REVOLUTIONIZING BIOMEDICAL RESEARCH: INTEGRATING CUTTING-EDGE AI/ML TO UNLEASH INNOVATION IN DRUG DISCOVERY AND THERAPEUTICS DEVELOPMENT

Discovery Of Non-canonical Protein Targets Of Fentanyl Across Tissues From Animal Models And Humans Using Photoaffinity Fentanyl Probes

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Synthetic opioids such as fentanyl and related analogs have been widely used for pain management. However, their negative side effects, including constipation, respiratory depression, and high potential for addiction underscore the need for a deeper understanding of fentanyl's interactions with various cellular receptors throughout the body. Fentanyl analogs bind and activate opioid receptors in the central and peripheral nervous systems, triggering a variety of downstream signaling pathways. Increasingly, fentanyl has been shown to interact with non-opioid receptors, and elucidation of these non-canonical fentanyl-protein interactions may provide insight into the mechanisms contributing to fentanyl's adverse effects and inform future drug designs or medical countermeasures. To identify proteins in mammalian tissues that may interact with fentanyl, we designed and synthesized three fentanyl photoaffinity probes for labeling and enrichment of fentanyl-binding proteins in tissues from various animal models. Protein targets were identified through bottom-up proteomics. Molecular docking simulations of protein target structures with fentanyl were used to assess these binding interactions computationally and prioritize proteins for biochemical validation. We performed protein expression, biostructural analyses, and biochemical assays to confirm the interaction of fentanyl with specific protein targets in vitro. Further study of these non-opioid receptor proteins and how fentanyl perturbs their functions will improve our understanding of affected biochemical pathways in the body and potential approaches to mitigate fentanyl's harmful effects.

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