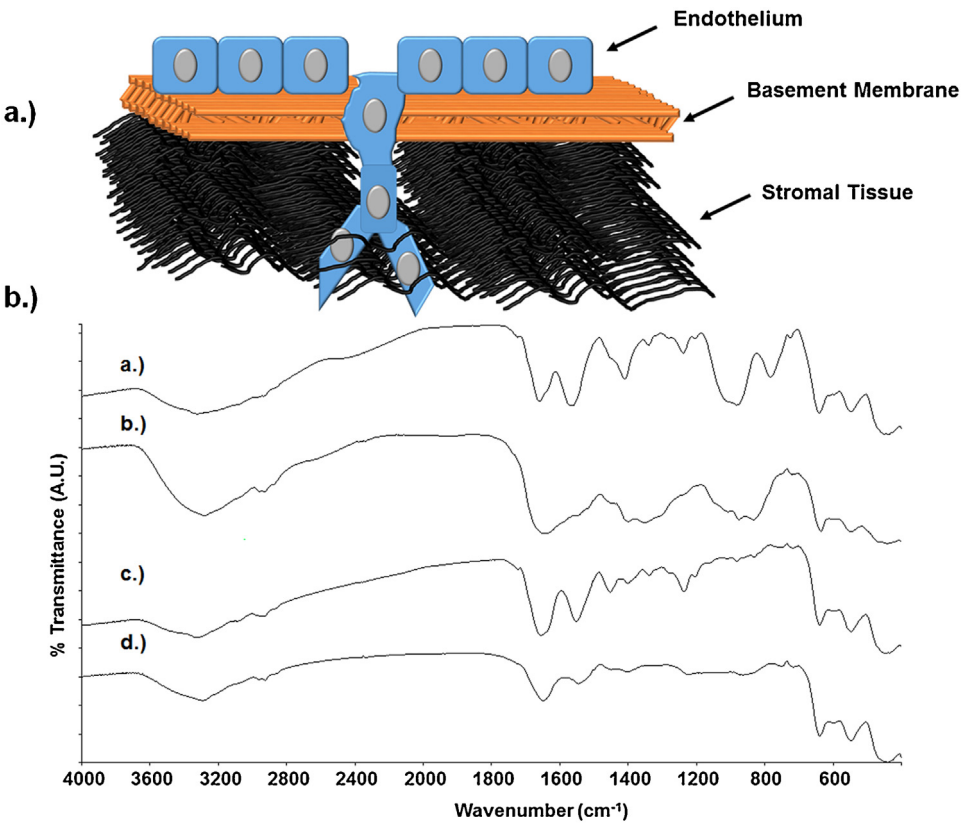

Fig 1. Process of Angiogenesis and IR Spectra of Proteins: a.) Schematic Representation of Angiogenesis Process: As basement membrane (BM) thins or becomes damaged EC begin to contact stromal tissue which induces proliferation, invasion, and angiogenesis b.) IR spectra of protein samples where a.) Collagen I prepared for cells b.) Matrigel® prepared for cells c.) Collagen I after DI water rinse d.) Matrigel® after DI water rinse.

Previously it has been empirically observed that ECs proliferate, migrate, or differentiate based on the ECM components they are in contact with. For instance, when cultured on equal concentrations of collagen I/III (stromal) and collagen IV/V (BM) the stromal type proteins resulted in EC monolayer formation while the BM proteins resulted in differentiated morphology [4]. When ECs were cultured on the BM side of isolated amniotic membranes they formed capillary networks whereas, when seeded on the stromal side, migration and proliferation ensued. Furthermore, it is well known that when cultured on tissue culture polystyrene (TCP) coated with Matrigel®, the commercially available BM protein mixture model, ECs rapidly form capillary networks (in-vitro angiogenesis), while they form monolayers when cultured on serum coated TCP [5]. These examples do not account for the effects of differential substratum chemistries or protein concentrations. For instance, thin Matrigel® films on glass and TCP differentially influenced the "stemness" of human embryonic stem cells (HESCs) and it was found that each substratum induced Matrigel® interface had different rheological properties [6].

One approach to understanding the role of ECM components in controlling cell adhesion and morphological characteristics has been the estimation of the non-specific intermolecular force characteristics of adsorbed proteins or tissues via contact angle measurements [7–9]. This is in contrast to the "biological approach" where the interactions between specific ligands and receptors are analyzed in a reductionist framework [10–12]. The complexity of the protein adsorption process and the interactions between ECM components and different substratum materials used for cell culture or TE applications warrants a broad outlook in terms of characterizing cell/protein/material interactions at different length scales in opposition to the rather "short-ranged" specific effects [13,14]. For instance, it is well known that substratum chemistry influences cell adhesion on adsorbed protein films, however, the interpretation of this data is conflicting [15]. There is some research indicating that passivation of the substratum influence occurs after just a few layers of deposited protein (albumin, thin-layer immunoassays, etc.) [11,16,17]. However, previous work has suggested that the substratum influence on attaching cells can extend over thousands of Å's of intervening deposited/adsorbed molecules, depending on substratum chemistry [18]. Estimation of differential interfacial energetics between substratum-ECM and cell-ECM is therefore an important and underutilized field of research in regenerative medicine.

In this study, we utilized contact angle measurements to estimate the surface energy of collagen I and Matrigel® interfaces (stromal and BM ECM type respectively) induced by substratum polyurethanes (PU) with differential surface chemistries. The versatility of PU stems from the two step-reaction which allows variation in the molecular weight (M.W.) of the polyol to control phase-characteristics and resultant macroscopic surface energy. The biocompatible PU family used consists of polycaprolactone (PCL) based soft-segment with aliphatic hexamethylene diisocyanate (HDI) and L-tyrosine based chain extender desaminotyrosyl tyrosine hexyl ester (DTH) based hard-segment. This family of pseudo poly (amino acids) was previously used to investigate the interfacial energetics of serum coated PU and the influence on EC behavior and morphological characteristics [19]. A combinatorial surface energetics approach based on contact angle analysis was pursued to account for the inherent heterogeneity of the PU surfaces, in the search for general empirical methods which can compare a wide variety of physically ideal (perfectly smooth, inert, chemically homogeneous) and non-ideal biomaterial surfaces [20]. It was