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Analysis of Multi Target Directed Non Peptidic Compounds as Potential Malaria Inhibitors against Plasmodium Proteases through Structure Activity Relationship and Virtual Screening Workflow

Babalola S Adewale*

Department of Pharmaceutical chemistry, ahmadu bello university, Zaira, Kaduna state, Nigeria

***Corresponding author:** Babalola S Adewale, Department of IT Engineering, Dezful Branch, Islamic Azad University, Dezful, Iran, E-mail: Hoseinpourmeghdad@gmail.com

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Abstract

Purpose: To design new antimalarial agents from both active synthetic and natural products fragments using in-silico software to determine their pharmacokinetics and pharmacodynamics profiles as therapeutic molecules against the deadly malaria causing parasite P. falciparum biological targets.

Methods: In this study, active fragments from crenelated and quinolinyl chaconnes with known antimalarial activities were hybridized through molecular hybridization. Four P. falciparum enzymes including plasmepsin II, Falcipain 2, Falcipain 3, and SUB1 which have been reported to be involved in malaria transmission are used as the biological targets for the ligands (prenylated-quinolinyl chalcones hybrids) during docking simulation. Receptor-ligand complexes were viewed. Web-based softwares (Moftsoft, SwissADME, AdmetSAR 1 and 2) were employed in drug-likeness and ADMET prediction.

Results: Hybridization of the active fragments resulted to a novel scaffold including 126 novel prenylated-quinolinyl chaconnes. Post-docking analysis revealed strong interactions of the compounds with the targets used. At least 25 of the compounds have high affinities for the targets.

Conclusion: The compounds exhibited plausible pharmacokinetics and The selected compounds demonstrated good drug-likeness and ADMET properties.

Keywords: Prenylated chalcones; Quinolinyl chalcones; SAR; Docking simulation; Molecular hybridization; Plasmodium falciparum; Antimalaria agents

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Introduction

Malaria remains one of the most prevalent diseases claiming more than 400,000 lives annually across the globe. Children and pregnant women are most vulnerable to this disease. Plasmodium falciparum remain the most dreadful malaria parasite which poses resistance with the current first-line antimalarial drugs. Scientists across the world are constantly searching for new principles to combat the emerging parasite strain. Several years of drug discovery and development of novel antimalarial drugs has led to increasing focus on elimination and eradication of malaria. In P. falciparum, the papain-family cysteine proteases falcipain-2 (FP-2) and falcipain-3 (FP-3) are known to catalyze the proteolysis of host hemoglobin, a process that is essential for the development of erythrocytic parasites.

Hemoglobin is degraded by a series of proteases in an acidic digestive vacuole of the parasite. The aspartic proteases found in the vacuole, named plasmepsin, make an initial attack on the hemoglobin molecule, followed by proteolysis of the large fragments into small peptides by a cysteine protease named falcipain. The initial cleavage by plasmepsin is critical to hemoglobin degradation because intact hemoglobin cannot be cleaved by falcipain unless it is first denatured. It is believed that this cleavage results in the unraveling of the hemoglobin molecule, allowing further proteolysis to proceed efficiently. Subtilisin-like serine protease called SUB1 is an enzyme which have been reported to promote egress. Minutes before egress, a calcium and cGMP-dependent signal triggers the discharge of a SUB1 from specialized secretory organelles of the still intracellular merozoite, called exonemes. On its release into the Parasitophorous Vacuole lumen, SUB1 proteolytically modifies a number of abundant merozoite surface and Parasitophorous Vacuole resident soluble proteins including SERA6, a putative cysteine protease involved

