Cancer is a heterogeneous disease that originates from normal tissue that undergoes a transformation to a diseased state. The transformation of normal tissue to tumor tissue requires acquisition of biological capabilities that result in certain hallmarks of cancer, including sustaining proliferative signaling, evading growth suppression signaling, resisting cell death, enabling cellular replicative immortality, activating invasion and metastasis, and inducing angiogenesis[1, 2]. Because tumors arise from normal tissue it is difficult to specifically treat cancer by systemic administration of chemotherapy drugs. However, the hallmarks of this transformation can give rise to opportunities to exploit the differentiation between normal and tumor tissue. Specifically for drug delivery, enhanced permeability and retention (EPR) of nanoparticles in the tumor environment arises due to tumor-associated angiogenesis.

Induction of angiogenesis, or new blood vessel formation, is required for tumors to grow beyond a few millimeters in size due to the need for oxygen and nutrients to support cell metabolism and proliferation[3]. Activation of the angiogenic switch causes existing vasculature to continually sprout based on signaling from expanding tumor and stromal cells[4]. Typically, the neovasculature resulting from angiogenesis results in excessive vessel sprouting, enlarged and distorted vessels, erratic blood flow, microhemorraging, leakiness, loose pericyte coverage, and poor lymphatic drainage[5-7]. These characteristics of tumor vasculature can lead to selective accumulation and increased resonance time of nanoparticles in the tumor environment due to the hyper-permeability of the vessels and lack of lymphatic drainage[8-18]. The enhanced permeability and retention of macromolecules in the tumor environment was first described in 1986, and has resulted in an ongoing field of research to characterize the EPR effect for cancer therapy[8, 19, 20]. Over the past 30 years a greater understanding of the physiology of the EPR effect has led to defining characteristics for nanoparticle design including biocompatibility, surface charge, size, shape, intracellular transport, and drug release.

In order for systemically administered nanotherapeutics to accumulate in the tumor environment they must first evade detection by the mononuclear phagocytic system (MPS). Nanoparticles must possess a biocompatible component to impart a stealth quality to escape immune surveillance. One of the most common ways to impart biocompatibility for nanotherapeutics is by surface modification with poly(ethylene glycol) (PEG)[21-25]. This chemically inert modification allows for evasion of the MPS, thus reducing immunogenicity and extending the blood circulation time of nanoparticles. The tumor accumulation of the nanoparticles due to the EPR effect is thereby increased by increasing the number of passes that a nanoparticle can make through the tumor, or tumor microenvironment. Biocompatibility of the nanoparticle is also charge dependent. Nanoparticles with negative or positive surface charges undergo opsonization, which is protein binding in the blood, which causes phagocytosis by MPS cells such as macrophages and Kupffer cells[26, 27]. Nanoparticles with neutral surface charge, as imparted by PEG surface modification, demonstrate inhibited binding of opsonin proteins and remain circulating in the blood compartment for extend periods of time.

Once nanoparticles make their way to the tumor and surrounding microenvironment the ability to transverse the epithelial lining of the tumor neovasculature is highly dependent on the size of the nanoparticle. The optimal size range for tumor accumulation has been shown to be above 12 nanometers and below 200 nanometers in hydrodynamic diameter[17, 28, 29]. Angiogenesis causes highly unorganized, leaky vasculature in the tumor and surrounding environment, which can be taken advantage of by nanoparticles in this size range. The particles
should be larger than the pores of vessels in normal tissue and large enough to escape renal clearance, which is typically above 12 nanometers in diameter[10, 28, 30-32]. In addition, the particles should be small enough to escape filtration by the liver and spleen, which is typically less than 200 nanometers in diameter[33-35].

The size and shape of the nanoparticle, as well as the difference between the microvascular and interstitial pressure play roles in the transvascular transport of nanoparticles from vessel to interstitial tumor space and tumor accumulation. The size and shape of the nanoparticles govern the interaction between the particle and vessel wall. The particle can have an affinity for the vessel wall due to steric collisions with the vessel or electrostatic interactions based on the charge of the particle and vessel wall, or there can be hydrodynamic forces induced by the motion of the particle within the fluid medium[36-38]. These interactions are all controlled by the ratio of particle size to vessel pore size, where smaller particles are less hindered than larger particles[37]. The interstitial fluid pressure within the tumor can also dictate the ability of the particle to diffuse from the vessel to the tumor. This pressure is tumor specific, and is controlled by the leakiness of the vessels and the lymphatic drainage of fluid within the tumor. If drainage is poor within the tumor then the interstitial fluid pressure may be as high, or higher, than the microvascular pressure resulting in a lack of pressure gradient to drive the particle into the tumor interstitial space[6, 37, 39]. In this case the particle size alone governs the tumor accumulation. However, in tumors with interstitial pressure less than the microvascular pressure, particles will actively be pushed through vessel pores resulting in increased accumulation of the nanoparticles[16, 36, 40, 41].

Particle size, charge, and interstitial pressure within the tumor also play important roles in the distribution of the nanoparticles, and subsequent drug delivery, within the tumor tissue and microenvironment. Nanoparticles that accumulate in tumors with high interstitial pressure may have difficulty penetrating deep into the tumor interior, or into areas that are hypovascular[9, 41, 42]. Tumor microenvironments contain fibrous elements from collagen that may limit distribution based on particle size, and can contain positively charged collagen elements and negatively charged hyaluronic acid which can give rise to electrostatic repulsion of charged nanoparticles[18, 43, 44]. While uniform penetration of particles throughout the tumor is important, ultimately the ability of the particles to release their drug payload is paramount to the efficacy of the therapeutic. Particles that rely on cellular uptake for drug release need to penetrate all regions of the tumor to provide uniform dosing of the therapeutic. Delivery systems that provide a triggered release of drug payload in the tumor as well as the tumor microenvironment rely less on uniform distribution throughout the tumor[25, 45]. This can be accomplished using a pH-dependent release of drug payload. The acidic tumor environment can allow for release of the encapsulated small molecule therapeutic, which can then distribute throughout the tumor and tumor microenvironment much easier than a macromolecule[46, 47].

Taken together, studies on the EPR effect over the past thirty years have provided potential advantages, as well as challenges, for the delivery of nanoparticles for cancer therapy. Specific design elements of nanoparticle delivery systems such as biocompatibility, size, shape, charge, and drug release all play an important role for maximizing antitumor efficacy. Other factors such as tumor vasculature, interstitial pressure, and microenvironment characteristics give rise to variables that are different among tumor sites and patients. Ultimately, the characteristics and utility of these systems will become better defined as more formulations make their way into use in the clinic.