

# Phenotypic Heterogeneity of the Endothelium

## II. Representative Vascular Beds

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**Abstract**—Endothelial cells, which form the inner cellular lining of blood vessels and lymphatics, display remarkable heterogeneity in structure and function. This is the second of a 2-part review on the phenotypic heterogeneity of blood vessel endothelial cells. The first part discusses the scope, the underlying mechanisms, and the diagnostic and therapeutic implications of phenotypic heterogeneity. Here, these principles are applied to an understanding of organ-specific phenotypes in representative vascular beds including arteries and veins, heart, lung, liver, and kidney. The goal is to underscore the importance of site-specific properties of the endothelium in mediating homeostasis and focal vascular pathology, while at the same time emphasizing the value of approaching the endothelium as an integrated system. (*Circ Res.* 2007;100:174-190.)

**Key Words:** endothelium ■ endothelial cells ■ heterogeneity

Endothelial cells (ECs) form the inner lining of blood vessels and lymphatics. Blood vessel endothelium traverses each and every tissue and, thus, transcends all clinical disciplines. Each vascular bed has unique structural and functional properties, and an understanding of these properties holds important clues to site-specific diagnostics and therapeutics. This is the second part of a 2-part review on phenotypic heterogeneity of blood vessel endothelium. The first part discusses the history of the field, examples of site-specific differences in endothelial structure and function, and proximate and evolutionary mechanisms of EC heterogeneity. In this part, each of these themes is discussed in the context of individual vascular beds or organs. Instead of touching superficially on each and every vascular bed, the aim here is to focus, in more detail, on a smaller number of vascular beds, including arteries and veins, the heart, lung, liver, and kidney. The goal is to highlight differences, while at the same time emphasizing the value of approaching the endothelium as an integrated system.

### Arteries and Veins

Although arteries and veins both function as conduits and are lined by continuous nonfenestrated endothelium, they differ in fundamental ways (Figure 1). Arteries have thick walls, and they pulsate. Veins have thin walls and do not pulsate. Veins have valves; arteries do not. Endothelial junctions in arteries are tighter compared with those in veins.<sup>1</sup> Arteries carry well oxygenated blood, whereas veins contain deoxygenated blood. An exception is the pulmonary circulation, where the oxygenation status is reversed. Compared with arteries, large veins have a greater capacity to mediate an

inflammatory response.<sup>2</sup> Discrete regions of the arterial tree, including branch points and large curvatures, are exposed to disturbed flow. These areas are primed for activation and serve as “hot spots” for inflammation, coagulation, and atherosclerosis<sup>3–5</sup> (and reviewed elsewhere<sup>6</sup>).

Arteries and veins express unique molecular markers. Genes that are preferentially expressed in arterial ECs include ephrinB2,<sup>7</sup> Delta-like 4 (Dll4),<sup>8</sup> activin-receptor-like kinase 1 (Alk1),<sup>9</sup> endothelial PAS domain protein 1 (EPAS1),<sup>10</sup> Hey1 and Hey2,<sup>11</sup> neuropilin 1 (NRP1),<sup>12</sup> and decidual protein induced by progesterone (Depp).<sup>13</sup> Venous EC-specific genes include EphB4,<sup>14</sup> neuropilin 2 (NRP2),<sup>15</sup> and COUP-TFII.<sup>16</sup> A recent study demonstrated that class III  $\beta$ -tubulin is expressed in ECs at the tip of venous valves, but not in the vein proper.<sup>17</sup>

The extent to which site-specific phenotypes of venous and arterial ECs are fixed or reversible remains a matter of conjecture. Studies in zebrafish embryos suggest that many artery- and vein-specific properties are epigenetically programmed before the onset of blood flow (reviewed elsewhere<sup>18</sup>). These findings have been used to argue against a role for hemodynamics in mediating arterial/venous identity. However, studies in avian models suggest that the arterial and venous phenotypes maintain a degree of plasticity.<sup>19,20</sup> The importance of the microenvironment in governing venous/arterial identity is further supported by cell culture studies in which flow induces an arterial phenotype in venous ECs.<sup>21,22</sup>

Coronary artery bypass grafting provides potential insights into the plasticity of venous endothelium. When veins are grafted into the arterial circulation, they acquire arterial-like properties, including a thickened wall and, in animal models,

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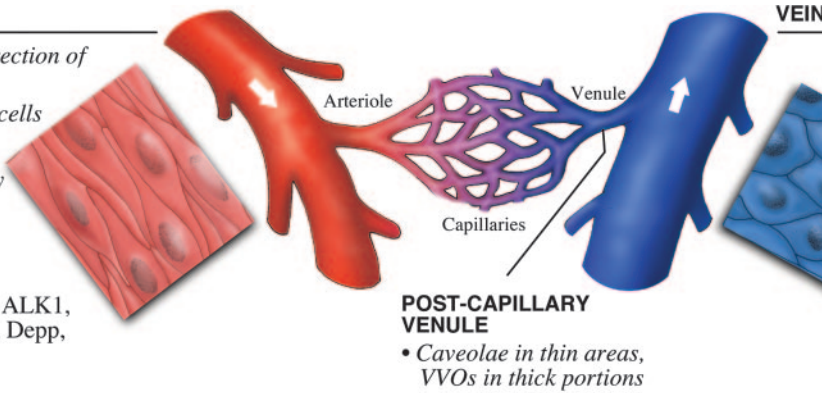
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**ARTERY**

- ECs aligned in direction of undisturbed flow
- Long and narrow cells
- Continuous endothelium with many tight junctions
- No valves
- Specific markers  
Ephrin B2, Dll4, ALK1, EPAS-1, Hey1/2, Depp, NRP1

**VEIN**

- Continuous endothelium
- Shorter, wider cells
- Not aligned in direction of blood flow
- Possess valves
- Specific markers  
EphB4, NRP2, COUP-TFII

**POST-CAPILLARY VENULE**

- Caveolae in thin areas, VVOs in thick portions

**CAPILLARY**

- More caveolae compared to artery and vein (except for the blood-brain barrier)
- ECs highly adapted to underlying tissues
- Many phenotypic differences between different vascular beds

TYPES		
CONTINUOUS		DISCONTINUOUS
Non-fenestrated	Fenestrated	
e.g. muscle; lung; skin; blood brain barrier	e.g. kidney glomerulus; gastrointestinal tract	e.g. liver; marrow sinus

**Figure 1.** ECs in arteries, veins, and capillaries. Shown are selected phenotypic differences between ECs in arteries, veins, postcapillary venules, and capillaries. ALK1 indicates activin-receptor-like kinase 1; Depp, decidual protein induced by progesterone; Dll4, delta-like 4; EPAS-1, endothelial PAS domain protein 1; NRP1, neuropilin 1; TE, transendothelial; VVOs, vesiculo-vacuolar organelles.

reduced permeability.<sup>23</sup> Venous arterialization is initially adaptive but may ultimately result in graft failure. Excised human saphenous veins exposed to arterial flow conditions demonstrated increased endothelial nitric oxide synthase (eNOS) protein<sup>24</sup> and reduced thrombomodulin.<sup>25</sup> In a rabbit model of jugular vein engraftment into the carotid artery circulation, thrombomodulin expression was downregulated by a wall tension-dependent mechanism.<sup>26</sup> Stress regulatory pathways in porcine vein grafts have also been implicated in the downregulation of prostacyclin production.<sup>27</sup> Human saphenous veins perfused ex vivo under arterial flow conditions produced increased matrix metalloproteinase-2 and -9.<sup>28</sup> In a related model, transplantation of the pulmonary valve into the aortic position in humans resulted in new expression of the arterial marker ephrinB2.<sup>29</sup> Taken together, these and other studies provide evidence that venous-arterial identity is mediated—at least to some extent—by differences in the microenvironment.

### Postcapillary Venules

Some postcapillary venules have endothelial-lined bicuspid microscopic valves.<sup>30</sup> They are identical in structure, location, and orientation to the valves of the larger veins, with the exception that their leaflets lack fibroblasts and myofibroblasts. Interestingly, the distribution of valves varies in different vascular beds. For example, in the legs, they are more frequent in venules overlying bone than muscle, the highest number being found in the big toe.<sup>30,31</sup>

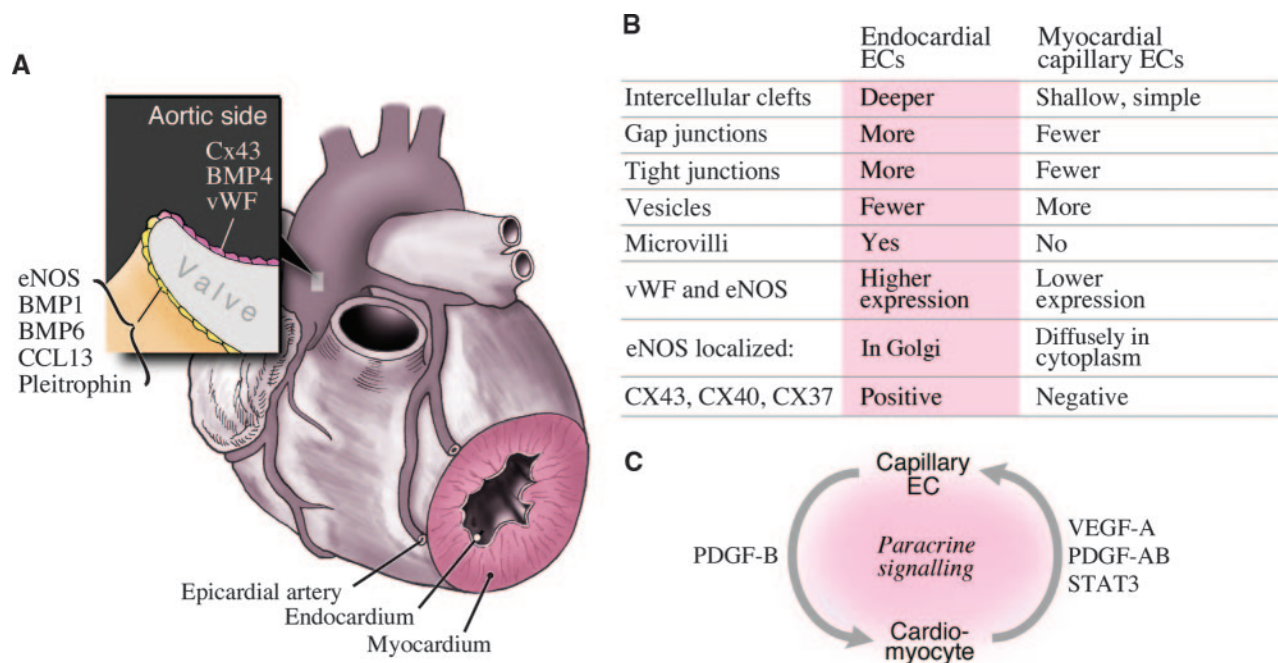
ECs of postcapillary venules are rich in vesiculo-vacuolar organelles (VVOs), particularly in the thicker portions of the blood vessels.<sup>32</sup> The intercellular junctions in the thinned areas are simple, short, and straight, whereas they are interdigitating in the thick part of the venules.<sup>32</sup>

Postcapillary venules are the preferred site for leukocyte trafficking and permeability in states of inflammation. In addition to leukocytes, platelets are also able to roll on activated postcapillary venular endothelium.<sup>33,34</sup> Hypercholesterolemia, hemorrhage shock, and ischemia/reperfusion causes increased platelet adhesion to postcapillary venules.<sup>35–37</sup> Colocalization of platelets and leukocytes is also observed in transient retinal ischemia, experimental models of chronic arterial hypertension, and sickle cell disease.<sup>38</sup>

An interesting exception to the prototypical EC phenotype of postcapillary venules is found in the high endothelial venules (HEVs) of secondary lymphoid organs. ECs in HEVs demonstrate unique structural, molecular, and functional properties. For example, compared with other ECs in the body, ECs lining HEVs are tall or cuboidal in shape. Moreover, they express a repertoire of site-specific adhesion molecules and chemokines that promote constitutive trafficking of lymphocytes between blood and lymph node (reviewed elsewhere<sup>39,40</sup>).

### Capillaries

Capillaries are the major exchange vessels in the circulation. The diameter of capillaries throughout the body is less than



10  $\mu$ m. In keeping with Fick's law of diffusion, capillaries comprise the majority of the surface area of the circulation. Moreover, their wall is extraordinarily thin, thus minimizing diffusional path length. Indeed, capillaries are essentially 3D tubes of flat ECs surrounded to a variable extent by occasional pericytes and extracellular matrix. Also, blood flow is slow through capillaries (it is said to "seep" rather than flow<sup>41</sup>), which optimizes the time for diffusion. More than any other vessel type, capillary ECs are uniquely adapted to the underlying tissue environment. Vascular bed-specific differences in the structure and function of capillary endothelium are discussed below, under the sections describing individual organ systems.

## Heart

The heart contains several endothelial compartments, including ECs of coronary arteries, capillaries, and endocardium (Figure 2). These subtypes of ECs differ in developmental origin, structure and function.

### Endocardium

The endocardium, which forms the inner lining of the heart chambers, arises (together with cardiomyocytes) from the cardiogenic mesoderm, which in turn derives from the rostral portion of the primitive streak. The endocardium and cardiomyocytes form the heart tube, well before the appearance of the epicardium and coronary vasculature. Endocardial ECs are larger than other types of ECs. They possess many microvilli, which project into the heart cavity. The luminal surface of the heart contains many trabeculae and furrows. Together, these features provide the endocardium with mark-

edly increased surface area. Compared with myocardial capillaries, the endocardium has more complex intercellular clefts (deeper and more tortuous), an increased number of gap junctions, and fewer vesicles (reviewed elsewhere<sup>42</sup>). Consistent with the relative abundance of gap junctions, Cx43, Cx40, and Cx37 are expressed by endocardium but not myocardial capillary endothelium.<sup>43</sup>

Like the pulmonary circulation, the endocardium is perfused by the entire blood volume. Indeed, it has been characterized as a "sensor device" for circulating blood entering and leaving the pulmonary circulation.<sup>42</sup> In addition, the endocardium modulates cardiac contractility, rhythmicity, and growth via paracrine and autocrine signaling (reviewed elsewhere<sup>42,44</sup>). Expression of von Willebrand factor (vWF) and eNOS is higher in endocardium compared with myocardial microvessels.<sup>45,46</sup> In endocardial ECs, eNOS is highly concentrated in the Golgi body, whereas in myocardial capillary ECs, eNOS is more diffusely distributed in the cytoplasm.<sup>47</sup>

### Heart Valves

During development, a subpopulation of endocardial cells participates in endocardial cushion formation, giving rise to the septum and heart valves. These cells have been shown to express JB3<sup>48</sup> and the "slug" transcription factor.<sup>49</sup> Heart valve development involves the swelling and deposition of extracellular matrix. Signals from myocardium overlying the heart valve region stimulate ECs in to undergo endocardial-to-mesenchymal transformation, in which they lose cell-cell contacts, delaminate, and invade into the extracellular matrix to form endocardial cushions. Only those ECs within this



region are capable of responding to these signals.<sup>50</sup> Previous studies have implicated Alk-2,<sup>51</sup> Notch-jagged,<sup>52</sup> and vascular endothelial growth factor (VEGF)/NF-AT signaling (reviewed elsewhere<sup>53,54</sup>) in this process.

