



HAL
open science

Novel Benzotriazole- β -lactam Derivatives as Antimalarial Agents: Design, Synthesis, Biological Evaluation and Molecular Docking Studies

Malihe Aye, Aliasghar Jarrahpour, Zahra Haghighijoo, Roghayeh Heiran, Roya Pournajati, Hamid Reza Karbalaei-Heidari, Veronique Sinou, Jean Michel Brunel, Mehmet Akkurt, Namık Özdemir, et al.

► To cite this version:

Malihe Aye, Aliasghar Jarrahpour, Zahra Haghighijoo, Roghayeh Heiran, Roya Pournajati, et al.. Novel Benzotriazole- β -lactam Derivatives as Antimalarial Agents: Design, Synthesis, Biological Evaluation and Molecular Docking Studies. *Chemistry and Biodiversity*, 2024, 21 (2), pp.e202301745. 10.1002/cbdv.202301745 . hal-04739635

HAL Id: hal-04739635

<https://hal.science/hal-04739635v1>

Submitted on 16 Oct 2024

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Novel Benzotriazole- β -lactam Derivatives as Antimalarial Agents: Design, Synthesis, Biological Evaluation and Molecular Docking Studies

Dr. Malihe Aye,^{a,b} Prof. Aliasghar Jarrahpour,^{a*} Dr. Zahra Haghijoo,^c Dr. Roghayeh Heiran,^{d,*} Dr. Roya Pournajati,^e Dr. Hamid Reza Karbalaee-Heidari,^e Dr. Veronique Sinou,^f Prof. Jean Michel Brunel,^f Prof. Mehmet Akkurt,^g Dr. Namik Özdemir,^h Prof. Edward Turos,ⁱ

^a Malihe Aye Department of Chemistry, College of Sciences, Shiraz University, Shiraz, 71946-84795 Iran (jarahpor@shirazu.ac.ir; aliasghar6683@yahoo.com)

^b Department of Civil and Environmental Engineering, Shiraz University, Shiraz, Iran

^c Department of pharmacology and Toxicology, University of Texas Medical Branch, Galveston, TX 77555, USA.

^d Estahban Higher Education Center- Shiraz University, Estahban, Iran (somaheheiran@gmail.com, r.heiran@saadi.shirazu.ac.ir)

^e Department of Biology, College of Sciences, Shiraz University, PO Box: 71467-13565, Shiraz 71454, Iran.

^f Aix Marseille Univ, INSERM, SSA, MCT, Faculté de Pharmacie, 27 bd Jean Moulin, 13385, Marseille, France.

^g Department of Physics, Faculty of Sciences, Erciyes University, 38039, Kayseri, Turkey.

^h Division of Physics Education, Department of Mathematics and Science Education, Faculty of Education, Ondokuz Mayıs University, TR-55139 Samsun, Turkey.

ⁱ Center for Molecular Diversity in Drug Design, Discovery, and Delivery, Department of Chemistry, CHE 207, 4202 East Fowler Avenue, University of South Florida, Tampa, FL 33620, USA.

Abstract: Many people around the world suffer from malaria, especially in tropical or subtropical regions. While malaria medications have shown success in treating malaria, there is still a problem with resistance to these drugs. Herein, we designed and synthesized some structurally novel benzotriazole- β -lactams using 2-(1H-benzotriazol-1-yl)acetic acid as a key intermediate. To synthesize the target molecules, the ketene-imine cycloaddition reaction was employed. First, the reaction of 1H-benzotriazol-1,2,3-triazole with 2-bromoacetic acid in aqueous sodium hydroxide yielded 2-(1H-benzotriazol-1-yl)acetic acid. Then, the treatment of 2-(1H-benzotriazol-1,2,3-triazol-1-yl)acetic acid with tosyl chloride, triethyl amine, and Schiff base provided new β -lactams in good to moderate yields. The formation of all cycloadducts was confirmed by elemental analysis, FT-IR, NMR and mass spectral data. Moreover, X-ray crystallography was used to determine the relative stereochemistry of 4a compound. The in vitro antimalarial activity test was conducted for each compound against *P. falciparum* K1. The IC_{50} values ranged from 5.56 to 25.65 μ M. A cytotoxicity profile of the compounds at 200 μ M final concentration revealed suitable selectivity of the compounds for malaria treatment. Furthermore, the docking study was carried out for each compound into the *P. falciparum* dihydrofolate reductase enzyme (PFDHFR) binding site to analyze their possible binding orientation in the active site.

