from deoxyribonucleic acid (DNA) templates inside the cell nucleus. Preprohormones, peptide hormone precursors, are then processed in several stages, typically in the endoplasmic reticulum, including the removal of the N-terminal signal sequence and sometimes glycosylation, resulting in prohormones. The prohormones are then packaged into membrane-bound secretory vesicles, which can be secreted from the cell by exocytosis in response to specific stimuli (e.g., an increase in Ca^{2+} and cyclic adenosine monophosphate concentration in the cytoplasm). These prohormones often contain superfluous amino acid residues that were needed to direct the folding of the hormone molecule into its active configuration but have no function once the hormone folds. Specific endopeptidases in the cell cleave the prohormone just before it is released into the bloodstream, generating the mature hormone form of the molecule. Mature peptide hormones then travel through the blood to all of the cells of the body, where they interact with specific receptors on the surfaces of their target cells.

The U.S. Food and Drug Administration (FDA) expects the sponsors to come up with a high level of scientific evaluation of their biosimilar candidate products, and it all begins with a full understanding of proteins as it is relevant to their development as biosimilar products. This chapter is not a primer on protein science, which I assume would be well understood by the sponsor, but a description of what is relevant to the development of biosimilars, as the FDA views it.

The first leg of establishing biosimilarity is a demonstration of structural similarity between a biosimilar candidate and the originator product. In this chapter, I describe the nature of the structural variants that are common, not necessarily always relevant, and the techniques available to establish this basic level of similarity. This is the step where we begin collecting evidence that will lead us to the totality of the evidence.

1.1.1 Developing biosimilars

If a new biological product development is akin to a horse running wild and reaching the goal post, biosimilar development is like running a horse on exactly the same track without any fences around the track. In one case, it is uncertain; in the other, extremely onerous. This chapter provides details of the critical aspects of protein and antibody structures that are relevant to establishing biosimilarity. This is not a primer on protein chemistry, as only those elements that are relevant to a regulatory development of biosimilar are provided here.

The nature of products that a biosimilar product developer will face greatly varies, even though they are all proteins. Our body cells exploit an enormous array of proteins, approximately 2000, to perform nearly every functional and structural role to stay alive. To date, more than 130 genuine and a similar number of modified therapeutic proteins are approved for clinical use in the European Union and the United States