There is strong evidence that chronic inflammation contributes to a number of conditions prevalent with aging, including peripheral vascular disease, rheumatoid arthritis, as well as Alzheimer’s disease. Yet despite being responsible for mediating immune responses, the lymphatic vasculature has largely escaped attention of biomedical engineers, research scientists, pharmaceutical companies, and health care professionals. The reason for this may be because the lymphatics are difficult to directly visualize in tissues. Interstitial fluid, immune cells, and foreign particles form colorless “lymph” when entering the epidermis and line all the major organs of the body. From there the lymph is transited through the collecting and conducting vessels, the latter of which consist of a series of “lymph hearts” or lymphangions that propel lymph through the main thoracic duct before emptying into the supraclavicular vein (Fig 1). Conventional lymphatic imaging employs the administration of contrast agents for visualizing the lymphatics and lymph nodes using intradermal administration of radiocolloid for lymphoscintigraphy, or using surgical procedures to inject gadolinium or iodinated contrast agents for magnetic resonance or CT imaging. These techniques are not routinely used and none are suitable for imaging the lymphatics in a mouse or rat, the predominant animal models used in discovery research. Beginning in 2005, we began imaging the dynamic activity of the lymphatics in humans following intradermal injection of indocyanine green, illumination of tissues with 1.9 mW/cm2 of 785 nm near-infrared (NIR) light, and collection of the dim 830 nm NIR fluorescent (NIRF) light using a InGaAs Gen III intensifier coupled conventional CCD or sCMOS detector. Shortly thereafter, we began imaging lymphatic activity in animal models of human disease. Because of the sensitivity of the fluorescence technique, images can be acquired in 50 -200 ms to non-invasively visualize the dynamic lymphangion activity in vessels as deep as 3-4 centimeters for the first time in human adults. As a result of the rapid, “point-of-care” imaging offered by NIRF, we are able to image infants and children without the need for sedation (Greives, et al., Pediatrics, 2017).

In humans, we used NIRF lymphatic imaging to show that lymphangion activity is impaired and showed “reflux” in both the early and late stages of peripheral vascular disease (Fig. 2) as compared to normal human subjects (Rasmussen, et al., J Vasc. Surg. VLD, 2016). Presumably due to a weak lymphatic pump, lymph “pooling” occurred in the ankles and other sites where at later stages of the disease, tissue breakdown and vascular wounds occur. In mice, we discovered that pro-inflammatory cytokines, TNF, IL-1β and IL-6 arrest lymphangion activity (Aldrich and Sevick-Muraca, Cytokines, 2013). In rats with early collagen induced arthritis, the lymphangions are inactive or pump in the opposite direction, resulting in swelling and destructive accumulation of immune cells that in later stages of the disease erode bone. Using nanotopography infusion devices to administer TNF inhibitor etanercept directly into the lymphatics of these animals, we showed recovery of lymphangion activity and reduction of swelling faster than when administered through conventional s.c. injection (Aldrich, et al., Arthritis Res. Ther., 2017). This and other works suggest that rescue of lymphangion activity as seen through NIRF lymphatic imaging could prove a new and potentially more effective strategy to treat chronic inflammatory conditions. Future work in this important area includes using imaging to understand the mechanisms behind what drives lymphangion activity so that pharmacological approaches to stimulate the lymphatic pump can be developed. Future engineering for the development of 3-D lymphangiography and automated real-time analysis of the lymphangion “pump” and reflux rates could make these technologies more widely available.