The endocardial lining of mature heart valves displays phenotypically distinct phenotypes. Transcriptional profiles of passage-5 porcine aortic ECs and porcine aortic valvular ECs cultured in the absence or presence of flow revealed significant differences between the 2 cell types.<sup>55</sup> Several studies have demonstrated differences in gene expression on the 2 sides of the aortic valve, which are subject to vastly different flow dynamics. One study demonstrated higher expression of eNOS on the ventricular side of porcine aortic valves.<sup>55</sup> (Another group reported the opposite finding.<sup>56</sup>) In rat heart valves, Cx43 is preferentially expressed on the upstream surfaces.<sup>57</sup> Transcription profiling of porcine aortic valve leaflets has demonstrated significant side-specific differences in gene expression.<sup>56</sup> Interestingly, some of these differences were preserved in multiply passaged cells, suggesting that they are epigenetically programmed (ie, mitotically stable).

### Coronary Vessels

Coronary arteries originate from the ascending aorta, with the ostia of the left and right coronary artery located in the sinuses of the aortic valves. Epicardial arteries give rise to small muscular arteries that penetrate the myocardium. These vessels, termed intramural arteries, ultimately branch into arterioles and a capillary network that surrounds the cardiomyocytes. Veins travel with arteries more than the surface of the heart. However, their path diverges when venous blood drains into the coronary sinus and into the right atrium. Coronary artery ECs derive from mesoderm-derived proepicardium (reviewed elsewhere<sup>58</sup>). At embryonic day 10.5 (E10.5) in mice, the proepicardium makes contact with the surface of the developing heart tube and undergoes directed migration to form a continuous epithelial sheet around the heart. Some of these cells undergo epithelial-to-mesenchymal transition, giving rise to endothelium and vascular smooth muscle cells. The proepicardium-derived mesenchymal cells then coalesce to form channels and become the endothelium of the epicardial vessels. In addition, these cells invade the myocardium and ultimately form the endothelium of myocardial resistance vessels and capillaries. The endothelium of the coronary conduit and resistance vessels is primarily responsible for controlling coronary supply to the myocardium. In many ways the coronary arteries, including their endothelial lining, are similar in structure and function to other arteries in the body.

### Myocardial Microvessels

Capillaries in the heart possess continuous endothelium. The capillary endothelium is in intimate contact with the cardiomyocytes. The number of myocardial ECs outnumbers cardiomyocytes by a ratio of 3:1 (reviewed elsewhere<sup>59</sup>). The distance between the capillary EC and the nearest cardiomyocyte is only 1  $\mu\text{m}$ . This architecture provides for optimal diffusion of oxygen and nutrients between blood and underlying muscle. Like the endocardium, myocardial capillary

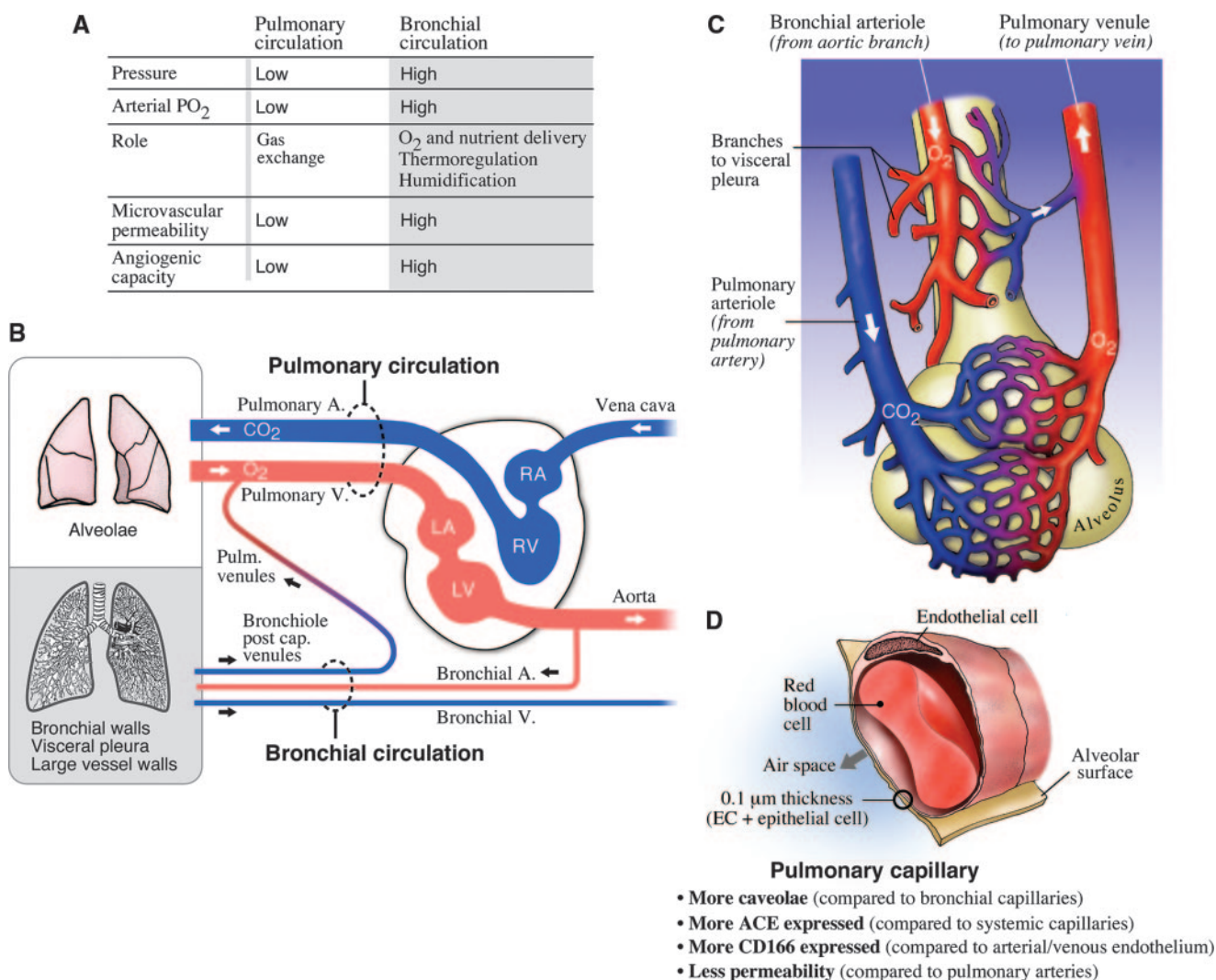
endothelium is intimately involved in reciprocal signaling with cardiomyocytes. Several endothelial-cardiomyocyte interactions are important during development. Endothelial-specific deletion of platelet-derived growth factor (PDGF)-B leads to myocardial abnormalities.<sup>60</sup> Cardiomyocyte-specific knockout of VEGF-A results in a thinned ventricular wall.<sup>61</sup> Cardiomyocyte-specific signal transducer and activator of transcription-3 (STAT3) deletion was shown to reduce LV capillarization, without altering the density of coronary resistance vessels.<sup>62</sup> There were no differences in hypoxia-inducible factor (HIF)-1 $\alpha$ , VEGF, or fibroblast growth factor (FGF)-2 expression in knockout and wild-type animals, suggesting that other downstream genes in cardiomyocytes (with paracrine function) are involved in mediating capillarization. Endothelial-derived nitric oxide (NO) leads to negative inotropy in the heart.<sup>63</sup> Cardiac microvascular ECs have been shown to promote cardiomyocyte survival.<sup>64</sup> Transgenic mouse studies have revealed the importance of cardiomyocyte-derived signals in mediating vascular bed-specific gene expression in myocardial capillaries.<sup>65</sup> Receptor-like tyrosine phosphatase  $\mu$  is expressed by ECs of heart capillaries (and arteries) but not veins or adult endocardium.<sup>66</sup> The arteriolar portion of the capillary bed demonstrates higher alkaline phosphatase activity, whereas the venular portion contains higher dipeptidylpeptidase activity.<sup>67</sup>

### Heart Endothelium in Disease

There is relatively little information about the role of the endocardium in human disease. Atrial fibrillation is associated with reduced expression of eNOS in the atrium.<sup>46</sup> Moreover, rapid atrial pacing in rats was shown to acutely downregulate the expression of thrombomodulin and tissue factor pathway inhibitor (TFPI) in the endocardium of the atrium but not the ventricle.<sup>68</sup> It is interesting to speculate that these effects are causally linked to the increased risk for thrombosis in atrial fibrillation. The notion that endocardial gene expression is sensitive to changes in hemodynamics, as occurs in chronic arrhythmias, is supported by studies of Kruppel-like factor (KLF)-2, endothelin (ET)-1, and eNOS in chicken hearts in which the venous return has been experimentally altered.<sup>69</sup>

It is now widely accepted that valvular degeneration represents a chronic inflammatory process. Valve pathology may be associated with local expression of vascular cell adhesion molecule (VCAM)-1 and E-selectin.<sup>70,71</sup> There is also evidence that aortic valve stenosis is associated with angiogenic activation of valvular ECs. In one study, normal valves were avascular, whereas stenotic aortic valves were shown to contain neovessels.<sup>72</sup> ECs lining these neovessels were consistently positive for platelet/EC adhesion molecule (PECAM)-1 (also known as CD31), but only a portion were positive for vWF.<sup>72</sup>

Coronary artery disease is a leading cause of mortality in the Western world. The nonrandom geometrically defined distribution of atherosclerotic lesions may be explained by regional differences in hemodynamics. Previous studies in animals have demonstrated that regions at high risk for atherosclerosis are exposed to disturbed flow and that ECs at these sites are primed for activation.<sup>4</sup> It is possible that



**Figure 3.** ECs in the lung. A, The lung has a dual circulation: the low-pressure, high-volume pulmonary vasculature, which is involved in gas exchange; and the high-pressure, low-volume bronchial circulation, which delivers oxygen to the bronchial tree. B, Schematic of the pulmonary and bronchial circulation emphasizing differences in oxygen content between veins and arteries. Target tissues are shown on the left. Intrapulmonary bronchial capillaries drain into the pulmonary vein, whereas those in the hilar region drain into true bronchial veins. C, Architectural arrangement of pulmonary and bronchial blood vessels relative to the bronchial tree. D, Unique properties of pulmonary capillary ECs. A indicates artery; cap, capillary; LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle; V, vein; ACE, angiotensin I-converting enzyme.

systemic changes in signal input associated with coronary risk factors (eg, smoke toxins, hypertension, high glucose, or hyperlipidemia) result in preferential activation of primed endothelium in regions of disturbed flow, leading to focal inflammation, monocyte extravasation and the development of fatty streaks (reviewed elsewhere<sup>73</sup>).

Many cardiac risk factors and diseases are associated with abnormalities in the microcirculation. Arteriolar dysfunction and/or narrowing has been described in patients with obesity, hypercholesterolemia, diabetes, hypertension, hypertrophic and dilated cardiomyopathy, and collagen vascular disease (reviewed elsewhere<sup>74</sup>). In rats, diabetes and hypertension are associated with functional and structural changes in myocardial capillaries, including a reduction in NO synthase.<sup>75</sup> In addition to hypertension and diabetes, changes in myocardial capillary endothelial phenotype have been described in hyperlipidemia, ischemia/reperfusion, and dilated idiopathic or

ischemic cardiomyopathy (reviewed elsewhere<sup>42</sup>). Moreover, left ventricular hypertrophy is associated with reduced capillary density.<sup>76</sup> However, the extent to which these various changes contribute to the underlying disease pathophysiology is unclear.

## Lung

The lung has a dual circulation: the low-pressure, high-volume pulmonary vasculature, which is involved in gas exchange; and the high-pressure, low-volume bronchial circulation, which delivers oxygen to the bronchial tree (Figure 3). In addition to taking up oxygen and releasing carbon dioxide, the lung plays an important role in host defense.

## Pulmonary Vessels

A unique feature of the pulmonary circulation is that it undergoes radical changes in the postnatal period. During

embryogenesis, the pulmonary vascular bed receives less than 10% of the cardiac output, whereas, after birth, the pulmonary artery is perfused with the entire cardiac output. Thus, the pulmonary circulation undergoes a sudden transition from low flow/high pressure/high resistance to high flow/low pressure/low resistance. Moreover, pulmonary capillary ECs, which were bathed during embryogenesis with maternally oxygenated blood, are now exposed to the highest oxygen environment of all vascular beds in the body. These sudden changes in the biomechanical and biochemical milieu undoubtedly elicit adaptive phenotypic changes within the endothelium.

During development, airway and pulmonary vascular development are closely interactive processes, thus ensuring close apposition of capillaries with terminal gas-exchanging units of the lung, the alveoli. Although the pulmonary arteries and pulmonary veins branch to the same extent, only the arteries follow the airway architecture. With some exceptions,<sup>77,78</sup> previous studies support a model of pulmonary vascular development in which the pulmonary arteries and veins arise from angiogenesis, with distal vascular development depending on vasculogenesis (reviewed elsewhere<sup>79</sup>).

