COMPUTER-AIDED LEAD MOLECULE IDENTIFICATION: SOME CASE STUDIES FOR MALARIA & NEURODEGENERATIVE DISEASES

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Computer-aided lead molecule identification: Some case studies for Malaria & Neurodegenerative Diseases

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Certificate

This is to certify that the thesis entitled, "**Computer-aided lead molecule identification: Some case studies for Malaria and Neurodegenerative Diseases**", being submitted by **Mr. Ashutosh Shandilya** to the Indian Institute of Technology, Delhi for the award of the degree of **Doctor of Philosophy** in Chemistry is a record of bonafide research work carried out by him. Ashutosh Shandilya has worked under my guidance and supervision and has fulfilled the requirements for the submission of this thesis, which to my knowledge has reached the requisite standard.

The results contained in this dissertation have not been submitted in part or full to any other University or Institute for the award of any degree or diploma.

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Abstract

This thesis focuses on identification of potential lead molecules against Malaria and neurodegenerative diseases providing atomic level computational insights on the mode of action of these molecules. A few novel potential lead molecules have been identified against malarial and neurodegenerative disease targets using computational approach. These designed molecules were further assayed experimentally and found to be in good conformity with computational predictions.

The thesis is divided into seven chapters. Chapter 1 discusses the current status of computer-aided drug design in general and its role in drug discovery process. A brief overview of the current state of research on malaria and neurodegenerative diseases has been reported as well.

Chapter 2 is devoted to identifying a plausible mechanism of action artemisinin, a widely used antimalarial, via exhaustive computational approaches. This chapter also includes a justification and modeling of the target enzyme and elucidates atomistic level interaction between ligand and target enzyme. Chapter 3 and Chapter 4 presents identification of some novel hit molecules against phosphoethanolamine N-methyl transferase enzyme (*PfPMT*). A *de novo* drug design strategy is adopted to target *PfPMT*. Designed molecules were synthesized and assayed against malarial cell lines in collaboration. Further *PfPMT* enzymes were expressed and purified. Isothermal titration calorimetry assays were carried out to understand the binding kinetics of ligands and protein. Cell based studies (schizont maturation inhibition assay) were carried to identify best inhibitors against

malaria. These molecules showed low nanomolar activities against malarial cell lines.

Chapter 5 focuses on identification of novel molecules against Alzheimer's disease. Calcium-calmodulin dependent kinase IV (CAMK4) enzyme was targeted for the treatment of Alzheimer's disease. Binding of curcumin and pyrimidine based derivatives were identified as good binders, and their modes of binding were also elucidated. Acetyl cholinesterase (ACh) is a widely known target for the structure based drug designing because of its direct association with Alzheimer's disease. A few triazine hybrid molecules were designed using computational approaches. Experimentally, they were found to be show good activity against Ach in low nanomolar range.

Chapter 6 discusses a detailed theoretical account that is substantiated with some new experimental investigations on the molecular origins of the differential affinities of iminocyclitols with various glycosidases. These newly designed compounds show interesting selectivity towards the target enzymes. One of the designed molecules is shown to be a stabilizer of α-galactosidase enzyme indicative of its chaperon activity. Finally in chapter 7, a summary and some perspectives emerging from the thesis work are discussed.

सारांश

यह शोधकार्य संगणकीय अन्तर्दृष्टि द्वारा आणविक पारस्परिक क्रिया को समझकर मलेरिया एवं न्यूरो डिजेनेरेटिव रोगो के संभावित दवाओं के पहचान पर केंद्रित है. संगणकीय दृष्टिकोण को ध्यान में रखकर मलेरिया एवं न्यूरो डिजेनेरेटिव रोगो के विरुद्ध कुछ नवीन एवं महत्वपूर्ण यौगिकों का डिज़ाइन किया गया है. इन नवीन डिज़ाइन यौगिकों को प्रयोगात्मक रूप से भी परखा गया और इनका संगणकीय पूर्वानुमान के साथ अच्छा अनुरूप पाया गया.

यह शोधकार्य सात अध्याय में बटा हुआ है. पहला अध्याय सामान्य रूप से संगणकीय सहायता प्राप्त दवा के डिज़ाइन की स्थिति और दवा के खोज में इसकी भूमिका के चर्चा करता है. इसके साथ-साथ मलेरिया एवं डिजेनेरेटिव रोगो पर अनुसन्धान की वर्तमान स्थिति का भी संक्षिप्त अवलोकन किया गया है. अध्याय २ मलेरिया के लिए व्यापक रूप से उपयोग में आने वाली दवा आर्टीमिसिनिन के संभावित कार्रवाई की विधि के आंकलन में समर्पित है. यह अध्याय लक्षित एंजाइम एवं आर्टीमिसिनिन के बीच आणविक पारस्परिक क्रिया को भी स्पष्ट करता हैं.

अध्याय ३ एवं ४ में *डि-नोवो* ड्रग डिज़ाइन नीति के तहत, एन-मिथाइल ट्रांस्फ़ेरेज़ (*PfPMT*) एंजाइम के विरुद्ध कुछ नवीन यौगिकों को डिज़ाइन किया गया है. तत्पश्चात इन डिजाइन्ड यौगिकों को संश्लेषित कर मलेरिया कोशिका परत पर परीक्षण किया गया. लिगेंड एवं एंजाइम के बाध्यकारी कैनेटीक्स का आंकलन करने के लिए समतापी कैलोरीमेट्री अनुमापन भी किया गया. इन डिज़ाइन यौगिकों में से कुछ यौगिक मलेरिया कोशिका एवं लक्षित एंजाइम के विरूद्ध नैनो-मोलर स्तर तक की बाध्यता दिखाई.

अध्याय ५ अल्ज़ाइमर रोग के विरुद्ध कुछ नवीन यौगिकों के पहचान पर केंद्रित है. अल्ज़ाइमर रोग के उपचार हेतु *CAMK4* एंजाइम को लक्षित किया गया. इसके लिए करक्यूमिन एवं पिरिमिडीन आधारित व्युत्पन्न यौगिकों को अच्छे बाइंडर के रूप में पहचान की गयी एवं उसके बाध्यकारी तंत्र को भी स्पष्ट किया गया. अल्ज़ाइमर रोग के लिए एसिटील कोलिन्स्टरेज (ACh) एक व्यापक रूप से ज्ञात लक्षित एंजाइम है. इस रोग के उपचार हेतु सगणकीय दृष्टिकोण से कुछ नवीन त्रिट्रिएज़िन (Triazine) संकरण आधारित नवीन यौगिक का डिज़ाइन किया गया. प्रयोगात्मक रूप में भी ACh के खिलाफ़ इन यौगिक ने नैनो-मोलर श्रेणी में बाध्यता दिखाई.

अध्याय ६ इमिनोसिक्लिटॉल्स (Iminocycltiols) की विभिन्न ग्लाइकोसिडेज़ के साथ विस्तृत सैद्धांतिक चर्चा करता है. यहाँ नवीन डिज़ाइन किये गए यौगिक लक्षित एंजाइम के प्रति दिलचस्प चयनात्मकता दिखते हैं. डिज़ाइन किये गए यौगिकों में से एक यौगिक अल्फा-गलैक्टोसिडेज़ (α- galactosidase) स्थिरकारी के रूप में इंगित होता है. अंततः अध्याय ७ इस शोधकार्य से उभर कर आये कुछ दृष्टिकोण एवं सारांश पर केंद्रित है.

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