though medicines successfully treated malaria, resistance is a serious issue. Considering malaria's still high prevalence and mortality rates, as well as drug resistance to known antimalarials, new and effective drugs are The widely demonstrated importance of the biological properties of heterocyclic compounds has led organic chemists to develop efficient methods for synthesizing many derivatives for testing and pharmaceutical Standing out prominently among these are 2-azetidinones (β -lactams), which constitute an important class of as

Introduction

Malaria is considered a serious public health issue in tropical and subtropical regions, affecting both indigenous populations and tourists. There are about 100 countries around the world that still suffer from this infectious disease.^[1,2] Malaria cases worldwide increased from 245 million in 2020 to 247 million in 2021, with most cases occurring in the WHO African region.^[3] The infection is transmitted via the bite of Anopheles mosquitoes carrying *Plasmodium* parasites. *Plasmodium falciparum* (*P. falciparum*) causes the most fatal form of human malaria, among different species of *Plasmodium*.^[1,2] To control malaria effectively, scientists around the world are working on a variety of techniques, including medicines, insecticides, vaccinations, and bed nets sprayed with pesticides.^[4-7] Even

urgently needed.^[8]

development.^[9]

compounds, especially due to their biological activities ^[10-15] antimicrobial, ^[16-19] antifungal, ^[20] antimalarial, ^[5,21] anti-inflammatory,^[17,22] and anticancer ^[19,23-25] agents. Extensive research has been devoted to the synthesis of the β -lactam ring and further modification of ring substituents

a second large class of heterocycles with impressive biological capabilities.^[27] Triazoles are a five member aromatic heterocyclic scaffolds containing three nitrogens. The diversity of biological profiles of triazoles has made them indispensable in medicinal chemistry. They have developed as potential enzyme inhibitors,^{[28][29]} anti-Alzheimer,^[30] anticancer,^{[31][32]} antiviral,^[33] antifungal,^[34] antibacterial,^[35] antidiabetic agents,^[36] and etc.^[37] Several triazole-based hybrids and conjugates have demonstrated potential antimalarial activity.^{[38][39]} Molecular hybridization is a method for rationally designing various prototypes that recognizes pharmacophoric subunits in two or more known bioactive derivatives. Consequently, hybrid architectures can be developed that retain the potency of the original templates.^[40] The potential activity of triazole and β -lactam cores led researchers to design molecular hybrids namely triazole- β -lactams, which have demonstrated promising biological activities,^[41] including enzyme inhibitory,^[42] antimicrobial,^[43] antifungal,^[44] anticancer,^[24] antitubercular activities^[45], and etc. Moreover, these hybrids offer a new frontier in the malaria treatment. Kumar et al. have reported β -lactams having appended triazole rings of the C-3 center with promising antimalarial activity.^[23,46,47] They

design 3-(1,2,3-triazol-1-yl)- β -lactam (I), which showed good antiplasmodial activity against four strains of *P. falciparum*. The presence of a phenyl group on the triazole ring markedly improved the activity profiles of triazole-tethered β -lactam.^[23] Moreover, a series of ferrocenylchalcone- β -lactam conjugates (II) demonstrated promising activity against 3D7 and W2 strains of *P. falciparum*.^[46] β -lactam-1,2,3-Triazole-7-chloroquinoline bifunctional hybrids (III) were also synthesized and evaluated as potential antimalarial agents against W2 strain of *P. falciparum*.^[47] Moreover, our laboratory have reported on the antimalarial properties of certain 1,2,3-triazole- β -lactams (IV, V). These β -lactam-1,2,3-Triazole hybrids demonstrated moderate to excellent antimalarial properties against chloroquine resistant K1 strains of *P. Falciparum*,^[48] and K14 strains of *P. Falciparum*.^[49]

In the present study, we used hybridization strategy and designed some new benzotriazole- β -lactam derivatives through Staudinger reaction, that have promising antimalarial capabilities against the drug-resistant K1 strain of *P. falciparum* by combining the triazole and β -lactam scaffolds as in molecules I-V (Figure 1).^[46-48]