Pulmonary capillaries are arranged as a densely interconnected net through which blood seeps as a sheet or film (in this analogy, the alveoli represent the holes in the net) (reviewed elsewhere<sup>41</sup>). Gas exchange takes place at the alveolo-capillary membrane (also termed the air-blood barrier), which consists of a layer of epithelium and endothelium separated by a thin basement membrane. The thickness of the gas diffusion surface (alveolar epithelium+interstitium+capillary endothelium) in humans is a mere 0.1  $\mu\text{m}$ . The pulmonary capillary endothelium contains large numbers of caveolae. In the intact lung, microvascular ECs uniquely bind *Griffonia simplicifolia*, a lectin that specifically interacts with  $\alpha$ -galactose.<sup>80</sup> Angiotensin I-converting enzyme is expressed in 100% of alveolar capillary ECs compared with 10% of systemic capillary ECs (including those of the bronchial circulation).<sup>81</sup> Activated leukocyte cell adhesion molecule (ALCAM, also known as CD166) is expressed in rat lung capillary endothelium, but not in larger pulmonary vessels.<sup>80</sup> Alveolar capillary ECs express PECAM-1/CD31 and CD34, but not vWF.<sup>82</sup> A recent study suggests that the clotting factor, factor VIII, may be expressed specifically by lung microvascular ECs.<sup>83</sup> In vivo mapping of the EC surface proteome identified more than 450 nonredundant proteins in rat lung.<sup>84</sup> Subtractive proteomic analysis of plasma membranes yielded a subset of EC proteins that were specifically expressed in lung endothelium, including aminopeptidase P and OX-45.<sup>85</sup> Most of these membrane proteins were shown to be expressed on capillary endothelium.<sup>86</sup>

Owing to the structural characteristics of the pulmonary circulation and mechanical properties of neutrophils, the transit time of neutrophils is prolonged in lung capillaries, compared with other vascular beds (reviewed elsewhere<sup>87</sup>). During inflammation, neutrophils become sequestered in the capillary bed of the lung, a process that is mediated by changes in neutrophil deformability, rather than selectin-induced rolling.<sup>87</sup> Prolonged firm adhesion and subsequent transmigration appears to be dependent, at least in part, on

CD11/CD18–intercellular adhesion molecule (ICAM)-1 interactions.

Infusion of thapsigargin, a plant alkaloid that activates store-operated calcium entry channels, into isolated perfused rat lungs resulted in gap formation and increased permeability in intermediate-to-large arteries and veins, but not capillaries.<sup>88</sup> Compared with rat pulmonary arterial ECs, cultured rat pulmonary microvascular ECs demonstrate reduced basal and inflammatory-mediated permeability to solutes.<sup>89</sup> These differences in permeability have been correlated with differences in hydraulic conductivity.<sup>90</sup> The retention of site-specific differences in permeability in multiply passaged ECs suggests that these properties are epigenetically programmed (reviewed elsewhere<sup>80</sup>). Consistent with this hypothesis, pulmonary micro- and macrovascular ECs demonstrate significant cell type-specific differences in DNA methylation pattern.<sup>80</sup> It is tempting to speculate that these differences reflect the distinct developmental origin of pulmonary artery ECs (angiogenesis) and pulmonary microvascular ECs (vasculogenesis).

### Bronchial Vessels

The bronchial circulation originates from the aorta or intercostal arteries. In contrast to the pulmonary circulation, little is known about the developmental origins of these blood vessels. The bronchial arteries are classified as intrapulmonary or extrapulmonary. The intrapulmonary bronchial arteries perfuse the airways from the level of the carina to the terminal bronchioles, pulmonary pleura, and the walls of the pulmonary artery and vein (ie, vasa vasorum). The extrapulmonary branches feed the esophagus, lobar bronchus, and hilar lymph node. Intrapulmonary capillaries drain into the pulmonary vein, whereas those in the hilar region drain into true bronchial veins. (It should be noted that mice do not have a functional bronchial circulation beyond the mainstem bronchi.<sup>91</sup>) In addition to delivering nutrients to these various structures, the bronchial circulation plays an active role in thermoregulation and humidification of ambient air, as well as facilitating the immune response to foreign material in the airway (by way of plasma exudation). In keeping with these latter functions, ECs in the bronchial vasculature are more leaky, are more responsive to inflammation, and have a far greater capacity for angiogenesis compared with ECs from the pulmonary vasculature (reviewed elsewhere<sup>79,91</sup>). Moreover, the abluminal interstitium is much thicker in the bronchial microcirculation compared with the pulmonary capillaries and contains many more cell types, including macrophages and fibroblasts.

There is less leukocyte margination in bronchial compared with pulmonary capillaries. This may be explained by the wide diameter of bronchial capillaries and/or increased blood pressure in the bronchial circulation (reviewed elsewhere<sup>92</sup>). As with most other systemic vascular beds, leukocyte trafficking in the bronchial circulation takes place in the post-capillary venules. However, in contrast to many vascular beds, the bronchial circulation has been shown to constitutively express E-selectin, suggesting that it is in a state of chronic activation, perhaps in response to inhaled antigens.<sup>93</sup>



Similar to the pulmonary circulation, ECs in the bronchial vasculature display site-specific properties. For example, bronchial microvascular ECs demonstrate increased permeability both at baseline and in response to edematogenic substances, including bradykinin and thrombin, compared with ECs in the bronchial artery.<sup>94</sup> Interestingly, airway venules demonstrate circumferential heterogeneity in inflammatory states. For example, ECs nearest the airway epithelium are phenotypically distinct from those that are on the opposite surface of the same vessel in mice infected with *Mycoplasma pulmonis*.<sup>95</sup>

### Lung Endothelium in Disease

Pathophysiological processes that interfere with simple diffusion in pulmonary capillaries may impair gas exchange and lead to hypoxemia. The endothelium plays an important role in mediating protein-rich pulmonary edema. For example, in sepsis-induced acute lung injury, the pulmonary endothelium demonstrates increased permeability and promotes leukocyte transmigration.<sup>96</sup> The endothelium has also been implicated in transfusion-related acute lung injury (reviewed elsewhere<sup>97</sup>) and ventilator-induced lung injury (reviewed elsewhere<sup>98,99</sup>).

Pulmonary hypertension is associated with hypertrophy and hyperplasia of smooth muscle cells in small precapillary pulmonary arteries. Recent evidence suggests that early pulmonary hypertension is associated with increased EC apoptosis, whereas more advanced pulmonary hypertension is characterized by reduced endothelial apoptosis and formation of plexiform lesions<sup>100</sup> (and reviewed elsewhere<sup>101</sup>). These changes in EC phenotype have been linked to abnormalities in bone morphogenetic protein and VEGF signaling (reviewed elsewhere<sup>101,102</sup>). In one study, medium conditioned with cultures of pulmonary artery ECs from patients with pulmonary hypertension induced a higher rate of smooth muscle cell proliferation compared with ECs from controls.<sup>103</sup> It is unclear whether these mitotically stable differences reflected different genetic background of patients versus controls, or whether they arose from disease-associated epigenetic changes in the endothelium. Patients with emphysema develop destruction of the alveolar septum. There is increasing evidence for a role of VEGF signaling in this process (reviewed elsewhere<sup>102</sup>).

Abnormalities in the bronchial circulation have been implicated in a number of disease states. For example, vascular engorgement and neoangiogenesis in airways may contribute to airflow obstruction in patients with asthma (reviewed elsewhere<sup>104,105</sup>). The bronchial circulation is responsible for more than 90% of all cases of hemoptysis. The disproportionate involvement of the bronchial circulation in hemoptysis may reflect the higher pressure in this vasculature and its greater capacity for neo-angiogenesis and inflammation. Many pulmonary diseases in which blood flow through the pulmonary circulation is compromised (eg, pulmonary hypertension, pulmonary artery thrombosis) are associated with remodeling of the bronchial arteries, including proliferation and enlargement.

Some diseases, such as hereditary hemorrhagic telangiectasia, may affect both pulmonary and bronchial circulations.

Hemorrhagic telangiectasia is associated with the development of arteriovenous malformations in selected organs, including the lung. Hemorrhagic telangiectasia is caused by mutations in endoglin or Alk-1 (reviewed elsewhere<sup>106</sup>). Endoglin is an ancillary receptor (or coreceptor) for transforming growth factor- $\beta_1$  and - $\beta_3$ , and is expressed predominantly by ECs. Alk-1 is a type I serine/threonine receptor kinase of the transforming growth factor- $\beta$  superfamily, whose expression is restricted to endothelium, particularly on the arterial side of the circulation.

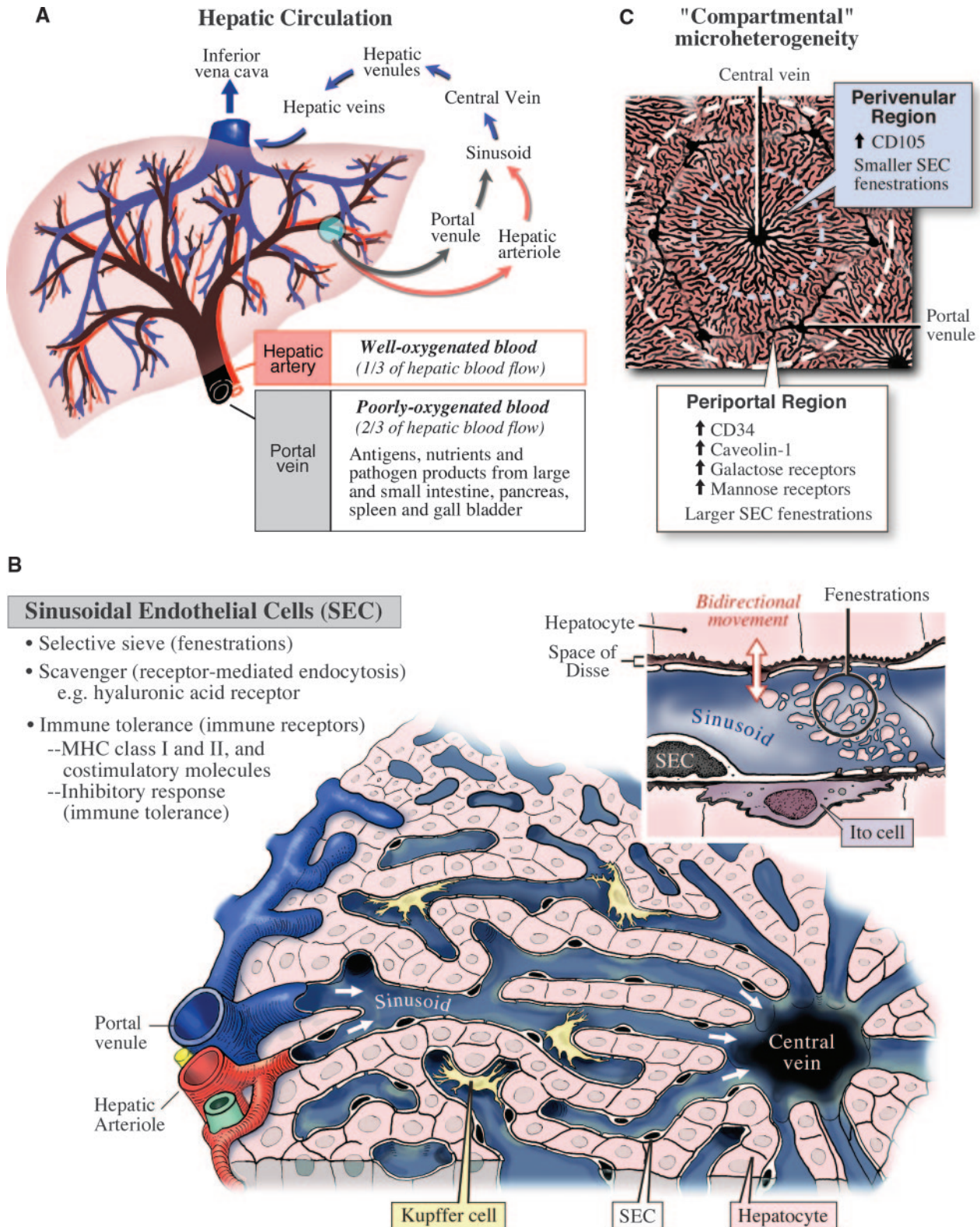
### Liver

The liver receives 15% to 20% of the cardiac output. Like the lung, the liver has a dual blood supply. The hepatic artery, which provides approximately one-third of the blood supply, delivers well oxygenated blood to the liver (Figure 4). The portal vein, representing two-thirds of the blood supply, delivers poorly oxygenated, nutrient-rich blood from several abdominal structures, including the intestine. In contrast to the usual arrangement of veins in the body, the portal vein branches into venules. The hepatic artery and portal vein both drain into the hepatic sinusoids, which represent the capillary network in the liver (occasional direct connections are seen between terminal arterioles and venules before connecting to the sinusoid) (reviewed elsewhere<sup>107</sup>). After circulating in the anastomosing sinuses, blood then empties into terminal hepatic venules, the hepatic vein, and ultimately the inferior vena cava and right atrium.

The endothelium displays a spectrum of phenotypes across the liver vasculature. As the portal vein gives rise to terminal portal venules, the ECs become spindle shaped, nonfenestrated, and possess short microvilli (reviewed elsewhere<sup>108</sup>). At the transition point between the terminal portal venule and hepatic sinusoid, the ECs are smooth and large and contain many actin fibers. Together with underlying Ito cells (the liver equivalents of pericytes), these ECs function as an inlet sphincter that can control sinusoidal blood flow. Hepatic arteries also open directly into the sinusoids. The arterial blood pressure in the terminal hepatic artery is estimated to be 20- to 40-fold higher than that of the sinusoids.<sup>108</sup> The pressure drop between hepatic arteriole and sinusoid is mediated, in part, by a precapillary sphincter, which consists of tall ECs and smooth muscle cells at the junction between the hepatic artery and sinusoid. In addition, it has been hypothesized that large-diameter fenestrae (which are more numerous in the periportal region of the sinusoid) serve to rapidly transport plasma into the space of Disse, thus offloading or cushioning the effects of the arterial jet stream.<sup>108</sup> There is also evidence that sinusoidal lining cells, including ECs and Kupffer cells (discussed below), are capable of contracting and/or swelling as a mechanism of regulating blood flow through the sinuses (reviewed elsewhere<sup>107</sup>).

### Hepatic Sinusoids

Hepatic sinusoidal ECs, which comprise 50% of the non-parenchymal cells of the liver, are discontinuous. They possess large (100 to 200 nm) membrane-bound, nondiaphragmed round cytoplasmic holes or fenestrae (occupying 6% to 8% of the endothelial surface<sup>109</sup>). The fenestrae are





arranged in sieve plates, which are approximately 0.1  $\mu\text{m}$  in diameter and comprise 20 to 50 aggregated pores. Sinusoidal ECs also display gaps and lack an organized basement membrane.

