Proteases are among the most critical of all proteins expressed by living organisms in mediating pathways of cell life and death. In fact, the initial interactions between protease and substrate, and the subsequent cleavage lies at base of a vast spectrum of essential biological events including thrombosis, coagulation, and apoptosis. Deregulated proteolysis has been implicated in many pathologies including cancer, autoimmune disease, inflammatory, and infectious disease. Thus, designing inhibitors to target proteases provides a sound and versatile approach toward new drugs discovery. In 1985, George P. Smith pioneered phage technology when he expressed short peptides on the surface of the filamentous bacteriophage M13 by inserting synthetic DNA sequences, coding for peptides, into the phage coat protein gene, gIIIp. This was accomplished without affecting phage infectivity and viability. Then random peptide libraries have been cloned into phages or into proteins fused to gIIIp, expanded to extremely large size, and screened in vitro for target protein binding. This type of approach has enabled the study of different types of protein-protein interactions including those between epitope and antibody (randomized variable regions), receptor and ligand (randomization of region for binding to the receptor), and protease and inhibitor (randomization of residues critical for interaction with protease). Matthews and Wells have modified the method to enable using proteolytic activity of proteases to identify novel corresponding substrates. This powerful technique is now known as phage display substrate.