in egress and invasion. Pharmacological inhibition of SUB1 discharge or catalytic activity blocks egress and/or reduces the invasive capacity of released merozoites. Drugs that target SUB1 activity should prevent disease progression and so have potential as a new class of antimalarial therapeutics, urgently needed in response to increasing drug resistance of the parasite. Scientists across the world are constantly searching for new principles to combat the emerging parasite resistant strains. Several years of drug discovery and development of novel antimalarial drugs has led to increasing focus on elimination and eradication of malaria. Since there is currently no licensed malaria vaccine available to make people immune, chemotherapy still offers the best solutions. Novel strategies in antimalarial treatment include the optimization of therapies with available drugs (e.g. artemisinin combination therapy), repurposing of medicines, developing analogs of existing drugs and the evaluation and use of chemosensitizers (drug-resistance reversers). There is, however, another strategy that has gained much attention in the field of contemporary medicinal chemistry. It involves the combination of two biologically active molecules (pharmacophores) into one single hybrid entity with a dual mode of action. These novel hybrid molecules have the potential to enhance efficacy, improve safety, be cost-effective and reduce the propensity to elicit resistance relative to the parent drugs. Hybridization concept has been proved as a lead for developing new antimalarial active agents. Drug discovery and development is a very complex and costly endeavor, which includes disease selection, target identification, and validation, lead discovery and optimization, preclinical and clinical trials. With the development of in silico methods in recent years, the number of new molecular entities (NMEs) approved by the U.S. Food and Drug Administration (FDA) has risen obviously.

However, there are still lots of drug candidates that fail to become drugs. Lack of efficacy and safety are the two major causes leading to drug failure which means the absorption, distribution, metabolism, excretion, and toxicity (ADMET) properties of chemicals play vital roles in every stage of drug discovery and development. Therefore, it is necessary to find efficacious molecules with better ADMET properties.

In this study, SAR guided design of six series of the novel scaffold of chalcone by extensive SAR survey of actively known quinolinyl chalcones and prenylated chalcones from the literature, molecular hybridization of the pharmacophoric fragments resulted to 126 novel prenylated-quinolinyl chalcone hybrids. Molecular docking pharmacodynamics studies were also conducted.

Methods

n-silico drug-likeness evaluation

The fragments were evaluated using rule of three (RO3) and are then hybridized selected compounds with best affinity for the receptors from the library were further analyzed on the basis of Lipinski's rule of five (RO5) Vebers number of rotatable bonds (NOR and polar surface area (PSA) and other physicochemical properties on Swiss ADME server. Drug scores were calculated using ORISIS property explorer and the drug-likeness scores were calculated using molsoft web-based calculator.

Vascular specialists can treat vascular structure conditions with current development. For example: Discouraged veins can be treated with little inflatable's to open the upset zone. A stent inside the conductor maintains its new broadened size. Bunch dissolving experts quicken ejection of blood groups. Stents covered with extraordinary material impersonating a fake course can be inserted inside an aneurysm to seal it and redirect the circulation system.

In-silico absorption, distribution, metabolism, elimination, and toxicity (ADMET studies

In-silico ADMET studies of selected compounds perform with web-based software AdmetSAR1 and Admet SAR2 and are validated using Swiss ADME scores Absorption parameters calculated were human intestinal absorption (HIA), caco-2 permeability. Distribution parameters calculated was plasma protein binding (PPB), p-glycoprotein substrate inhibitor, blood-brain barrier penetration. For metabolism CYP inhibitory promiscuity, cytochrome P450 (CYP450) substrate inhibitor Substrate: CYP2C9, 2D6, 3A4 Inhibitor: CYP1A2, 2D6, 2C9, 2C19, 3A4 were the parameters calculated. Human either-a-go-go-related gene inhibition, oral acute toxicity, AMES toxicity, carcinogenesis, fish toxicity, tetrahymena pyriformis toxicity, honey bee toxicity, and biodegradation were the parameters examined.

Docking simulation

Both the fragments and the designed hybrids were drawn using Marvin JS Chemsketch. The drawn hybrid structures were later optimized to 3D in PDB format with the same software. Four malarial biological targets were used for the docking study; Pf Plasmepsin II, Pf Falcipain 2, Pf Falcipain 3 and Pf SUB1. The 3D crystal structures of the targets were retrieved from Protein Data Bank, Plasmepsin II, Falcipain 2, Falcipain 3. Both the hybrids and receptors were prepared for docking using UCSF Chimera docking software. Incomplete side chains of the target molecules were replaced using the Dunbrack rotamer library, and the charges were computed using antechamber. The prepared ligands and receptors were converted to PDBQT format and the co-ordinates of the active sites of the receptors were deduced using grid box using Autodock tool 1.5.6. Subsequently, the ligands were docked on each receptor through virtual screening with bash command using Autodock Vina in cygwin. The bindings were visualized using discovery studio visualizer (**Table 1**).