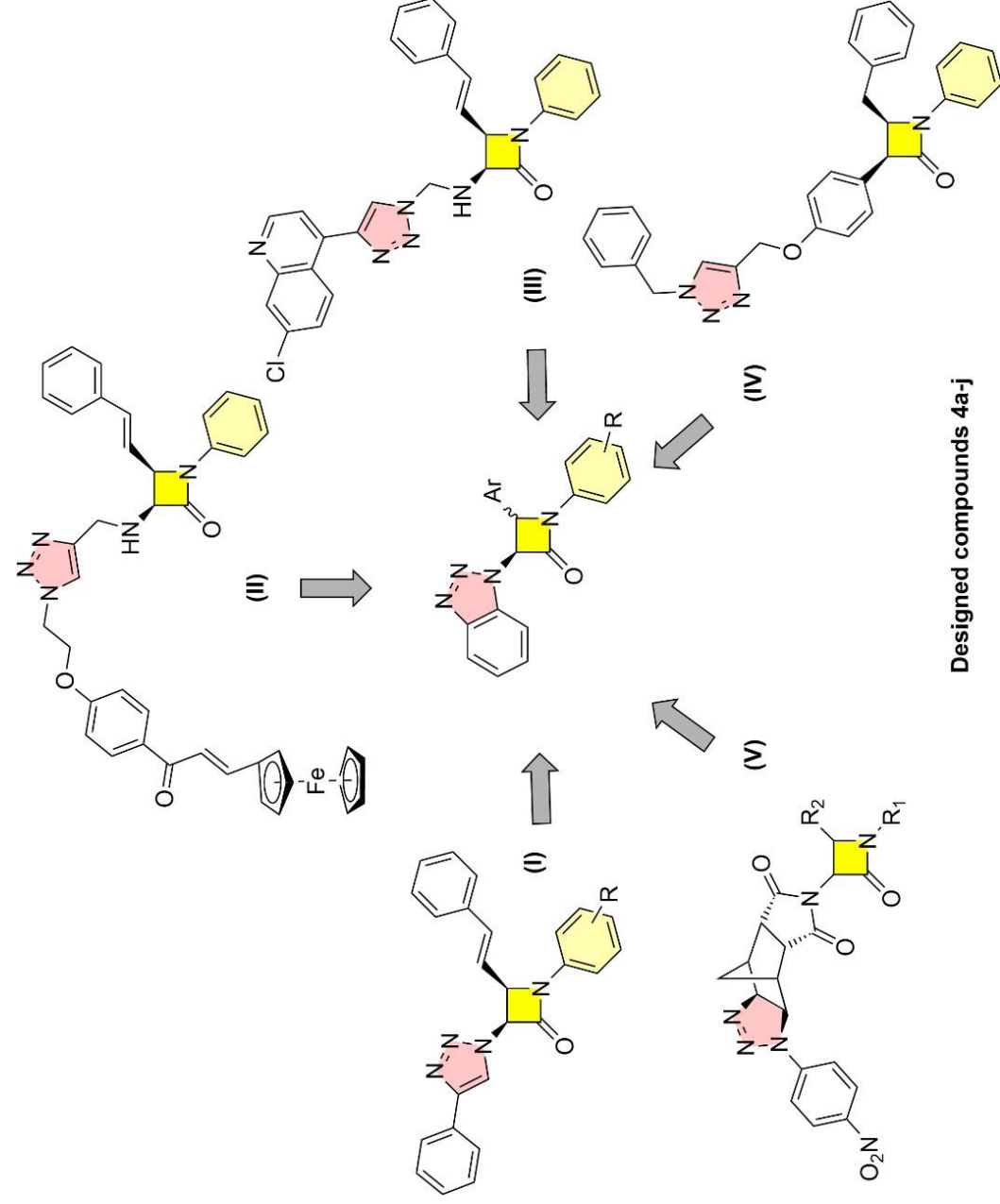


Figure 1. Analogue-based pharmacophore design of antimalarial benzotriazole- β -lactams **4a-j**.

