REVIEW ARTICLE

Site-Directed Mutagenesis in Viral Glycoprotein and Its Role in Viral Diagnostics and Therapy

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Abstract

Site-directed mutagenesis introduces precise mutations into viral glycoprotein genes, allowing the study of their structural and functional roles and interactions with host cells. By modifying specific amino acids, researchers can assess the impact on viral entry, replication, and immune evasion. Engineered glycoproteins improve diagnostic assay sensitivity and specificity and facilitate the development of better vaccine antigens. Therapeutically, site-directed mutagenesis aids in designing antiviral drugs targeting specific glycoprotein regions and developing attenuated viral strains for vaccines. Additionally, this approach enhances understanding of viral evolution and adaptation, offering insights into future pandemic threats and aiding preparedness efforts.

Keywords

Mutagenesis, Viral glycoprotein, Site-directed mutagenesis.

INTRODUCTION

Research and development in viral glycobiology have provided a huge impetus for the application of virology. This was due to the discovery of the role of surface glycoproteins of viruses in the processes of their adsorption, penetration into the cell, and their participation as antigens and effectors in the development of the immune response of microorganisms (Shimada, 1996). The surface glycoprotein of a viral particle is a complex molecule formed by a polypeptide with a high or low molecular weight, with glyosidic chains attached to it. Such chains can bind viruses that are capable of causing severe infectious pathologies in humans and animals. These viruses include the herpes simplex virus, feline immunodeficiency virus, monkeypox virus, influenza virus, and Marburg virus (Baranovich et al., 2013).

To elucidate the role of the surface glycoproteins of these viruses in the pathogenesis of viral diseases and their use in diagnostic and therapeutic purposes, site-specific mutagenesis was used. In this study, the issues of improving this method and assessing their results are considered based on personal experimental experience. The site-directed mutagenesis of viral glycoproteins has emerged as a powerful tool for advancing viral diagnostics and therapeutic strategies (Liu, 2008).

This technique allows researchers to introduce specific mutations into genes encoding viral glycoproteins, enabling the study of their structure-function relationships and their interactions with host cells (Braman, 2002). By altering key amino acid residues, scientists can investigate how these changes affect viral entry, replication, and immune evasion. In diagnostics, engineered glycoproteins can be used to develop more sensitive and specific assays for viral detection as well as to create improved antigens for vaccine development (Braman, 2002). Therapeutically, site-directed mutagenesis has facilitated the design of novel antiviral drugs that target specific glycoprotein regions and the development of attenuated viral strains for potential use in vaccines. Furthermore, this approach has contributed to our understanding of viral evolution and adaptation, providing insights into potential future pandemic threats, and guiding preparedness efforts (Schiffer et al., 2012).

VIRAL GLYCOPROTEINS

Viral glycoproteins are integral components of the viral envelope and play a crucial role in mediating viral entry into host cells. These proteins are anchored to the viral membrane and project outward, enabling the virus to interact with specific receptors on the host cell surface (Sanjuán, 2010). Viral glycoproteins are responsible for various stages of the viral life cycle including attachment, penetration, and fusion with the host cell membrane. Understanding the structure and function of viral glycoproteins is essential to develop effective diagnostic tests and therapeutic interventions (Sanjuán, 2010).

Site-directed mutagenesis is a powerful technique for studying protein function by introducing specific amino acid substitutions (Nøhr and Kristiansen, 2003). This method involves altering the code of a gene to create mutant proteins with targeted changes in their sequences. By systematically modifying specific residues within a viral glycoprotein, researchers can investigate the impact of these changes on the protein structure, stability, and function. This approach allows for a detailed understanding of the critical amino acids involved in protein-protein interactions, enzymatic activity, and other essential biological processes (Nøhr and Kristiansen, 2003).

