1 Introduction

Optical phantoms are a widely used tool to validate optical instrumentation. In essence, phantoms are "false tissues" made of various materials and can be liquid, solid, or gelatinous. Generally, phantoms are made to either simulate a tissue's optical, mechanical, chemical, or physical properties, or a combination of these. These structures are typically comprised of a base substrate material, which can be doped with certain additives that give the material specific optical, mechanical, or chemical properties. Additionally, depending on the substrate material used and desired geometry, optical phantoms can be molded into a variety of shapes and sizes for different applications. ^{1–3}

This chapter opens with a review of optical phantoms as validation tools for imaging and spectroscopy platforms, with a majority of the discussed optical instrumentation being probe based. This will provide insight into the use of optical phantoms within the context of current biomedical optics research. Following this, the "diffuse" and "sub-diffuse" scattering regimes governing light transport through tissue will be addressed. Distinguishing between the "diffuse" and "sub-diffuse" scattering regimes is important for several reasons, including: (1) generating design requirements for probe-based instrumentation, especially for determining appropriate source-detector separations in spectroscopy probes, (2) evaluating the accuracy of computational or numerical models of light transport, and (3) understanding the optical properties of target human tissues and designing optical phantoms to mimic such tissues. Finally, this chapter concludes with a tutorial on how to construct thin, solid, multilayer tissue-simulating phantoms using a spin-coating technique for a variety of applications, and a tutorial on how to construct liquid phantoms to build a lookup table (LUT) inverse model to extract optical properties using diffuse or sub-diffuse reflectance spectroscopy.

1.1 Optical phantoms in probe-based systems

Each subtype of optical phantom discussed in this section will be described based on their intended application, substrate material, scattering and absorbing agents, corresponding reduced scattering coefficients (μ_s) and absorption coefficients (μ_a), and experimental design. Additionally, the probe and/or detector used to acquire data will be briefly described. Optical phantoms for validating instrumentation in high-resolution microendoscopy, hyperspectral imaging, diffuse optical tomography (DOT), reflectance spectroscopy, fluorescence spectroscopy, and Raman spectroscopy will be discussed.

1.1.1 High-resolution microendoscopy

High-resolution microendoscopy is a noninvasive imaging technique that couples a light source to a small image-fiber probe. The probe is placed in contact with an exogenously stained sample, is excited via an illumination source, and then