The sinusoidal endothelium functions as a selective sieve. The fenestrae act as a dynamic filter for fluids, solutes, and particles, allowing for passage of small particles (up to medium-sized chylomicrons) from blood to hepatocytes via the space of Disse (transport across the sinusoidal endothelium also occurs via transcytosis, as discussed below). Although the fenestrae are not large enough to accommodate leukocyte transmigration, they do allow cytoplasmic extensions from such cells to penetrate and “touch” underlying matrix, stellate cells, and hepatocytes. Blood flow velocity in the sinusoids has been estimated to be 400 to 450 m/sec, compared with 500 to 1000 m/sec in “true capillaries” (reviewed elsewhere<sup>108</sup>). The relatively low flow would be predicted to prolong interactions between blood and sinusoidal ECs and thus promote filtration. Sieve function may also be enhanced by the action of red blood cells (“forced sieving”) and white blood cells (“endothelial massage”) (reviewed elsewhere<sup>109</sup>). Sieving plays a particularly important role in lipoprotein metabolism. Fenestrae allow for efficient transfer of lipoproteins and small chylomicron remnants between blood and the space of Disse, where they are taken up by hepatocytes via receptor-mediated endocytosis. Loss of sinusoidal EC fenestrations was shown to impair passage of lipoprotein remnants out of the sinusoids and into the space of Disse.<sup>110</sup>

Sinusoidal ECs also function as scavengers, eliminating soluble waste macromolecules from portal venous blood. Indeed, sinusoidal EC have many endocytotic vesicles and lysosome-like vacuoles, consistent with a well-developed endocytotic activity.<sup>109</sup> Among the macromolecules that are cleared are hyaluronan (and other circulating extracellular matrix compounds), acetylated low-density lipoprotein (LDL), denatured albumin, advanced glycation end products, ovalbumin, and other modified or denatured macromolecules. This process involves receptor-mediated endocytosis. Some of these receptors have been identified, including the mannose receptor, scavenger receptor, and the hyaluronan receptors HARE (Hyaluronan Receptor for Endocytosis) and stabilin-2.<sup>111,112</sup>

A unique feature of the liver sinusoid is the presence of monocyte-derived resident macrophages (termed Kupffer cells) on the luminal side of the endothelium. Kupffer cells account for 15% to 20% of the nonparenchymal cells of the liver and 50% of all tissue macrophages in the body (reviewed elsewhere<sup>113</sup>). The sinusoidal EC and Kupffer cell play complementary roles in scavenging function, with the sinusoidal EC removing soluble material via receptor-mediated endocytosis and the Kupffer cell engulfing particulate matter through phagocytosis. Kupffer cells are located primarily in the periportal sinusoids, where they are poised to filter incoming blood from the portal vein. As alluded to above, their proximity to the openings between portal vein/hepatic arteriole and the sinusoids promotes intermittent blockage of flow through the sinusoids, thus increasing the

time of contact between blood-borne cells/soluble substances and Kupffer cells/sinusoidal ECs.<sup>114</sup>

Another important function of liver sinusoidal ECs relates to immunity. Sinusoidal ECs take up antigen via the scavenger receptor and mannose receptor. Like other ECs, sinusoidal ECs present antigen to CD4<sup>+</sup> T cells via major histocompatibility complex (MHC) class II molecules (reviewed elsewhere<sup>115</sup>). In addition, liver sinusoidal ECs constitutively express MHC class I molecules, together with costimulatory molecules CD40, CD80, and CD86. Thus, they have the capacity to present exogenous antigen to CD8<sup>+</sup> T cells, a process that is otherwise restricted to professional antigen-presenting cells (APCs) (reviewed elsewhere<sup>116</sup>). A critical difference between the liver sinusoidal EC and other APCs is that sinusoidal endothelial MHC class I-mediated cross-presentation of antigens to CD8<sup>+</sup> T cells results in antigen-specific induction of CD8<sup>+</sup> T cell tolerance, rather than enhanced immunity.<sup>117,118</sup> Immune tolerance prevents response to innocuous oral antigens and bacterial components from the gastrointestinal tract, as well as neoantigens expressed by neighboring hepatocytes.<sup>115</sup>

Repeated stimulation of sinusoidal ECs by gut-derived bacterial degradation products may underlie refractoriness to lipopolysaccharide signaling.<sup>119</sup> Lipopolysaccharide tolerance is mediated—at least in part—by the paracrine action of Kupffer cell–derived interleukin-10 and tumor necrosis factor- $\alpha$  on sinusoidal ECs (reviewed elsewhere<sup>116</sup>). In this way, sinusoidal ECs can clear lipopolysaccharide without inducing a local inflammatory reaction.

Hepatic sinusoidal ECs play a key role in mediating organogenesis and liver regeneration (reviewed elsewhere<sup>120</sup>). Sinusoidal ECs produce hepatocyte growth factor, which acts in a paracrine manner to induce hepatocyte proliferation. Hepatocytes in turn express VEGF, which binds to VEGF receptor-2 (Flk-1/KDR), leading to EC proliferation. VEGF has also been shown to induce hepatocyte proliferation through VEGF receptor-1 (Flt-1)—dependent sinusoidal EC-derived paracrine mediators, including hepatocyte growth factor and interleukin-6.<sup>121</sup>

Next to the lung, the liver is the site of greatest leukocyte margination in the body. Although postcapillary venules in the liver are capable of mediating leukocyte adhesion and transmigration, the sinusoids support the majority of leukocyte trafficking. As is the case with lung capillaries, leukocyte accumulation and adhesion in the liver sinusoids is not preceded by rolling.<sup>122</sup> Transient leukocyte plugging occurs more frequently in the periportal sinusoids, which are narrower and more tortuous compared with their perivenular counterparts. Liver sinusoidal ECs do not express selectins, but the density of cell surface ICAM-1 is comparable to that of postcapillary (central) venules.<sup>123</sup> Knock out of ICAM-1, but not P/E-selectin, resulted in reduced leukocyte adhesion to sinusoidal endothelium.<sup>122</sup> In addition, activated Kupffer cells have been shown to promote leukocyte recruitment in liver sinusoids, either by altering the shear forces within the microvasculature or by inducing the expression of endothelial and/or leukocyte adhesion molecules.<sup>124</sup> Unlike other vascular beds, PECAM-1/CD31 is not necessary for leukocyte transmigration in the liver.<sup>125</sup> Rather, recent evidence impli-

cates a role for the junctional adhesion molecule-1 in this process.<sup>126</sup> Together, these data suggest that the mechanisms of leukocyte trafficking in the liver sinusoids differ in fundamental from those of other vascular beds.

Sinusoidal ECs also contribute to vasomotor tone. Sinusoidal ECs express the ET-1 receptor ETB. Binding of ET-1 to sinusoidal EC ETB results in NO release and stellate cell-dependent vasodilation at the sinusoidal level.<sup>127,128</sup>

### Compartmentalization of Endothelial Cell Phenotypes in the Liver

The endothelial lining of liver sinusoids demonstrates microheterogeneity. For example, the diameter of fenestrae decreases, but their frequency increases (with net increase in porosity), in sinusoidal ECs from periportal to centrilobular regions.<sup>129</sup> In rats, caveolin-1 expression is increased in periportal sinusoidal ECs.<sup>130</sup> In human liver, CD34 is expressed predominantly in the periportal region, whereas CD105 (also known as endoglin) is present on sinusoidal ECs in the direct vicinity of portal veins.<sup>82,131</sup> In response to ischemia/reperfusion, junctional adhesion molecule-1 is up-regulated in sinusoidal ECs in the perivenular sinusoids (as well as the postcapillary venules).<sup>126</sup> The pattern of lectin-binding sites in sinusoidal ECs differs from periportal to perivenous zones.<sup>132</sup> Sinusoidal ECs in the periportal region express more galactose and mannose receptors and are more efficient in removing apoptotic peripheral lymphocytes.<sup>132</sup> As discussed above, Kupffer cells are more numerous in the periportal sinusoids. In addition, Kupffer cells display different structural and functional properties in periportal versus perivenular regions (reviewed elsewhere<sup>113</sup>). Interestingly, hepatocytes also demonstrate heterogeneity in gene expression depending on their location relative to the portal and central vein (so-called zonal heterogeneity). It has been hypothesized that these phenotypic differences are driven, in part, by signals derived from ECs lining the central vein.<sup>133</sup>

### Liver Endothelium in Disease

In liver fibrosis, the sinusoids may undergo a process termed capillarization, characterized by progressive loss of fenestrae and the formation of a continuous basement membrane; a similar, though less-pronounced process (referred to as pseudocapillarization) occurs with aging.<sup>134–136</sup> These changes probably impair oxygen diffusion and transport of solutes and fluids across the sinusoid. Indeed it has been suggested that aging-related pseudocapillarization results in decreased chylomicron clearance and an increased risk for atherosclerosis.<sup>137,138</sup>

Sinusoidal obstructive syndrome, also known as hepatic venoocclusive disease, occurs most commonly in response to conditioning regimens (chemotherapy with or without irradiation) used in bone marrow transplantation. The first lesions appear in the sinusoids. Experimental models suggest that toxins result in swelling and rounding up of sinusoidal ECs, followed by entry of red blood cells into the space of Disse.<sup>139</sup> In the space of Disse, blood flow essentially dissects the sinusoidal endothelial lining away from underlying parenchymal cells, which results in embolization of sinusoidal ECs and occlusion of the downstream vessel. In an experimental

model of sinusoidal obstructive syndrome, these changes were prevented by administration of glutathione.<sup>139</sup>

Liver injury may lead to underproduction of NO in liver sinusoids, which in turn may contribute to portal hypertension.<sup>140,141</sup> Recent studies point to an important role for G protein-coupled receptor kinase-2-mediated inhibition of Akt in reducing eNOS activity in injured liver sinusoidal ECs.<sup>142</sup> Cirrhosis is associated with increased expression of PECAM-1/CD31 (and/or redistribution to cell surface),<sup>143</sup> as well as new expression of cyclooxygenase-2.<sup>144</sup> Consistent with the role of liver sinusoidal ECs in immune tolerance, portosystemic shunting leads to loss of oral tolerance and increased production of antibodies against intestinal bacterial antigens.<sup>145,146</sup>

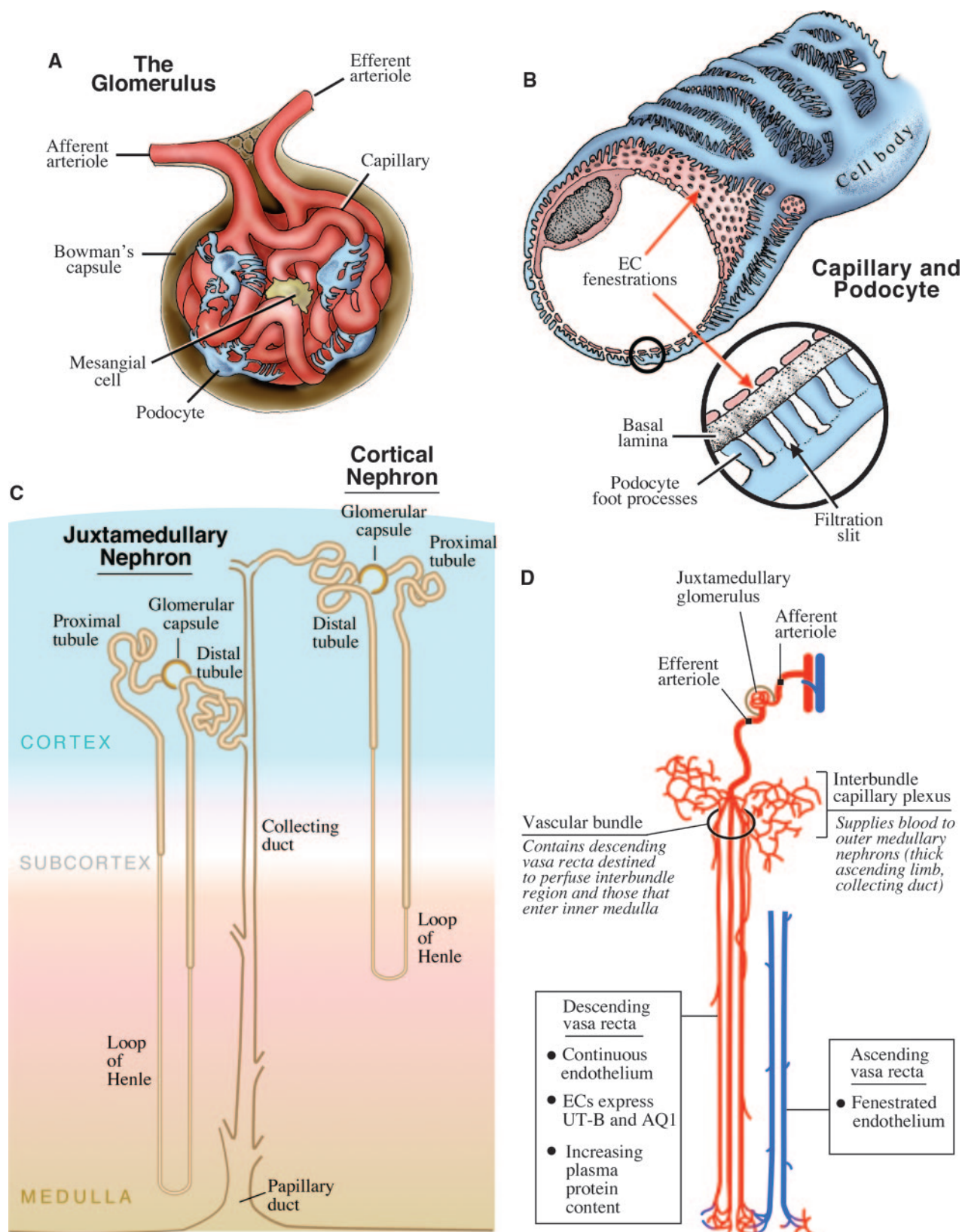
Liver fenestrae are capable of contracting and dilating, a process that appears to be mediated by a calcium/calmodulin/actomyosin-dependent mechanism (reviewed elsewhere<sup>109</sup>). Certain drugs (eg, acetylcholine and ethanol) cause an increase in pore diameter, raising the possibility of therapeutically modulating pore-dependent properties such as cholesterol metabolism (reviewed elsewhere<sup>109</sup>). Drugs such as nicotine and epinephrine decrease endothelial porosity and may contribute to drug-related atherogenesis.<sup>138</sup> As noted above, liver disease (eg, induced by hepatotoxins such as chronic alcohol use) is associated with defenestration, which in turn results in hyperlipidemia.<sup>147,148</sup>

### Kidney

The kidney receives approximately 20% of the cardiac output. Blood enters via the renal artery which then branches sequentially into interlobar, arcuate, and interlobular arteries. Afferent arterioles from the interlobular artery enter the capillary tufts of the glomeruli and exit as efferent arterioles (Figure 5). Efferent arterioles then give rise to the peritubular plexus of capillaries or the vasa recta as described below.

