target the viral entry process, particularly fusion or various functions of the RNA transcription/replication complex.

A significant unmet medical need exists for the development of a convenient, safe and effective antiviral therapy for RSV infection. The goal of an antiviral therapeutic would be to block the progression to a lower respiratory tract infection and reduce the number and duration of hospitalizations. The development of direct-acting antivirals for RSV has been slow but a common theme has been the ability to identify inhibitors of the virus fusion process with its target cell. This chapter reviews the challenges, both pre-clinical and clinical, and recent progress that has been made in the development of RSV fusion inhibitors.

2.2 Challenges in the Development of RSV Antivirals

The challenges associated with the discovery and development of RSV fusion (F) inhibitors range from understanding the fundamental interactions of inhibitors with the fusion protein to clinical trial design and demonstration of clinical efficacy. The RSV fusion protein is a critical component of the fusion process along with the attachment protein. The mechanism under which an uninfected cell becomes infected with the virus is poorly understood, in part because it is a dynamic, energy-driven process involving large structural changes of the fusion protein. The F protein is a type I, single-pass integral membrane protein that is embedded in the viral envelope and inserts into the host cell membrane. Until recently, the identity of the target cell surface protein that might be involved in cell recognition and attachment was not known. However, recent reports now suggest that nucleolin, a nuclear receptor that is shuttled from the nucleus to the cell surface, might play a key role in mediating RSV infection.[10,11]

The fact that the fusion protein is a membrane-bound protein with a dynamic mode of action has made access to structural information, especially small-molecule X-ray structure, difficult to obtain. The first report of an X-ray structure of a F protein–inhibitor complex appeared only recently and provides some insight into one potential binding interaction that is consistent with data concerning residues that mutate to reduce inhibitor binding.[12] Other reports have identified sites at which monoclonal antibodies, *e.g.*, motavizumab may also bind to residues of the RSV fusion protein.[13] At present, however, the utility of structural information that can guide structure-based design approaches to inhibitors of the RSV F protein is very limited, which presents a challenge in the design of optimal fusion inhibitors. In addition to the lack of structural information, there is also a limitation in biochemical assays available for evaluation of binding interactions. Therefore, to date, small-molecule inhibitors that have been identified and optimized have been discovered by screening of libraries using a virus replication assay, followed by empirical approaches to optimization. Indeed, compared with many other viruses such as HIV and HCV, the amount of X-ray structural data to drive RSV inhibitor design has been minimal. Given the wide variety of small-molecule structures