RESEARCH IN MICROCIRCULATION IN HEALTH AND DISEASE:
AN OVERVIEW

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Introduction
In this brief review the basic structure of the microcirculation is presented. Some relatively recent research findings made in this field will be also reviewed with regards to their impact on diseases or pathologies of clinical significance. In each process described below, the focus will be on cellular and vascular events. In many (if not all) cases a host of molecular events takes place simultaneously but they will not be reviewed due to the limited scope of this overview.

One of the reasons for convening in this conference is that for years the treatment of patients has been based on the recovery of systemic parameters like arterial blood pressure. Unfortunately, sometimes the patient dies despite this recovery. Perhaps we have been looking at the wrong parameter, i.e., the blood pressure was reestablished but the microcirculation remained shut down. Currently the best parameters to be monitored for the best outcome after some life-threatening emergencies are still unknown. Some answers may lie in the microcirculation! Similar statements may be true for the treatment of many diseases. Modulation of microvessel activity may be essential to alter the whole organ function.

The survival, integration and proper functioning of organs depends upon suitable transport and exchange of various molecules between the flowing blood and parenchymal cells. This continuous process takes place at the level of the microcirculation, and includes gases such as oxygen (O$_2$) and carbon dioxide (CO$_2$), nutrients, waste products, hormones as well as various molecules and cells linked to the immune response, inflammation and coagulation. Since the system is so deeply involved in nearly all physiological functions, it is not surprising that all pathologies (and potential treatments) will implicate one or more elements of the microcirculation.

Microcirculation – Structure and Function
The microcirculation includes the most distal part of the vascular system, typically comprising vessels with an internal diameter of less than 100 µm (Fig. 1). These microvessels (arterioles, capillaries, or venules - depending on anatomic features and direction of blood flow) form complex networks in most tissues (Popel et al. 2005). Several arterioles connect to many capillaries that drain to numerous venules. Many vessels are interconnected and the internal diameter of each input arteriole is variable. Therefore, local variables such as pressure, flow, oxygenation, and hematocrit, will vary depending on the balance of constrictive and dilating influences actuating on different arterioles. Moreover, rheological factors such as blood viscosity and leukocyte adhesion may play a role in local blood flow and O$_2$ distribution. Since the microcirculatory vessels are embedded within the organs, the communication between parenchymal tissue cells and the flowing blood is a critical function provided by the microcirculation. This exchange maintains cell survival and integration through processes that include passage of water, solutes, proteins, and even some blood cells through the microvascular wall. The microcirculation is involved in the transport of drugs and medications as well as in the control of temperature. The lymphatic vessels are also included in several microcirculatory studies.
Glycocalyx

The endothelial glycocalyx is a layer covering the lumen of the blood vessels. The glycocalyx protects and exerts several physiological functions related to permeability, flow, cell adhesion, and coagulation. The glycocalyx can be studied in vivo using microscopy imaging techniques. A compound (fluorescent Dextran) that does not penetrate the glycocalyx is injected into the bloodstream. The lumen of the vessel and the diameter of the fluorescent column are measured using the images; by comparing the difference between the two measurements, the glycocalyx thickness is established (Torres Filho et al. 2013). This thickness can decrease after hemorrhage (post-hemorrhage degradation or shedding).

These are several situations and diseases where the glycocalyx thickness has been shown to decrease or disappear (e.g., dialysis, high fat diets, hyperglycemia, diabetes, inflammation). The glycocalyx can also decrease following hemorrhage (post-hemorrhage degradation or shedding), both in humans and experimental animals. Treatment with solutions like lactated Ringer’s or saline offers no improvement but plasma can allow the glycocalyx to recover (Torres et al. 2013; 2016a). This is important for situations when hemorrhages occur, e.g., car crash or soldiers’ injuries on the battlefield. Which solution should be used (in case of hemorrhage) to recover the small vessels (microcirculation) as well as the arterial blood pressure?