### Glomerular Capillaries

The glomerular circulation functions as a size- and charge-selective filter. Although highly permeable to water and small solutes, the glomerulus is relatively impermeable to macromolecules. The filtration barrier consists of three layers or components: endothelium with glycocalyx, glomerular basement membrane, and podocytes (reviewed elsewhere<sup>149</sup>). Glomerular ECs form the initial barrier to filtration. These cells possess fenestrae that are 60 to 80 nm in diameter,<sup>150</sup> and cover 20% of the endothelial surface.<sup>151</sup> As with nonglomerular continuous fenestrated endothelium, these fenestrae appear to possess a diaphragm.<sup>150</sup> However, unlike other fenestrated endothelium (including that of neighboring peritubular capillaries), the bridging diaphragms of glomerular ECs lack the protein PV-1.<sup>152</sup> Glomerular ECs actively synthesize glycocalyx and basement membrane. The glycocalyx, a 60- to 300-nm thick surface layer of membrane-associated proteoglycans, glycosaminoglycans, glycolipids and associated plasma proteins, provides a filtration barrier with charge selectivity.<sup>153</sup> Under in vivo conditions, human glomerular ECs are uniformly positive for PECAM-1/CD31 and CD34 but not vWF.<sup>82</sup> ECs in the glomerulus contribute to the control of vasomotor tone, by releasing factors such as NO, prostaglandins, and ET-1.



**Figure 5.** ECs in the kidney. A and B, The glomerular circulation functions as a size- and charge-selective filter, with fenestrated ECs forming the initial barrier to filtration. Efferent arterioles from the outer- and mid-cortex form a peritubular plexus of capillaries (not shown). These microvessels surround the proximal and distal convoluted tubules and serve to accommodate reabsorption of glomerular filtrate. C and D, Efferent arterioles from the juxtamedullary glomeruli terminate in vasa recta, which enter the medulla as descending arterioles (DVR) and exit the medulla as ascending veins (AVR). The descending and ascending limbs of the vasa recta are connected by a sparse capillary plexus. The microcirculation of the vasa recta provides the medulla with countercurrent exchange, which is important for preserving medullary hypertonicity. Shown are phenotypic differences between ECs in the DVR and AVR.



There is increasing evidence supporting an important role for crosstalk between podocytes and ECs not only in mediating barrier properties but also in promoting “cell health.” Most notably, podocyte-specific deletion of VEGF resulted in profound phenotypic alterations of glomerular ECs and loss of barrier function.<sup>154</sup> In addition to podocytes and ECs, glomeruli contain mesangial cells. Recent studies suggest that mesangial cells communicate with glomerular ECs and appear to be important for their proliferation, neovascularization, and mechanical stability of the capillary.<sup>155–157</sup>

Efferent arterioles from the outer- and mid-cortex form a peritubular plexus of capillaries. These microvessels surround the proximal and distal convoluted tubules and serve to accommodate reabsorption of glomerular filtrate.

### Vasa Recta

Efferent arterioles from the juxtamedullary glomeruli terminate in vasa recta, which enter the medulla as descending arterioles (descending vasa recta [DVR]) and exit the medulla as ascending veins (ascending vasa recta [AVR]). The descending and ascending limbs of the vasa recta are connected by a sparse capillary plexus. The microcirculation of the vasa recta provides the medulla with countercurrent exchange, which is important for preserving medullary hypertonicity (reviewed elsewhere<sup>158</sup>). For the most part, osmotically active solutes (eg, NaCl and urea) are taken up from the interstitium of the medulla in the DVR and released back into the interstitium in the AVR, effectively “trapping” solute in the interstitium by recycling between ascending and descending capillary, thus avoiding a wash out of hypertonicity.

These functional differences are reflected by site-specific differences in endothelial phenotypes. For example, the endothelium of the DVR is continuous and nonfenestrated. NaCl and other small hydrophilic solutes enter the DVR via diffusive paracellular transport. In contrast, DVR ECs use transcellular pathways to transport urea and water. Urea transport from interstitium to lumen is mediated by the facilitated urea transporter, UT-B. This is the same urea transporter that is expressed in red blood cells but differs from the major urea receptors expressed in epithelial cells (UT-A1–4). The transporter serves to take up and recycle urea that diffuses from the AVR. Genetic deletion of UT-B in mice results in impaired urine concentrating ability,<sup>159</sup> implicating a role for DVR UT-B in preserving countercurrent exchange. In contrast to NaCl and urea, which move from AVR to interstitium to DVR, water moves from DVR to interstitium to AVR. This process is facilitated by the water channel AQP1. It has been hypothesized that the net loss of water from DVR results in reduced blood flow to the inner medulla, thus improving the plasma-interstitial equilibrium and optimizing countercurrent exchange. Mice lacking AQP1 manifest defective urinary concentrating ability.<sup>160</sup>

In contrast to DVR, ECs of the ascending vasa recta are fenestrated (more so in the inner versus outer medulla). Compared with DVR, ECs in the AVR have higher hydraulic conductivity and lower reflection coefficients for small solutes. These properties promote net movement of solutes (eg, NaCl and urea) from blood to interstitium. The excess of water that is transported by AQP1 from DVR into

interstitium is absorbed by the AVR. Because the inner medulla does not contain lymphatics, interstitial albumin is taken up by AVR.

Another important function of the vasa recta is to deliver oxygen and nutrients to medullary tissue. A trade off of countercurrent exchange is that oxygen and nutrients are shunted from DVR to AVR, which results in profound hypoxia in the inner medulla ( $P_{O_2}$  in the medulla is as low as 10 to 25 mm Hg) (reviewed elsewhere<sup>161</sup>). The threat of ischemia is offset by the capacity of the kidney to tightly regulate perfusion to the outer and inner medulla, particularly at the level of the DVR. Indeed, DVR functions both as an exchange vessel (capillary) and a resistance vessel (arteriole).

In addition to serving countercurrent exchange functions, other properties of the vasa rectal ECs are likely to reflect their adaptation to extremes in the medullary environment, including profound hypoxia and hyper-osmolarity (reviewed elsewhere<sup>162</sup>).

### Compartmentalization of Endothelial Cell Phenotypes in the Kidney

As with the liver, the kidney microvasculature displays remarkable compartmentalization. Consider the following path through the renal microcirculation: well-oxygenated blood enters the glomerular capillary via the afferent renal arteriole. In contrast to other vascular beds, the glomerular capillaries serve primarily to filter fluids and solutes, not to exchange oxygen and nutrients. In an unusual arrangement, the glomerular capillaries coalesce into yet another arteriolar system, the efferent arterioles. Because approximately 30% of the blood volume is filtered by the glomerulus, blood entering the efferent arterioles will have markedly increased viscosity. The efferent arteriole then gives rise to the DVR, which are hybrid resistance and exchange vessels. Net exchange of water from DVR into interstitium would be predicted to increase the plasma protein concentration and viscosity of the blood even further. In the hyperosmolar, hypoxic depths of the inner medulla, the DVR branch into yet another capillary network, from which blood ultimately drains back to the cortex via the AVR. At each step along the path, ECs perform different functions to maintain kidney homeostasis. Moreover, they are exposed to vastly different input signals from the extracellular environment.

In keeping with their functional heterogeneity, the different microvascular compartments express distinct molecular markers. For example, eNOS immunoreactivity is greater in ECs of the renal medulla (vasa recta) compared with the cortex (glomeruli and peritubular capillaries).<sup>163</sup> Interestingly, the corticomedullary pattern is reversed in embryonic kidneys, with highest expression in the cortex.<sup>163</sup> Gap junctional proteins also vary across the renal vasculature. For example, connexin 37 and 40 are expressed in the endothelium of afferent, but not efferent, arterioles of the mouse kidney; connexin 43 is expressed in both afferent and efferent arterioles; and none of the above connexins are present in the glomerular capillaries.<sup>164</sup> Claudin-10 and claudin-15 are expressed in the endothelium of the vasa recta, but not afferent or efferent arterioles.<sup>165</sup> As discussed above, the urea transporter UT-B1 and the water channel AQP1, are specifically

expressed in ECs of the DVR.<sup>166</sup> Expression of UT-B ends quite abruptly at the distal end of DVR, where these vessels connect to the capillary plexus before forming the AVR.<sup>167</sup> In addition to AQ1 and UT-B, DVR ECs specifically express transient receptor potential channel-4 and an isoform of the Na/H exchanger regulator factor NHERF-2.<sup>168</sup> The endothelium of the AVR (and the very terminal end of DVR and capillary plexus) possess fenestrae and express PV-1.<sup>167</sup>

### Kidney Endothelium in Disease

In a rat model of renal microvascular endothelial injury, the chemokine interferon-inducible protein-10/CXCL10 was expressed in ECs in the tubulointerstitial area, but not in glomerular ECs, whereas monocyte chemoattractant protein-1/CCL2 was induced in both populations of ECs.<sup>169</sup> The differential expression of chemokines by ECs in different compartments of the kidney correlated with differences in T-cell infiltration. Consistent with its organ predilection, Wegener's granulomatosis is associated with the presence of anti-EC antibodies that specifically target ECs from the nose, kidney, and lung.<sup>170</sup> Hemolytic uremic syndrome is characterized by microangiopathic hemolytic anemia, thrombocytopenia, and acute renal failure. The classic form is caused by Shiga-like, toxin-producing bacterial infection (reviewed elsewhere<sup>171</sup>). Atypical forms of hemolytic uremic syndrome are associated with certain drugs and infectious agents. A cardinal feature of both classic and atypical hemolytic uremic syndrome is endothelial damage. Glycolipid receptors for Shiga toxins are preferentially expressed in ECs of peritubular capillaries.<sup>172</sup> Diabetic nephropathy arises from a complex interaction among metabolic, hemodynamic factors, and growth regulating peptides. In animal models of diabetes and in patients with type 1 diabetes, VEGF expression is increased.<sup>173,174</sup> In diabetic animals, neutralizing antibodies to VEGF improve glomerular volume and urinary albumin excretion.<sup>175</sup> Also in animal models, diabetes is associated with upregulation of fractalkine,<sup>176</sup> calcineurin-A- $\alpha$ ,<sup>177</sup> adrenomedullin, and its receptor RMAP2,<sup>178</sup> in glomerular ECs. In a mouse model of lupus nephritis, the pattern of adhesion molecule expression in glomerular ECs varies according to the severity and diversity of the histopathology.<sup>179</sup>

### Conclusions

Owing to space limitations, the current review has necessarily focused on selected vascular beds. There are, of course, many other vascular beds of interest to the biomedical practitioner. Among the more thoroughly studied are the blood-brain barrier (reviewed elsewhere<sup>180,181</sup>), high endothelial venules (reviewed elsewhere<sup>182</sup>), and lymphatic blood vessels (reviewed elsewhere<sup>183</sup>). Each vascular bed has its own "story to tell." ECs from different organs demonstrate unique structural and functional properties, as well as distinct developmental programs, roles in pathophysiology, and potential for targeted therapy in patients with vascular disease.

Advances in our understanding of EC phenotypes have depended on (and in turn engendered) organ-specific approaches to the endothelium. For example, cardiologists have focused primarily on the coronary arteries; neurologists on the cerebral circulation/blood-brain barrier; hepatologists on

the liver circulation; and dermatologists on the skin microvasculature. However, there also are advantages in approaching the endothelium as an integrated system. Insights gleaned from studies of one vascular bed may be more readily applied to an understanding of the biology, propensity for disease, and/or therapeutic potential of the endothelium in another organ. For example, the study of tumor vasculature has provided enormously valuable insights into the structure and function of postcapillary venules.<sup>32,184</sup> Investigations of the placenta and kidney have yielded important insight into the mechanisms by which VEGF deficiency (eg, as occurs in patients with cancer receiving anti-VEGF therapy) promotes hypertension and proteinuria. Knowledge of the molecular basis of scavenging activity in liver sinusoidal ECs may help to therapeutically modulate particle clearance in other vascular beds. An understanding of the mechanisms underlying transcytosis in non-blood-brain barrier endothelium may provide a foundation for drug delivery in neurological disease. The ECs in the inner medulla of the kidney are exposed to profoundly low levels of oxygen. It is likely that these cells are uniquely adapted to this extreme degree of hypoxia. Perhaps an understanding of these adaptive mechanisms might provide clues about how to render ECs of the lung and other vascular beds less vulnerable to hypoxia in critically ill states.

A final, albeit philosophical, reason for embracing the endothelium as an integrated system relates to the fragmented nature of clinical medicine. With the rapid expansion of knowledge necessitating increasing degrees of specialization, the resulting tendency to compartmentalize the human body (conceptually, and from the standpoint of diagnosis and therapy) has precluded an appreciation for the critical interactions between individual organs. Because the endothelium is spatially distributed throughout the body, because it communicates with each and every tissue, and because it is involved in most disease states, it represents a powerful organizing principle in human health and disease.

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### References

1. Simionescu M, Simionescu N, Palade GE. Segmental differentiations of cell junctions in the vascular endothelium. Arteries and veins. *J Cell Biol.* 1976;68:705–723.
2. Eriksson EE, Karlof E, Lundmark K, Rotzius P, Hedin U, Xie X. Powerful inflammatory properties of large vein endothelium in vivo. *Arterioscler Thromb Vasc Biol.* 2005;25:723–728.
3. Lupu C, Westmuckett AD, Peer G, Ivanciu L, Zhu H, Taylor FB Jr, Lupu F. Tissue factor-dependent coagulation is preferentially up-regulated within arterial branching areas in a baboon model of *Escherichia coli* sepsis. *Am J Pathol.* 2005;167:1161–1172.
4. Hajra L, Evans AI, Chen M, Hyduk SJ, Collins T, Cybulsky MI. The NF-kappa B signal transduction pathway in aortic endothelial cells is primed for activation in regions predisposed to atherosclerotic lesion formation. *Proc Natl Acad Sci U S A.* 2000;97:9052–9057.