MW; Molecular Weight	NORT: Number of rotatable bonds HA: Heavy atoms
logP: Partition co-efficient	MR: Molar refractivity
logS: Aqeous solubility	AA: Aromatic atoms
HBA: Hydrogen bond acceptor	PSA:Polar surface area
HBD: Hydrogen bond donor	RO5: Rule of five

Insilico admet prediction

There is, however, another strategy that has gained much attention in the field of contemporary medicinal chemistry. It

involves the combination of two biologically active molecules (pharmacophores) into one single hybrid entity with a dual mode of action. These novel hybrid molecules have the potential to enhance efficacy, improve safety, be cost-effective and reduce the propensity to elicit resistance relative to the parent drugs. Hybridization concept has been proved as a lead for developing new antimalarial active agents. Drug discovery and development is a very complex and costly endeavor, which includes disease selection, target identification, and validation, lead discovery and optimization, preclinical and clinical trials. With the development of in silico methods in recent years, the number of new molecular entities (NMEs) approved by the U.S. Food and Drug Administration (FDA) has risen obviously.

Rule of Three (RO3) has been useful in ensuring that fragment libraries really do consist of compounds with active fragment-like properties. The parameters of 'Rule of Three' are such that a molecule with molecular weight ≤ 300, the number of hydrogen bond donors is ≤ 3, the number of hydrogen bond acceptors is ≤ 3, ClogP is ≤ 3, NROT (≤ 3) and PSA (≤ 60) would be an active fragment. Any fragment that passes these rules on the average could be useful when constructing fragment libraries for efficient lead discovery. The RO3 was applied to the fragment library to assess whether our derived active fragments possess the parameters to exhibit active fragment-like properties. All the fragments passed the RO3 as indicated, fragments 5, 6, 7, 11, 13, 15, 16, and 20 passes the RO3 only the on the average.

Six series of a novel scaffold comprising of 126 hybrids resulted from the hybridization of the screened fragments as listed above. The best twenty five hybrids with high affinity for at least two biological targets were selected from the docking score ranking (unpublished). Nineteen of these compounds have higher affinities for three receptors (Falcipain 2, Falcipain 3, and SUB1) compared to the respective receptors' natural inhibitors.

The binding interactions as viewed on Discovery studio visualizer show that all the selected compounds have good poses on the receptors. While compounds 19d and 21d had five and seven hydrogen bonds respectively with plasmepsin II residues in addition to other hydrophobic bonds which is in line with their low binding energy.

Compounds 13d, 15d, and 21d had three hydrogen bonds each with the Falcipain 2 residues in addition to other hydrophobic interactions. With Falcipain 3, compounds 13d had five hydrogen bonds in addition to other hydrophobic interactions whereas compounds 4b, 4d, 7c, 8c, 11d, 13a, 18d, and 21d had three hydrogen bonds each with the receptor residues.

Compounds 7c and 11d had six and four hydrogen bonds respectively with SUB1 residues in addition to other hydrophobic interactions. From the molecular simulation results of the hybrids, it stems out that compounds having the quinoline fragment with additional hydroxyl group at position 8 (fragment d) shows a great binding affinity for all the receptors. This is in accordance to the (fragment d) high ranking in the fragments binding energy ranking (unpublished). This is indicating its potential as possible nucleus for new antimalarial agents (**Figure 1**).

