Molecular pathology selected abstracts

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Ability of glioblastoma mutations to alter EGFR dimers to prevent ligand bias

April 2022—Mutations of the epidermal growth factor receptor gene are common in the aggressive brain tumor glioblastoma multiforme and occur in approximately a quarter of all such cases. However, unlike *EGFR* mutations in lung cancer, which typically occur in the intracellular portion of the EGFR protein (intracellular tyrosine kinase domain), *EGFR* mutations in glioblastoma multiforme tend to occur in the extracellular portion of the protein. Furthermore, the role of extracellular *EGFR* mutations in glioblastoma multiforme is less clear than that of *EGFR* mutations in lung cancer, in which the gene serves as an important oncogenic driver alteration. The authors published a detailed analysis of glioblastoma multiforme-specific *EGFR* mutations, focusing on how the mutations contribute to disease progression. They compared ligands, such as EGF, that bind with high affinity to the extracellular domain of the EGFR protein to ligands that have a low affinity for binding to EGFR, such as epiregulin (EREG), amphiregulin (AREG), and epithelial mitogen (EPGN). The authors discovered that common mutations, such as R84K and A265V, in the extracellular portion of EGFR in glioblastoma multiforme can change the strength with which various types of ligand molecules bind and activate this receptor. For example, extracellular ligands that normally bind very weakly to the normal EGFR receptor (including EREG, AREG, and EPGN) can show as much as 10-fold higher binding to the mutated receptor, thereby inappropriately triggering signals for cell growth that likely contribute to the glial cells in the brain becoming rapidly growing tumor cells. Furthermore, the authors' results reveal additional mechanisms of activation of mutant *EGFR* in glioblastoma multiforme. Dimerization of EGFR refers to two molecules of EGFR binding to each other. The authors demonstrated in in vitro experiments that mutation-induced changes in the shape of EGFR strengthen the formation of EGFR dimers by several hundredfold. This study explains the mechanism of EGFR activation in glioblastoma multiforme. Specifically, the authors show that mutations in the extracellular domain of EGFR allow low-affinity ligands, such as EREG, and high-affinity ligands, such as EGF, to impact signaling similarly. Therefore, these extracellular *EGFR* mutations in glioblastoma multiforme appear to thwart the ability of EGFR to distinguish between high-affinity and low-affinity ligands. Dysregulation of signaling mediated by low-affinity ligands, such as EREG, may disrupt the normal differentiation of progenitor cells, thereby promoting the development of glioblastoma multiforme. The authors' findings may contribute to the development of glioblastoma multiforme-specific EGFR-targeted therapeutics in the future.

Hu C, Leche CA II, Kiyatkin A, et al. Glioblastoma mutations alter EGFR dimer structure to prevent ligand bias. *Nature*. 2022;602:518–522. doi: 10.1038/s41586-021-04393-3.

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Impact of prior influenza virus infection on response to influenza vaccination

A major challenge in developing vaccines that target such RNA viruses as influenza and SARS-CoV-2 is antigenic drift. Each cycle of viral RNA replication may lead to mutations in the RNA genetic code that are passed on to successive generations of viruses. Therefore, components of a newer strain of an influenza virus, such as the major cell surface protein hemagglutinin, may differ from those of older strains of the same virus. Because influenza vaccines target specific viral proteins, including hemagglutinin, they may lose their efficacy against newer strains

of a virus that have undergone antigenic drift. The authors conducted a study to determine the impact of the influenza virus A(H3N2) strain on immunity that had been induced by prior vaccination. The study focused on a cohort of adults from Northern Vietnam who had (72 participants) or did not have (28 participants) documented influenza A(H3N2) infection during the preceding nine years. Response to influenza vaccination was determined by serially assessing the production of antiviral proteins, or antibodies, by immune cells in response to 40 influenza A(H3N2) strains circulating in humans from 1968 (when the strain emerged) to 2018. Influenza vaccine-induced antibodies may be generated by induction or activation of naïve immune cells (B cells) that have not been previously exposed to the virus or by further maturation of immune cells (memory B cells) that have been previously exposed to the virus. The results of this study suggest that memory B cells may outperform naïve B cells in mounting an effective immune response against a new influenza A(H3N2) virus strain. Antibody levels against the influenza A(H3N2) strain rose substantially within seven days of vaccinating study participants. This is the opposite of what is typically observed in young children who previously have not been exposed to the influenza virus. The authors documented several observations that demonstrate how recent infection with influenza positively affects immunity. For instance, vaccine-induced antibody levels were significantly higher in participants who recently had been infected with influenza, and vaccine-induced antibodies frequently targeted portions of viruses, or epitopes, that had circulated between 2013 and 2015 versus epitopes from strains that had circulated between 2009 and 2012. More importantly, among vaccinated study participants, positive cases were disproportionately higher for people who were not recently infected than for those who were recently infected. These findings are critical for refining future vaccine strategies to counter new strains of the influenza virus that may exhibit significant antigenic drift. Hypothetically, these results also may be valid for other RNA viruses that exhibit antigenic drift, such as SARS-CoV-2, and, therefore, require additional study.

Auladell M, Phuong HVM, Mai LTQ, et al. Influenza virus infection history shapes antibody responses to influenza vaccination. *Nat Med*. 2022. doi: 10.1038/s41591-022-01690-w.

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