5. Passerini AG, Polacek DC, Shi C, Francesco NM, Manduchi E, Grant GR, Pritchard WF, Powell S, Chang GY, Stoeckert CJ Jr, Davies PF. Coexisting proinflammatory and antioxidative endothelial transcription profiles in a disturbed flow region of the adult porcine aorta. *Proc Natl Acad Sci U S A*. 2004;101:2482–2487.
6. Aird WC. Mechanisms of endothelial cell heterogeneity in health and disease. *Circ Res*. 2006;98:159–162.
7. Gale NW, Baluk P, Pan L, Kwan M, Holash J, DeChiara TM, McDonald DM, Yancopoulos GD. Ephrin-B2 selectively marks arterial vessels and neovascularization sites in the adult, with expression in both endothelial and smooth-muscle cells. *Dev Biol*. 2001;230:151–160.
8. Krebs LT, Xue Y, Norton CR, Shutter JR, Maguire M, Sundberg JP, Gallahan D, Closson V, Kitajewski J, Callahan R, Smith GH, Stark KL, Gridley T. Notch signaling is essential for vascular morphogenesis in mice. *Genes Dev*. 2000;14:1343–1352.
9. Seki T, Yun J, Oh SP. Arterial endothelium-specific activin receptor-like kinase 1 expression suggests its role in arterialization and vascular remodeling. *Circ Res*. 2003;93:682–689.
10. Tian H, McKnight SL, Russell DW. Endothelial PAS domain protein 1 (EPAS1), a transcription factor selectively expressed in endothelial cells. *Genes Dev*. 1997;11:72–82.
11. Nakagawa O, Nakagawa M, Richardson JA, Olson EN, Srivastava D. HRT1, HRT2, and HRT3: a new subclass of bHLH transcription factors marking specific cardiac, somitic, and pharyngeal arch segments. *Dev Biol*. 1999;216:72–84.
12. Mukoyama YS, Gerber HP, Ferrara N, Gu C, Anderson DJ. Peripheral nerve-derived VEGF promotes arterial differentiation via neuropilin 1-mediated positive feedback. *Development*. 2005;132:941–952.
13. Shin D, Anderson DJ. Isolation of arterial-specific genes by subtractive hybridization reveals molecular heterogeneity among arterial endothelial cells. *Dev Dyn*. 2005;233:1589–1604.
14. Gerety SS, Wang HU, Chen ZF, Anderson DJ. Symmetrical mutant phenotypes of the receptor EphB4 and its specific transmembrane ligand ephrin-B2 in cardiovascular development. *Mol Cell*. 1999;4:403–414.
15. Yuan L, Moyon D, Pardanaud L, Breant C, Karkkainen MJ, Alitalo K, Eichmann A. Abnormal lymphatic vessel development in neuropilin 2 mutant mice. *Development*. 2002;129:4797–4806.
16. You LR, Lin FJ, Lee CT, DeMayo FJ, Tsai MJ, Tsai SY. Suppression of Notch signalling by the COUP-TFII transcription factor regulates vein identity. *Nature*. 2005;435:98–104.
17. Kang J, Lee I. TuJ1 (class III beta-tubulin) as phenotypic marker of lymphatic and venous valves. *Cardiovasc Pathol*. 2006;15:218–221.
18. Torres-Vazquez J, Kamei M, Weinstein BM. Molecular distinction between arteries and veins. *Cell Tissue Res*. 2003;314:43–59.
19. Othman-Hassan K, Patel K, Papoutsi M, Rodríguez-Niedenfuhr M, Christ B, Wilting J. Arterial identity of endothelial cells is controlled by local cues. *Dev Biol*. 2001;237:398–409.
20. Moyon D, Pardanaud L, Yuan L, Breant C, Eichmann A. Plasticity of endothelial cells during arterial-venous differentiation in the avian embryo. *Development*. 2001;128:3359–3370.
21. Tsukurov OI, Kwolek CJ, L'Italien GJ, Benbrahim A, Milinazzo BB, Conroy NE, Gertler JP, Orkin RW, Abbott WM. The response of adult human saphenous vein endothelial cells to combined pressurized pulsatile flow and cyclic strain, in vitro. *Ann Vasc Surg*. 2000;14:260–267.
22. Dai G, Kaazempur-Mofrad MR, Natarajan S, Zhang Y, Vaughn S, Blackman BR, Kamm RD, Garcia-Cardena G, Gimbrone MA Jr. Distinct endothelial phenotypes evoked by arterial waveforms derived from atherosclerosis-susceptible and -resistant regions of human vasculature. *Proc Natl Acad Sci U S A*. 2004;101:14871–14876.
23. Kwei S, Stavrakis G, Takahas M, Taylor G, Folkman MJ, Gimbrone MA Jr, Garcia-Cardena G. Early adaptive responses of the vascular wall during venous arterialization in mice. *Am J Pathol*. 2004;164:81–89.
24. Golledge J, Turner RJ, Harley SL, Springall DR, Powell JT. Circumferential deformation and shear stress induce differential responses in saphenous vein endothelium exposed to arterial flow. *J Clin Invest*. 1997;99:2719–2726.
25. Gosling M, Golledge J, Turner RJ, Powell JT. Arterial flow conditions downregulate thrombomodulin on saphenous vein endothelium. *Circulation*. 1999;99:1047–1053.
26. Sperry JL, Deming CB, Bian C, Walinsky PL, Kass DA, Kolodgie FD, Virmani R, Kim AY, Rade JJ. Wall tension is a potent negative regulator of in vivo thrombomodulin expression. *Circ Res*. 2003;92:41–47.
27. Jeremy JY, Dashwood MR, Mehta D, Izzat MB, Shukla N, Angelini GD. Nitric oxide, prostacyclin and cyclic nucleotide formation in externally stented porcine vein grafts. *Atherosclerosis*. 1998;141:297–305.
28. Mavromatis K, Fukai T, Tate M, Chesler N, Ku DN, Galis ZS. Early effects of arterial hemodynamic conditions on human saphenous veins perfused ex vivo. *Arterioscler Thromb Vasc Biol*. 2000;20:1889–1895.
29. Rabkin-Aikawa E, Aikawa M, Farber M, Kratz JR, Garcia-Cardena G, Kouchoukos NT, Mitchell MB, Jonas RA, Schoen FJ. Clinical pulmonary autograft valves: pathologic evidence of adaptive remodeling in the aortic site. *J Thorac Cardiovasc Surg*. 2004;128:552–561.
30. Caggiani A, Phillips M, Lametschwandner A, Allegra C. Valves in small veins and venules. *Eur J Vasc Endovasc Surg*. 2006;32:447–452.
31. Dunn RM, Fudem GM, Walton RL, Anderson FA Jr, Malhotra R. Free flap valvular transplantation for refractory venous ulceration. *J Vasc Surg*. 1994;19:525–531.
32. Feng D, Nagy JA, Dvorak HF, Dvorak AM. Ultrastructural studies define soluble macromolecular, particulate, and cellular transendothelial cell pathways in venules, lymphatic vessels, and tumor-associated microvessels in man and animals. *Microsc Res Tech*. 2002;57:289–326.
33. Frenette PS, Johnson RC, Hynes RO, Wagner DD. Platelets roll on stimulated endothelium in vivo: an interaction mediated by endothelial P-selectin. *Proc Natl Acad Sci U S A*. 1995;92:7450–7454.
34. Mori M, Salter JW, Vowinkel T, Kriegelstein CF, Stokes KY, Granger DN. Molecular determinants of the prothrombotic phenotype assumed by inflamed colonic venules. *Am J Physiol Gastrointest Liver Physiol*. 2005;288:G920–G926.
35. Tailor A, Granger DN. Hypercholesterolemia promotes leukocyte-dependent platelet adhesion in murine postcapillary venules. *Microcirculation*. 2004;11:597–603.
36. Cooper D, Russell J, Chitman KD, Williams MC, Wolf RE, Granger DN. Leukocyte dependence of platelet adhesion in postcapillary venules. *Am J Physiol Heart Circ Physiol*. 2004;286:H1895–1900.
37. Russell J, Cooper D, Tailor A, Stokes KY, Granger DN. Low venular shear rates promote leukocyte-dependent recruitment of adherent platelets. *Am J Physiol Gastrointest Liver Physiol*. 2003;284:G123–G129.
38. Wood KC, Heibel RP, Granger DN. Endothelial cell P-selectin mediates a proinflammatory and prothrombotic phenotype in cerebral venules of sickle cell transgenic mice. *Am J Physiol Heart Circ Physiol*. 2004;286:H1608–H1614.
39. von Andrian UH, Mempel TR. Homing and cellular traffic in lymph nodes. *Nat Rev Immunol*. 2003;3:867–878.
40. Miyasaka M, Tanaka T. Lymphocyte trafficking across high endothelial venules: dogmas and enigmas. *Nat Rev Immunol*. 2004;4:360–370.
41. Kilner PJ. Pulmonary resistance in cardiovascular context. *Int J Cardiol*. 2004;97(suppl 1):3–6.
42. Brutsaert DL. Cardiac endothelial-myocardial signaling: its role in cardiac growth, contractile performance, and rhythmicity. *Physiol Rev*. 2003;83:59–115.
43. Brutsaert DL, Fransen P, Andries LJ, De Keulenaer GW, Sys SU. Cardiac endothelium and myocardial function. *Cardiovasc Res*. 1998;38:281–290.
44. Paulus WJ. Endothelial control of vascular and myocardial function in heart failure. *Cardiovasc Drugs Ther*. 1994;8:437–446.
45. Yamamoto K, de Waard V, Fearn C, Loskutoff DJ. Tissue distribution and regulation of murine von Willebrand factor gene expression in vivo. *Blood*. 1998;92:2791–2801.
46. Cai H, Li Z, Goette A, Mera F, Honeycutt C, Feterik K, Wilcox JN, Dudley SC Jr, Harrison DG, Langberg JJ. Downregulation of endothelial nitric oxide synthase expression and nitric oxide production in atrial fibrillation: potential mechanisms for atrial thrombosis and stroke. *Circulation*. 2002;106:2854–2858.
47. Andries LJ, Brutsaert DL, Sys SU. Nonuniformity of endothelial constitutive nitric oxide synthase distribution in cardiac endothelium. *Circ Res*. 1998;82:195–203.
48. Wunsch AM, Little CD, Markwald RR. Cardiac endothelial heterogeneity defines valvular development as demonstrated by the diverse expression of JB3, an antigen of the endocardial cushion tissue. *Dev Biol*. 1994;165:585–601.
49. Romano LA, Runyan RB. Slug is a mediator of epithelial-mesenchymal cell transformation in the developing chicken heart. *Dev Biol*. 1999;212:243–254.
50. Runyan RB, Markwald RR. Invasion of mesenchyme into three-dimensional collagen gels: a regional and temporal analysis of interaction in embryonic heart tissue. *Dev Biol*. 1983;95:108–114.
51. Wang J, Sridurongrit S, Dudas M, Thomas P, Nagy A, Schneider MD, Epstein JA, Kaartinen V. Atrioventricular cushion transformation is