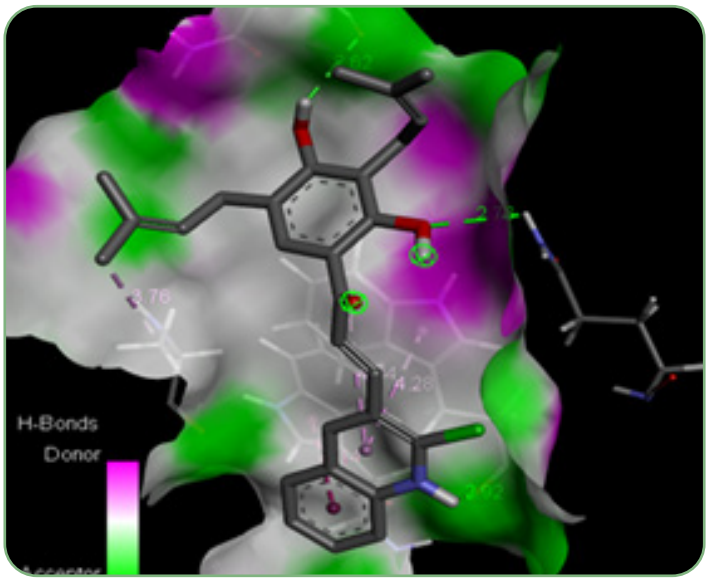

Figure 1: Binding interaction of 4b with falcipain in 3D.

Results and Discussion

Accounted for the development of orally bioavailable drug candidates. The rule of five depicted the relationship between bioavailability and physicochemical properties, settling the concept of drug likeness. According to the Rule of Five, a molecule would not be orally active if it violates two or more of the four rules. These rules have been improved by others, such as veber and co-workers. Who discovered that the number of rotatable bonds (NROT) is an essential parameter, a maximum of seven as optimal for oral bioavailability? Ghose and co-worker's molar refractivity (MR≤130) and the total number of atoms between 20 and 70 literature also indicates that polar surface area (PSA) is another important property passively absorbed molecules with a PSA of 110–140 Å2 are thought to have low oral bio-availabilities. An optimal range of each property for bioavailability of a drug-like molecule. All the selected compounds are in accordance with Lipinski's Rule of Five for oral bioavailability and permeability. However, compounds 4b, 10e, 13b, and 13f showed higher lipophilicity scores 4.20, 4.93, 4.59, 4.21 respectively than the marked score (MlogP≤4.15). These compounds also met optimal conditions for bioavailability. All of the compounds fulfilled Ghose's total number of atoms, and all of the them are in accordance with the molar refractivity score (MR≤130) except 4a, 4b, 4d, and 4e with a margin increase 133.04, 138.05, 135.06, and 133.04 respectively. The drug-likeness score depicts the structural similarity features of the compounds with the existing marketed drugs using Molsoft's chemical fingerprints. The training set for this model consisted of five thousand of marketed drugs from WDI (positives) ten thousand of carefully selected non-drug compounds (negatives). All the compounds exhibit good drug-likeness score except for compounds 10e and 21a with 0.58 and 0.10 respectively. During the time and resource-consuming processes of drug discovery and development, enormous number of molecular compounds is evaluated using diverse parameters in order to determine the selection of which chemicals to synthetize, test and promote, with the sole aim of identifying

those with the best likelihood of becoming an effective medicine for the patients. The molecules must show high biological activity together with low toxicity. Chemical absorption, distribution, metabolism, excretion, and toxicity (ADMET), play key roles in drug discovery and development. A good drug candidate should not only have effective efficacy against the therapeutic target, but also show teeming ADMET properties at a therapeutic dose In silico ADMET analysis was conducted to further predict the pharmacokinetics and physico-dynamic profile of the selected compounds. The permeability glycoprotein P-gp is essential to allowing active efflux through biological membranes, for instance from the gastrointestinal wall to the lumen or from the brain. Specifically, P-gp functions to protect the central nervous system (CNS) from xenobiotics. Therefore, it is important to determine whether a molecule is an agonist or antagonist of this glycoprotein. Cytochromes P450 (CYP) is a group of isoenzymes responsible for drug elimination through metabolic biotransformation. It has been reported that CYP and P-gp can transform small molecules synergistically to increase protection of tissues and organisms. It has been estimated that 50 to 90% of drug molecules are agonist of five major isoforms (depending on the author) (CYP1A2, CYP2C19, CYP2C9, CYP2D6, CYP3A4).