Vasomotion and flowmotion

Vasomotion is the spontaneous cyclical variations in the lumen of vessels. These oscillations can have various frequencies but should not be confounded with different, unrelated phenomena such as heart rate. In the case of shock resulting from blood loss the vasomotion increased in animals treated with hypertonic saline solution both in vessels that had vasomotion before the treatment during hypotension and in vessels that did not (Torres Filho et al., 2001). The treatment that improved outcome in these animals also improved vasomotion. While cause and effect could not be precisely established in these experiments, these findings
deserve further investigation. Moreover, vasomotion occurs in normal healthy people and animals.

When diameter in a vessel changes, the flow produced downstream also changes. Changes in diameter are called vasomotion, while downstream changes are named flowmotion. In a recent study, diabetic ulcer patients treated with hyperbaric oxygen therapy (HBOT) showed improved condition of the ulcer and simultaneous changes in vasomotion/flowmotion activity (Fig. 2). The authors concluded that flowmotion dynamics may partly explain the positive effect of HBOT on the healing process of a diabetic ulcer (Balaz et al., 2016).

![Graph showing changes in diameter and flowmotion](image)

**Figure 2.** Hyperbaric oxygen therapy changes flowmotion dynamics that may be involved in the healing of ulcers in diabetic patients. Reproduced with permission from Balaz et al. (2016). Colors and arrows added for clarity and emphasis.

**Thrombus formation in vivo**

When the inside of a vessel is damaged using a laser, passing blood platelets immediately attach to the vessel wall (endothelium). It is a precursor to mitigate vessel perforation, i.e., the platelets form a clot. Since some of these platelets have been made fluorescent, they are easier to visualize, record and count. This experimental procedure allows in vivo testing of the functionality of platelets, and the efficacy of platelets that have been under various conditions of storage (different temperatures and periods of time). These techniques use a special microscope, and may help to find fluids to treat soldiers with hemorrhage on the battlefield (Torres Filho et al., 2016b).

**Inflammation**

Many steps of the inflammatory process occur at the level of the microcirculation. Leukocytes (white blood cells), as part of the defense mechanisms, intermittently roll along the inside wall of post-capillary venules and scan for bacteria, foreign particles, etc. Various other leukocyte activities have been described such as firm adhesion to endothelium and transmigration (Popel et al. 2005). The inflammatory process also includes changes in microvascular permeability, diameter and flow.

**Oxygenation**

Oxygenation can be studied through PO$_2$ (oxygen partial pressure) and SO$_2$ (oxygen saturation) measurements in the microcirculation. Vessel diameter and blood flow can also be measured in arterioles and venules allowing the determination of O$_2$ delivery and O$_2$ consumption. PO$_2$ measurements in normal tissues and human tumors have been made with techniques such as the phosphorescence quenching (Torres Filho et al., 1993). The PO$_2$ in certain areas within a tumor starts high but rapidly decreases, becoming very low over relatively small distances (Torres Filho et al., 1994). This is critical because when oxygenation is low (hypoxia) tumors become resistant to radiotherapy.
Moy et al. (2011) and Styp-Rekowska et al. (2007) measured SO₂ using different methodologies and achieved results that allowed relatively detailed maps of the SO₂ distribution in large areas of the microcirculation.

Human Microcirculation

Using non-invasive techniques such as orthogonal polarization spectral imaging and side stream dark field microscopy the human microcirculation (especially the sublingual mucosa) can be studied (Ward et al., 2010). Different situations have been examined such as septic shock, hemorrhagic shock, heart failure, and sickle cell disease. These methods have been extensively used in Europe but also in North and South America (Massey et al., 2016).

Summary

There are new methodologies for examining various microcirculatory functions in vivo. These data suggest that the modulation of the microcirculation can affect or improve the quality of life and in many circumstances mean the difference between life and death.

Disclaimer

The opinions and assertions contained herein are the private views of the author and are not to be construed as official or as reflecting the views of the Department of the Army or the U.S. Department of Defense. This study has been conducted in compliance with the Animal Welfare Act, the implementing Animal Welfare Regulations, and the principles of the Guide for the Care and Use of laboratory Animals, using protocols approved by the Institutional Animal Care and Use Committee of the U.S. Army ISR.

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