- mediated by ALK2 in the developing mouse heart. *Dev Biol.* 2005;286:299–310.
52. Noseda M, McLean G, Niessen K, Chang L, Pollet I, Montpetit R, Shahidi R, Dorovini-Zis K, Li L, Beckstead B, Durand RE, Hoodless PA, Karsan A. Notch activation results in phenotypic and functional changes consistent with endothelial-to-mesenchymal transformation. *Circ Res.* 2004;94:910–917.
  53. Armstrong EJ, Bischoff J. Heart valve development: endothelial cell signaling and differentiation. *Circ Res.* 2004;95:459–470.
  54. Lambrechts D, Carmeliet P. Sculpting heart valves with NFATc and VEGF. *Cell.* 2004;118:532–534.
  55. Butcher JT, Tressell S, Johnson T, Turner D, Sorescu G, Jo H, Nerem RM. Transcriptional profiles of valvular and vascular endothelial cells reveal phenotypic differences: influence of shear stress. *Arterioscler Thromb Vasc Biol.* 2006;26:69–77.
  56. Simmons CA, Grant GR, Manduchi E, Davies PF. Spatial heterogeneity of endothelial phenotypes correlates with site-specific vulnerability to calcification in normal porcine aortic valves. *Circ Res.* 2005;96:792–799.
  57. Inai T, Mancuso MR, McDonald DM, Kobayashi J, Nakamura K, Shibata Y. Shear stress-induced upregulation of connexin 43 expression in endothelial cells on upstream surfaces of rat cardiac valves. *Histochem Cell Biol.* 2004;122:477–483.
  58. Reese DE, Mikawa T, Bader DM. Development of the coronary vessel system. *Circ Res.* 2002;91:761–768.
  59. Hsieh PC, Davis ME, Lisowski LK, Lee RT. Endothelial-cardiomyocyte interactions in cardiac development and repair. *Annu Rev Physiol.* 2006;68:51–66.
  60. Bjarnegard M, Enge M, Norlin J, Gustafsdottir S, Fredriksson S, Abramsson A, Takemoto M, Gustafsson E, Fassler R, Betsholtz C. Endothelium-specific ablation of PDGFB leads to pericyte loss and glomerular, cardiac and placental abnormalities. *Development.* 2004;131:1847–1857.
  61. Giordano FJ, Gerber HP, Williams SP, VanBruggen N, Bunting S, Ruiz-Lozano P, Gu Y, Nath AK, Huang Y, Hickey R, Dalton N, Peterson KL, Ross J Jr, Chien KR, Ferrara N. A cardiac myocyte vascular endothelial growth factor paracrine pathway is required to maintain cardiac function. *Proc Natl Acad Sci U S A.* 2001;98:5780–5785.
  62. Hilfiker-Kleiner D, Hilfiker A, Fuchs M, Kaminski K, Schaefer A, Schieffer B, Hillmer A, Schmiedl A, Ding Z, Podewski E, Podewski E, Poli V, Schneider MD, Schulz R, Park JK, Wollert KC, Drexler H. Signal transducer and activator of transcription 3 is required for myocardial capillary growth, control of interstitial matrix deposition, and heart protection from ischemic injury. *Circ Res.* 2004;95:187–195.
  63. Barouch LA, Harrison RW, Skaf MW, Rosas GO, Cappola TP, Kobeissi ZA, Hobai IA, Lemmon CA, Burnett AL, O'Rourke B, Rodriguez ER, Huang PL, Lima JA, Berkowitz DE, Hare JM. Nitric oxide regulates the heart by spatial confinement of nitric oxide synthase isoforms. *Nature.* 2002;416:337–339.
  64. Narmoneva DA, Vukmirovic R, Davis ME, Kamm RD, Lee RT. Endothelial cells promote cardiac myocyte survival and spatial reorganization: implications for cardiac regeneration. *Circulation.* 2004;110:962–968.
  65. Aird WC, Edelberg JM, Weiler-Guettler H, Simmons WW, Smith TW, Rosenberg RD. Vascular bed-specific expression of an endothelial cell gene is programmed by the tissue microenvironment. *J Cell Biol.* 1997;138:1117–1124.
  66. Koop EA, Lopes SM, Feiken E, Bluysen HA, van der Valk M, Voest EE, Mummery CL, Moolenaar WH, Gebbink MF. Receptor protein tyrosine phosphatase mu expression as a marker for endothelial cell heterogeneity; analysis of RPTPmu gene expression using LacZ knock-in mice. *Int J Dev Biol.* 2003;47:345–354.
  67. Gao M, Shirato H, Miyasaka K, Koyama T. Effect of irradiation on enzymes of the capillary bed in rat ventricles. *Adv Exp Med Biol.* 2003;530:527–533.
  68. Yamashita T, Sekiguchi A, Iwasaki YK, Sagara K, Hatano S, Iinuma H, Aizawa T, Fu LT. Thrombomodulin and tissue factor pathway inhibitor in endocardium of rapidly paced rat atria. *Circulation.* 2003;108:2450–2452.
  69. Groenendijk BC, Hierck BP, Vrolijk J, Baiker M, Pourquie MJ, Gittenberger-de Groot AC, Poelmann RE. Changes in shear stress-related gene expression after experimentally altered venous return in the chicken embryo. *Circ Res.* 2005;96:1291–1298.
  70. Ghaisas NK, Foley JB, O'Brian DS, Crean P, Kelleher D, Walsh M. Adhesion molecules in nonrheumatic aortic valve disease: endothelial expression, serum levels and effects of valve replacement. *J Am Coll Cardiol.* 2000;36:2257–2262.
  71. Muller AM, Cronen C, Kupferwasser LI, Oelert H, Muller KM, Kirkpatrick CJ. Expression of endothelial cell adhesion molecules on heart valves: up-regulation in degeneration as well as acute endocarditis. *J Pathol.* 2000;191:54–60.
  72. Chalajour F, Treede H, Ebrahimnejad A, Lauke H, Reichenspurner H, Ergun S. Angiogenic activation of valvular endothelial cells in aortic valve stenosis. *Exp Cell Res.* 2004;298:455–464.
  73. Aird WC. Endothelial cell heterogeneity and atherosclerosis. *Curr Atheroscler Rep.* 2006;8:69–75.
  74. Abarquez RF Jr, Cinco JE. Microcirculation: target therapy in cardiovascular diseases—a clinical perspective. *Clin Hemorheol Microcirc.* 2003;29:157–165.
  75. Okruhlicova L, Tribulova N, Weismann P, Sotnikova R. Ultrastructure and histochemistry of rat myocardial capillary endothelial cells in response to diabetes and hypertension. *Cell Res.* 2005;15:532–538.
  76. Levy BI, Duriez M, Samuel JL. Coronary microvasculature alteration in hypertensive rats. Effect of treatment with a diuretic and an ACE inhibitor. *Am J Hypertens.* 2001;14:7–13.
  77. Schachtner SK, Wang Y, Scott Baldwin H. Qualitative and quantitative analysis of embryonic pulmonary vessel formation. *Am J Respir Cell Mol Biol.* 2000;22:157–165.
  78. Parera MC, van Dooren M, van Kempen M, de Krijger R, Grosveld F, Tibboel D, Rottier R. Distal angiogenesis: a new concept for lung vascular morphogenesis. *Am J Physiol Lung Cell Mol Physiol.* 2005;288:L141–L149.
  79. Stenmark KR, Abman SH. Lung vascular development: implications for the pathogenesis of bronchopulmonary dysplasia. *Annu Rev Physiol.* 2005;67:623–661.
  80. Gebb S, Stevens T. On lung endothelial cell heterogeneity. *Microvasc Res.* 2004;68:1–12.
  81. Balyasnikova IV, Metzger R, Visintine DJ, Dimasius V, Sun ZL, Berestetskaya YV, McDonald TD, Curiel DT, Minshall RD, Danilov SM. Selective rat lung endothelial targeting with a new set of monoclonal antibodies to angiotensin I-converting enzyme. *Pulm Pharmacol Ther.* 2005;18:251–267.
  82. Pusztaszeri MP, Seelentag W, Bosman FT. Immunohistochemical expression of endothelial markers CD31, CD34, von Willebrand factor, and Fli-1 in normal human tissues. *J Histochem Cytochem.* 2006;54:385–395.
  83. Jacquemin M, Neyrinck A, Hermanns MI, Lavend'homme R, Rega F, Saint-Remy JM, Peerlinck K, Van Raemdonck D, Kirkpatrick CJ. FVIII production by human lung microvascular endothelial cells. *Blood.* 2006;108:515–517.
  84. Durr E, Yu J, Krasinska KM, Carver LA, Yates JR, Testa JE, Oh P, Schnitzer JE. Direct proteomic mapping of the lung microvascular endothelial cell surface in vivo and in cell culture. *Nat Biotechnol.* 2004;22:985–992.
  85. Oh P, Li Y, Yu J, Durr E, Krasinska KM, Carver LA, Testa JE, Schnitzer JE. Subtractive proteomic mapping of the endothelial surface in lung and solid tumours for tissue-specific therapy. *Nature.* 2004;429:629–635.
  86. Valadon P, Garnett JD, Testa JE, Bauerle M, Oh P, Schnitzer JE. Screening phage display libraries for organ-specific vascular immunotargeting in vivo. *Proc Natl Acad Sci U S A.* 2006;103:407–412.
  87. Doerschuk CM. Mechanisms of leukocyte sequestration in inflamed lungs. *Microcirculation.* 2001;8:71–88.
  88. Chetham PM, Babal P, Bridges JP, Moore TM, Stevens T. Segmental regulation of pulmonary vascular permeability by store-operated Ca<sup>2+</sup> entry. *Am J Physiol.* 1999;276:L41–L50.
  89. Kelly JJ, Moore TM, Babal P, Diwan AH, Stevens T, Thompson WJ. Pulmonary microvascular and macrovascular endothelial cells: differential regulation of Ca<sup>2+</sup> and permeability. *Am J Physiol.* 1998;274:L810–L819.
  90. Parker JC, Stevens T, Randall J, Weber DS, King JA. Hydraulic conductance of pulmonary microvascular and macrovascular endothelial cell monolayers. *Am J Physiol Lung Cell Mol Physiol.* 2006;291:L30–L37.
  91. Mitzner W, Wagner EM. Vascular remodeling in the circulations of the lung. *J Appl Physiol.* 2004;97:1999–2004.
  92. Doerschuk CM. Leukocyte trafficking in alveoli and airway passages. *Respir Res.* 2000;1:136–140.
  93. Feuerhake F, Fuchsl G, Bals R, Welsch U. Expression of inducible cell adhesion molecules in the normal human lung: immunohistochemical

- study of their distribution in pulmonary blood vessels. *Histochem Cell Biol.* 1998;110:387–394.
94. Moldobaeva A, Wagner EM. Heterogeneity of bronchial endothelial cell permeability. *Am J Physiol Lung Cell Mol Physiol.* 2002;283:L520–L527.
  95. Murphy TJ, Thurston G, Ezaki T, McDonald DM. Endothelial cell heterogeneity in venules of mouse airways induced by polarized inflammatory stimulus. *Am J Pathol.* 1999;155:93–103.
  96. Andonegui G, Bonder CS, Green F, Mullaly SC, Zbytnuik L, Raharjo E, Kubes P. Endothelium-derived Toll-like receptor-4 is the key molecule in LPS-induced neutrophil sequestration into lungs. *J Clin Invest.* 2003;111:1011–1020.
  97. Silliman CC. The two-event model of transfusion-related acute lung injury. *Crit Care Med.* 2006;34:S124–S131.
  98. Tremblay LN, Slutsky AS. Ventilator-induced lung injury: from the bench to the bedside. *Intensive Care Med.* 2006;32:24–33.
  99. Marini JJ, Hotchkiss JR, Broccard AF. Bench-to-bedside review: microvascular and airspace linkage in ventilator-induced lung injury. *Crit Care.* 2003;7:435–444.
  100. Teichert-Kuliszewska K, Kutryk MJ, Kuliszewski MA, Karoubi G, Courtman DW, Zucco L, Granton J, Stewart DJ. Bone morphogenetic protein receptor-2 signaling promotes pulmonary arterial endothelial cell survival: implications for loss-of-function mutations in the pathogenesis of pulmonary hypertension. *Circ Res.* 2006;98:209–217.
  101. Michelakis ED. Spatio-temporal diversity of apoptosis within the vascular wall in pulmonary arterial hypertension: heterogeneous BMP signaling may have therapeutic implications. *Circ Res.* 2006;98:172–175.
  102. Voelkel NF, Vandivier RW, Tuder RM. Vascular endothelial growth factor in the lung. *Am J Physiol Lung Cell Mol Physiol.* 2006;290:L209–L221.
  103. Eddahibi S, Guignabert C, Barlier-Mur AM, Dewachter L, Fadel E, Darteville P, Humbert M, Simonneau G, Hanoun N, Saurini F, Hamon M, Adnot S. Cross talk between endothelial and smooth muscle cells in pulmonary hypertension: critical role for serotonin-induced smooth muscle hyperplasia. *Circulation.* 2006;113:1857–1864.
  104. Goldie RG, Pedersen KE. Mechanisms of increased airway microvascular permeability: role in airway inflammation and obstruction. *Clin Exp Pharmacol Physiol.* 1995;22:387–396.
  105. Wilson J. The bronchial microcirculation in asthma. *Clin Exp Allergy.* 2000;30(suppl 1):51–53.
  106. Abdalla SA, Letarte M. Hereditary haemorrhagic telangiectasia: current views on genetics and mechanisms of disease. *J Med Genet.* 2006;43:97–110.
  107. McCuskey RS. Morphological mechanisms for regulating blood flow through hepatic sinusoids. *Liver.* 2000;20:3–7.
  108. Oda M, Yokomori H, Han JY. Regulatory mechanisms of hepatic microcirculation. *Clin Hemorheol Microcirc.* 2003;29:167–182.
  109. Braet F, Wisse E. Structural and functional aspects of liver sinusoidal endothelial cell fenestrae: a review. *Comp Hepatol.* 2002;1:1.
  110. Carpenter B, Lin Y, Stoll S, Raffai RL, McCuskey R, Wang R. VEGF is crucial for the hepatic vascular development required for lipoprotein uptake. *Development.* 2005;132:3293–3303.
  111. Zhou B, Weigel JA, Fauss L, Weigel PH. Identification of the hyaluronan receptor for endocytosis (HARE). *J Biol Chem.* 2000;275:37733–37741.
  112. Politz O, Gratchev A, McCourt PA, Schledzewski K, Guillot P, Johansson S, Svineng G, Franke P, Kannicht C, Kzyshkowska J, Longati P, Velten FW, Johansson S, Goerdts S. Stabilin-1 and -2 constitute a novel family of fasciclin-like hyaluronan receptor homologues. *Biochem J.* 2002;362:155–164.
  113. Naito M, Hasegawa G, Ebe Y, Yamamoto T. Differentiation and function of Kupffer cells. *Med Electron Microsc.* 2004;37:16–28.
  114. MacPhee PJ, Schmidt EE, Groom AC. Intermittence of blood flow in liver sinusoids, studied by high-resolution in vivo microscopy. *Am J Physiol.* 1995;269:G692–G698.
  115. Knolle PA, Limmer A. Neighborhood politics: the immunoregulatory function of organ-resident liver endothelial cells. *Trends Immunol.* 2001;22:432–437.
  116. Racanelli V, Rehmann B. The liver as an immunological organ. *Hepatology.* 2006;43:S54–S62.
  117. Limmer A, Ohl J, Kurts C, Ljunggren HG, Reiss Y, Groettrup M, Momburg F, Arnold B, Knolle PA. Efficient presentation of exogenous antigen by liver endothelial cells to CD8<sup>+</sup> T cells results in antigen-specific T-cell tolerance. *Nat Med.* 2000;6:1348–1354.
  118. Limmer A, Ohl J, Wingender G, Berg M, Jungerkes F, Schumak B, Djandji D, Scholz K, Klevenz A, Hegenbarth S, Momburg F, Hammerling GJ, Arnold B, Knolle PA. Cross-presentation of oral antigens by liver sinusoidal endothelial cells leads to CD8 T cell tolerance. *Eur J Immunol.* 2005;35:2970–2981.
  119. Uhrig A, Banafsche R, Kremer M, Hegenbarth S, Hamann A, Neurath M, Gerken G, Limmer A, Knolle PA. Development and functional consequences of LPS tolerance in sinusoidal endothelial cells of the liver. *J Leukoc Biol.* 2005;77:626–633.
  120. Cleaver O, Melton DA. Endothelial signaling during development. *Nat Med.* 2003;9:661–668.
  121. LeCouter J, Moritz DR, Li B, Phillips GL, Liang XH, Gerber HP, Hillan KJ, Ferrara N. Angiogenesis-independent endothelial protection of liver: role of VEGFR-1. *Science.* 2003;299:890–893.
  122. Wong J, Johnston B, Lee SS, Bullard DC, Smith CW, Beaudet AL, Kubes P. A minimal role for selectins in the recruitment of leukocytes into the inflamed liver microvasculature. *J Clin Invest.* 1997;99:2782–2790.
  123. Iigo Y, Suematsu M, Higashida T, Oheda J, Matsumoto K, Wakabayashi Y, Ishimura Y, Miyasaka M, Takashi T. Constitutive expression of ICAM-1 in rat microvascular systems analyzed by laser confocal microscopy. *Am J Physiol.* 1997;273:H138–H147.
  124. Granger DN. Cell adhesion and migration. II. Leukocyte-endothelial cell adhesion in the digestive system. *Am J Physiol.* 1997;273:G982–G986.
  125. Chosay JG, Fisher MA, Farhood A, Ready KA, Dunn CJ, Jaeschke H. Role of PECAM-1 (CD31) in neutrophil transmigration in murine models of liver and peritoneal inflammation. *Am J Physiol.* 1998;274:G776–G782.
  126. Khandoga A, Kessler JS, Meissner H, Hanschen M, Corada M, Motoike T, Enders G, Dejana E, Krombach F. Junctional adhesion molecule-A deficiency increases hepatic ischemia-reperfusion injury despite reduction of neutrophil transendothelial migration. *Blood.* 2005;106:725–733.
  127. Clemens MG, Zhang JX. Regulation of sinusoidal perfusion: in vivo methodology and control by endothelins. *Semin Liver Dis.* 1999;19:383–396.
  128. Kamoun WS, Karaa A, Kresge N, Merkel SM, Korneszcuk K, Clemens MG. LPS inhibits endothelin-1-induced endothelial NOS activation in hepatic sinusoidal cells through a negative feedback involving caveolin-1. *Hepatology.* 2006;43:182–190.
  129. Wisse E, De Zanger RB, Jacobs R, McCuskey RS. Scanning electron microscope observations on the structure of portal veins, sinusoids and central veins in rat liver. *Scan Electron Microsc.* 1983;(pt 3):1441–1452.
  130. Ogi M, Yokomori H, Oda M, Yoshimura K, Nomura M, Ohshima S, Akita M, Toda K, Ishii H. Distribution and localization of caveolin-1 in sinusoidal cells in rat liver. *Med Electron Microsc.* 2003;36:33–40.
  131. Theuerkauf I, Zhou H, Fischer HP. Immunohistochemical patterns of human liver sinusoids under different conditions of pathologic perfusion. *Virchows Arch.* 2001;438:498–504.
  132. Dini L, Carla EC. Hepatic sinusoidal endothelium heterogeneity with respect to the recognition of apoptotic cells. *Exp Cell Res.* 1998;240:388–393.
  133. Hailfinger S, Jaworski M, Braeuning A, Buchmann A, Schwarz M. Zonal gene expression in murine liver: lessons from tumors. *Hepatology.* 2006;43:407–414.
  134. Mori T, Okanoue T, Sawa Y, Hori N, Ohta M, Kagawa K. Defenestration of the sinusoidal endothelial cell in a rat model of cirrhosis. *Hepatology.* 1993;17:891–897.
  135. Bhunchet E, Fujieda K. Capillarization and venularization of hepatic sinusoids in porcine serum-induced rat liver fibrosis: a mechanism to maintain liver blood flow. *Hepatology.* 1993;18:1450–1458.
  136. Le Couteur DG, Cogger VC, Markus AM, Harvey PJ, Yin ZL, Ansellin AD, McLean AJ. Pseudocapillarization and associated energy limitation in the aged rat liver. *Hepatology.* 2001;33:537–543.
  137. Le Couteur DG, Fraser R, Cogger VC, McLean AJ. Hepatic pseudocapillarization and atherosclerosis in ageing. *Lancet.* 2002;359:1612–1615.
  138. Fraser R, Dobbs BR, Rogers GW. Lipoproteins and the liver sieve: the role of the fenestrated sinusoidal endothelium in lipoprotein metabolism, atherosclerosis, and cirrhosis. *Hepatology.* 1995;21:863–874.
  139. DeLeve LD, Ito Y, Bethea NW, McCuskey MK, Wang X, McCuskey RS. Embolization by sinusoidal lining cells obstructs the microcirculation in rat sinusoidal obstruction syndrome. *Am J Physiol Gastrointest Liver Physiol.* 2003;284:G1045–G1052.