Conclusion

Overall results have proven that selected compounds with the best affinities for the selected malaria biological targets (PlmII, Falcipain 2 and 3, and SUB1) exhibited plausible drug-able properties which raise them to be possible antimalarial drug candidates.

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References

1 Macintyre F, Ramachandruni H, Burrows JN, Holm R, Thomas A Sajid M et al (2018) Injectable anti-malarials revisited: discovery and development of new agents to protect against malaria. Malar J. 17: 1-8.

2 White NJ, Duong TT, Uthaisin C, Nosten F, Phyo AP Sajid M et al (2016) Antimalarial activity of KAF156 in falciparum and vivax malaria. N Engl J Med. 375: 1152-1160.

3 Kerr ID, Lee JH, Farady CJ, Marion R, Rickert M, Sajid M (2009). Vinyl sulfones as antiparasitic agents and a structural basis for drug design. J Biol Chem. 284: 25697-25703.

4 Chen X, Aledia AS, Popson SA, Him L, Hughes CC, George SC (2010) Rapid anastomosis of endothelial progenitor cell–derived vessels with host vasculature is promoted by a high density of cotransplanted fibroblasts. Tis Eng 16: 585-594.

5 Asojo OA, Gulnik SV, Afonina E, Yu B, Ellman JA, Haque TS et al (2003) Novel uncomplexed and complexed structures of plasmepsin II, an aspartic protease from Plasmodium falciparum. J Mol Biol. 327:173-81.

6 Withers-Martinez C, Strath M, Hackett F, Haire LF, Howell SA, Walker PA et al (2014). The malaria parasite egress protease SUB1 is a calcium-dependent redox switch subtilisin. Nature communications. 5: 1-1.

7 Agarwal S, Singh MK, Garg S, Chitnis CE, Singh S (2013) Ca2+ mediated exocytosis of subtilisin like protease 1: a key step in egress of plasmodium falciparum merozoites. Cell Micro Biol. 15: 910-921.

8 Koussis K, Withers-Martinez C, Yeoh S, Child M, Hackett F, Knuepfer E et al (2009) A multifunctional serine protease primes the malaria parasite for red blood cell invasion. EMBO J. 28: 725-735.

9 Ruecker A, Shea M, Hackett F, Suarez C, Hirst EM, Milutinovic K et al (2012) Proteolytic activation of the essential parasitophorous vacuole cysteine protease SERA6 accompanies malaria parasite egress from its host erythrocyte. J Biol Chem. 287: 37949-37963.

10 Thomas JA, Tan MS, Bisson C, Borg A, Umrekar TR, Hackett F et al (2018) A protease cascade regulates release of the human malaria parasite Plasmodium falciparum from host red blood cells. Nature microbiology. 2018 Apr;3(4):447-55.

11 Schlitzer M (2008) Antimalarial drugs–what is in use and what is in the pipeline Archiv der Pharmazie: Int J Pharm Med Chem. 341: 149-163.

12 Builders MI (2018) In search of a potent antimalarial agent: Antiplasmodial assessment of three herbs with folkloric antimalarial claims. Int J Biomedi Clini Scie. 3: 40-46.

13 Walsh JJ, Bell A (2009) Hybrid drugs for malaria. Curr Pharm Des. 15: 2970-2985.

14 Muregi FW, Ishih A (2010) Next-generation antimalarial drugs: hybrid molecules as a new strategy in drug design. Dru Develop Res. 71: 20-32.

15 Menezes CM, Anna CM, Rodrigues CR, Barreiro EJ (2002) Molecular modeling of novel 1H-pyrazolo [3, 4-b] pyridine derivatives designed as isosters of the antimalarial mefloquine. J Mol Str. 579: 31-9.