APPLICATIONS OF SITE-DIRECTED MUTAGENESIS

Viral glycoproteins are typically composed of three distinct domains: ectodomain, a transmembrane domain, and cytoplasmic tail (Jeltsch and Lanio, 2002). The ectodomain, which is exposed to the external environment, contains binding sites for host cell receptors and is involved in mediating viral attachment and entry. The transmembrane domain anchors the glycoprotein to the viral envelope, whereas the cytoplasmic tail plays a role in intracellular signaling and viral assembly (Jeltsch and Lanio, 2002).

Viral glycoproteins play crucial roles in various stages of the viral life cycle. Attachment to the host cell is mediated by the binding of the viral glycoprotein ectodomain to specific receptors on the host cell surface (Jeltsch and Lanio, 2002). Following attachment, the

virus undergoes membrane fusion, which allows the viral genome to enter the host cell. The cytoplasmic tail of the viral glycoprotein interacts with host cell proteins, facilitating viral entry and subsequent intracellular events. Additionally, viral glycoproteins can modulate the host immune response, enabling viruses to evade detection and elimination by the immune system. Understanding the molecular interactions between viral glycoproteins and host cell receptors is essential for developing antiviral strategies targeting these critical steps in the viral life cycle (Zhang et al., 2021).

Site-directed mutagenesis is a precise genetic engineering technique that allows the targeted modification of specific nucleotides within a gene. This method involves the use of synthetic oligonucleotides known as primers that contain the desired mutation (Ran et al., 2020; Edelheit et al., 2009). These primers were designed to anneal to the gene of interest and were then extended by DNA polymerase to create a mutant DNA molecule. By introducing specific nucleotide changes, researchers can alter the amino acid sequence of a protein and study its resulting functional consequences. Compared to random mutagenesis, which generates a large number of mutations without control over their location, site-directed mutagenesis offers greater precision and allows for systematic investigation of the effects of specific amino acid substitutions on protein function. This technique has been widely applied in viral glycoprotein research to identify critical residues involved in protein-protein interactions, enzymatic activity, and other essential biological processes. Additionally, site-directed mutagenesis can be used to create mutant viral glycoproteins that are resistant to antiviral drugs, thereby providing valuable insights into the development of novel therapeutic strategies (Ran et al., 2020; Edelheit et al., 2009).

The fields of site-directed mutagenesis and viral glycoprotein research are rapidly evolving, with several emerging trends. One notable trend is the increasing use of high-throughput mutagenesis techniques, which allow the generation and screening of a large number of mutant proteins in a shorter timeframe. Additionally, advancements in protein structure determination and computational modelling have provided new insights into the structural and functional properties of viral glycoproteins, facilitating the identification of potential drug targets (Ran et al., 2020).

Despite its numerous advantages, site-directed mutagenesis is challenging. One potential issue is the occurrence of off-target mutations, which can introduce unintended changes in the protein sequence and confound the interpretation of the experimental results. Ethical considerations also arise when working with viruses, particularly those with a pandemic potential. It is essential to ensure that research involving these pathogens is conducted in a responsible and ethical manner with appropriate biosafety measures in place (Oka et al., 2011).

Future research directions in this field include the development of more efficient and precise mutagenesis techniques, exploration of novel drug targets within viral glycoproteins, and studying the role of viral glycoproteins in immune evasion and pathogenesis. Additionally, greater emphasis should be placed on understanding the interactions between viral glycoproteins and host cell factors as these interactions are crucial for viral entry, replication, and dissemination. By addressing these challenges and pursuing research directions, scientists can continue to make significant advancements in our understanding of viral glycoproteins and develop innovative therapeutic strategies for combating viral infections (Liun and Naismith, 2008).

CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare. All co-authors have seen and agreed with the contents of the manuscript. There are no financial interests to disclose. We certify that the submission is an original work and is not under review at any other publication.

DISCLOSURE

The authors did not receive any form of commercial support, either in the form of compensation or financial assistance, for this case report. The authors have no financial interest in any of the products, devices, or drugs mentioned in this article.

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