140. Rockey DC, Chung JJ. Reduced nitric oxide production by endothelial cells in cirrhotic rat liver: endothelial dysfunction in portal hypertension. *Gastroenterology*. 1998;114:344–351.
141. Shah V, Toruner M, Haddad F, Cadelina G, Papapetropoulos A, Choo K, Sessa WC, Groszmann RJ. Impaired endothelial nitric oxide synthase activity associated with enhanced caveolin binding in experimental cirrhosis in the rat. *Gastroenterology*. 1999;117:1222–1228.
142. Liu S, Premont RT, Kontos CD, Zhu S, Rockey DC. A crucial role for GRK2 in regulation of endothelial cell nitric oxide synthase function in portal hypertension. *Nat Med*. 2005;11:952–958.
143. Couvelard A, Scoazec JY, Feldmann G. Expression of cell-cell and cell-matrix adhesion proteins by sinusoidal endothelial cells in the normal and cirrhotic human liver. *Am J Pathol*. 1993;143:738–752.
144. Mohammed NA, Abd El-Aleem SA, El-Hafiz HA, McMahon RF. Distribution of constitutive (COX-1) and inducible (COX-2) cyclooxygenase in postviral human liver cirrhosis: a possible role for COX-2 in the pathogenesis of liver cirrhosis. *J Clin Pathol*. 2004;57:350–354.
145. Yang R, Liu Q, Grosfeld JL, Pescovitz MD. Intestinal venous drainage through the liver is a prerequisite for oral tolerance induction. *J Pediatr Surg*. 1994;29:1145–1148.
146. Bjorneboe M, Prytz H, Orskov F. Antibodies to intestinal microbes in serum of patients with cirrhosis of the liver. *Lancet*. 1972;1:58–60.
147. Horn T, Christoffersen P, Henriksen JH. Alcoholic liver injury: defenestration in noncirrhotic livers—a scanning electron microscopic study. *Hepatology*. 1987;7:77–82.
148. Clark SA, Angus HB, Cook HB, George PM, Oxner RB, Fraser R. Defenestration of hepatic sinusoids as a cause of hyperlipoproteinaemia in alcoholics. *Lancet*. 1988;2:1225–1227.
149. Levidiotis V, Power DA. New insights into the molecular biology of the glomerular filtration barrier and associated disease. *Nephrology (Carlton)*. 2005;10:157–166.
150. Rostgaard J, Qvortrup K. Electron microscopic demonstrations of filamentous molecular sieve plugs in capillary fenestrae. *Microvasc Res*. 1997;53:1–13.
151. Deen WM, Lazzara MJ, Myers BD. Structural determinants of glomerular permeability. *Am J Physiol Renal Physiol*. 2001;281:F579–F596.
152. Stan RV, Kubitz M, Palade GE. PV-1 is a component of the fenestral and stomatal diaphragms in fenestrated endothelia. *Proc Natl Acad Sci U S A*. 1999;96:13203–13207.
153. Jeansson M, Haraldsson B. Morphological and functional evidence for an important role of the endothelial cell glycocalyx in the glomerular barrier. *Am J Physiol Renal Physiol*. 2006;290:F111–F116.
154. Eremina V, Sood M, Haigh J, Nagy A, Lajoie G, Ferrara N, Gerber HP, Kikkawa Y, Miner JH, Quaggin SE. Glomerular-specific alterations of VEGF-A expression lead to distinct congenital and acquired renal diseases. *J Clin Invest*. 2003;111:707–716.
155. Eng E, Holgren C, Hubchak S, Naaz P, Schnaper HW. Hypoxia regulates PDGF-B interactions between glomerular capillary endothelial and mesangial cells. *Kidney Int*. 2005;68:695–703.
156. Lopez-Ongil S, Diez-Marques ML, Grier M, Rodriguez-Puyol M, Rodriguez-Puyol D. Crosstalk between mesangial and endothelial cells: angiotensin II down-regulates endothelin-converting enzyme 1. *Cell Physiol Biochem*. 2005;15:135–144.
157. Kitahara T, Hiromura K, Ikeuchi H, Yamashita S, Kobayashi S, Kuroiwa T, Kaneko Y, Ueki K, Nojima Y. Mesangial cells stimulate differentiation of endothelial cells to form capillary-like networks in a three-dimensional culture system. *Nephrol Dial Transplant*. 2005;20:42–49.
158. Pallone TL, Turner MR, Edwards A, Jamison RL. Countercurrent exchange in the renal medulla. *Am J Physiol Regul Integr Comp Physiol*. 2003;284:R1153–R1175.
159. Yang B, Bankir L, Gillespie A, Epstein CJ, Verkman AS. Urea-selective concentrating defect in transgenic mice lacking urea transporter UT-B. *J Biol Chem*. 2002;277:10633–10637.
160. Ma T, Yang B, Gillespie A, Carlson EJ, Epstein CJ, Verkman AS. Severely impaired urinary concentrating ability in transgenic mice lacking aquaporin-1 water channels. *J Biol Chem*. 1998;273:4296–4299.
161. Pallone TL, Zhang Z, Rhinehart K. Physiology of the renal medullary microcirculation. *Am J Physiol Renal Physiol*. 2003;284:F253–F266.
162. Neuhofer W, Beck FX. Cell survival in the hostile environment of the renal medulla. *Annu Rev Physiol*. 2005;67:531–555.
163. Han KH, Lim JM, Kim WY, Kim H, Madsen KM, Kim J. Expression of endothelial nitric oxide synthase in developing rat kidney. *Am J Physiol Renal Physiol*. 2005;288:F694–F702.
164. Zhang J, Hill CE. Differential connexin expression in preglomerular and postglomerular vasculature: accentuation during diabetes. *Kidney Int*. 2005;68:1171–1185.
165. Inai T, Sengoku A, Guan X, Hirose E, Iida H, Shibata Y. Heterogeneity in expression and subcellular localization of tight junction proteins, claudin-10 and -15, examined by RT-PCR and immunofluorescence microscopy. *Arch Histol Cytol*. 2005;68:349–360.
166. Lucien N, Bruneval P, Lasbennes F, Belair MF, Mandet C, Cartron JP, Bailly P, Trinh-Trang-Tan MM. UT-B1 urea transporter is expressed along the urinary and gastrointestinal tracts of the mouse. *Am J Physiol Regul Integr Comp Physiol*. 2005;288:R1046–R1056.
167. Pannabecker TL, Dantzer WH. Three-dimensional architecture of inner medullary vasa recta. *Am J Physiol Renal Physiol*. 2006;290:F1355–F1366.
168. Lee-Kwon W, Wade JB, Zhang Z, Pallone TL, Weinman EJ. Expression of TRPC4 channel protein that interacts with NHERF-2 in rat descending vasa recta. *Am J Physiol Cell Physiol*. 2005;288:C942–C949.
169. Panzer U, Steinmetz OM, Reinking RR, Meyer TN, Fehr S, Schneider A, Zahner G, Wolf G, Helmchen U, Schaefer P, Stahl RA, Thaiss F. Compartment-specific expression and function of the chemokine IP-10/CXCL10 in a model of renal endothelial microvascular injury. *J Am Soc Nephrol*. 2006;17:454–464.
170. Holmen C, Christensson M, Pettersson E, Bratt J, Stjerne P, Karrar A, Sumitran-Holgersson S. Wegener's granulomatosis is associated with organ-specific antiendothelial cell antibodies. *Kidney Int*. 2004;66:1049–1060.
171. Ray PE, Liu XH. Pathogenesis of Shiga toxin-induced hemolytic uremic syndrome. *Pediatr Nephrol*. 2001;16:823–839.
172. Okuda T, Tokuda N, Numata S, Ito M, Ohta M, Kawamura K, Wiels J, Urano T, Tajima O, Furukawa K, Furukawa K. Targeted disruption of Gb3/CD77 synthase gene resulted in the complete deletion of globo-series glycosphingolipids and loss of sensitivity to verotoxins. *J Biol Chem*. 2006;281:10230–10235.
173. Cooper ME, Vranes D, Youssef S, Stacker SA, Cox AJ, Rizkalla B, Casley DJ, Bach LA, Kelly DJ, Gilbert RE. Increased renal expression of vascular endothelial growth factor (VEGF) and its receptor VEGFR-2 in experimental diabetes. *Diabetes*. 1999;48:2229–2239.
174. Hovind P, Tarnow L, Oestergaard PB, Parving HH. Elevated vascular endothelial growth factor in type 1 diabetic patients with diabetic nephropathy. *Kidney Int Suppl*. 2000;75:S56–S61.
175. Flyvbjerg A, Dagnaes-Hansen F, De Vriese AS, Schrijvers BF, Tilton RG, Rasch R. Amelioration of long-term renal changes in obese type 2 diabetic mice by a neutralizing vascular endothelial growth factor antibody. *Diabetes*. 2002;51:3090–3094.
176. Kikuchi Y, Ikee R, Hemmi N, Hyodo N, Saigusa T, Namikoshi T, Yamada M, Suzuki S, Miura S. Fractalkine and its receptor, CX3CR1, upregulation in streptozotocin-induced diabetic kidneys. *Nephron Exp Nephrol*. 2004;97:e17–e25.
177. Gooch JL, Pergola PE, Guler RL, Abboud HE, Barnes JL. Differential expression of calcineurin A isoforms in the diabetic kidney. *J Am Soc Nephrol*. 2004;15:1421–1429.
178. Hiragushi K, Wada J, Eguchi J, Matsuoka T, Yasuhara A, Hashimoto I, Yamashita T, Hida K, Nakamura Y, Shikata K, Minamino N, Kangawa K, Makino H. The role of adrenomedullin and receptors in glomerular hyperfiltration in streptozotocin-induced diabetic rats. *Kidney Int*. 2004;65:540–550.
179. Nakatani K, Fujii H, Hasegawa H, Terada M, Arita N, Ito MR, Ono M, Takahashi S, Saiga K, Yoshimoto S, Iwano M, Shiiki H, Saito Y, Nose M. Endothelial adhesion molecules in glomerular lesions: association with their severity and diversity in lupus models. *Kidney Int*. 2004;65:1290–1300.
180. Pardridge WM. Molecular biology of the blood-brain barrier. *Mol Biotechnol*. 2005;30:57–70.
181. Abbott NJ, Ronnback L, Hansson E. Astrocyte-endothelial interactions at the blood-brain barrier. *Nat Rev Neurosci*. 2006;7:41–53.
182. Luster AD, Alon R, von Andrian UH. Immune cell migration in inflammation: present and future therapeutic targets. *Nat Immunol*. 2005;6:1182–1190.
183. Oliver G, Detmar M. The rediscovery of the lymphatic system: old and new insights into the development and biological function of the lymphatic vasculature. *Genes Dev*. 2002;16:773–783.
184. Feng D, Nagy JA, Pyne K, Hammel I, Dvorak HF, Dvorak AM. Pathways of macromolecular extravasation across microvascular endothelium in response to VPF/VEGF and other vasoactive mediators. *Microcirculation*. 1999;6:23–44.



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