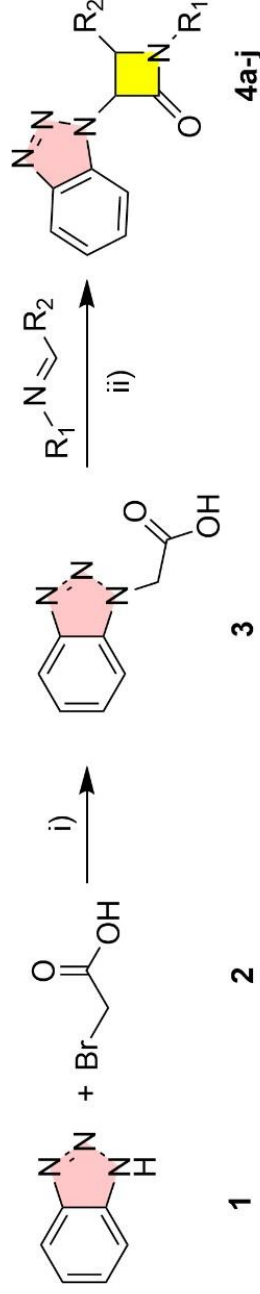
Derivative **4a** indicates normal bond lengths and angles comparable to those observed in related compounds.^{[51][52][53]} The β -lactam ring of **4a** is nearly planar [r.m.s deviation = 0.003 Å] and its mean plane makes dihedral angles of 2.00 (2), 78.69 (19) and 79.10 (2) ° with the phenyl ring, the naphthalene and 1*H*-benzotriazole ring systems, respectively. In the crystal of **4a** exists evidence of C \cdots H ... π and van der Waals interactions, generating a three-dimensional structure. ¹H NMR spectral analysis for **4a-j** demonstrates that the protons on the β -lactam ring appeared as two discrete doublets in the spectral region from 5.47-6.67 ppm. Under these experimental conditions, the *cis* and *trans* stereoisomers were easily identified from the observed coupling constants of the vicinal protons on the β -lactam ring, such that the smaller *J* value (1.75–2.5 Hz) is for the *trans*- β -lactam isomer.^{[54][55][56]} ¹³C NMR spectroscopic data showed the β -lactam carbonyl signal at 158.4-160.7 ppm, whereas C-3 resonated at 66.5-77.4 ppm, and C-4 at 60.0-65.4 ppm. Furthermore, the FT-IR spectra of derivatives **4a-j** confirmed the formation of cycloadducts. We found the characteristic absorption band at 1705-1766 cm⁻¹, attributed to the β -lactam carbonyl group.

Results and Discussion

Chemistry

As part of this work, diastereoselective ketene-imine cycloaddition reaction was employed to synthesize a selection of benzotriazole- β -lactams. Table 1 shows the structure of synthesized compounds. First, 1*H*-benzo[*d*][1,2,3]triazole (**1**) was converted into 2-(1*H*-benzo[*d*][1,2,3]triazol-1-yl)acetic acid (**3**) by refluxing in the presence of 2-bromoacetic acid in aqueous sodium hydroxide.^[50] Subsequent reaction of 2-(1*H*-benzo[*d*][1,2,3]triazol-1-yl)acetic acid (**3**) with Schiff base, tosyl chloride, and triethyl amine (Et₃N) provided β -lactams **4a-j** in moderate to good yields (Figure 2).

After purification by column chromatography, the synthesized compounds have been characterized by mass spectroscopy, ¹H NMR, ¹³C NMR, and FT-IR spectroscopy, in addition to elemental analysis. Derivative **4a** was also studied by X-ray crystallography, as shown in Figure 3.



Conditions: i) NaOH, reflux ii) TsCl, Et₃N, CH₂Cl₂, rt

Figure 2. Synthesis of 1*H*-benzo[*d*][1,2,3]triazole- β -lactams **4a-j**.

Table 1. Synthesis of benzotriazole- β -lactam derivatives **4a-j**.

Entry	Structure	Yield (%)	Entry	Structure	Yield (%)
4a		45	4f		65
4b		23	4g		17

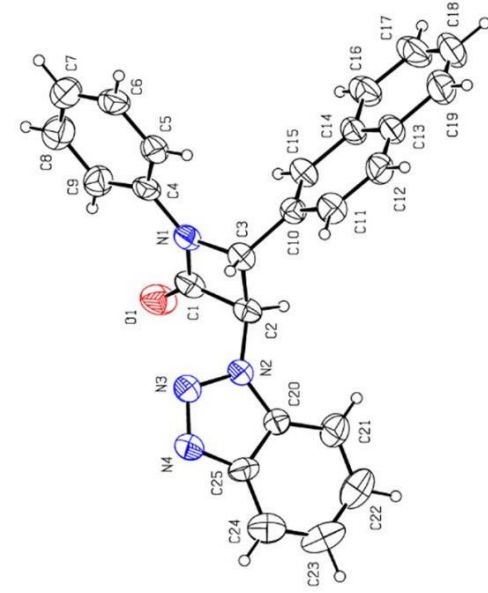
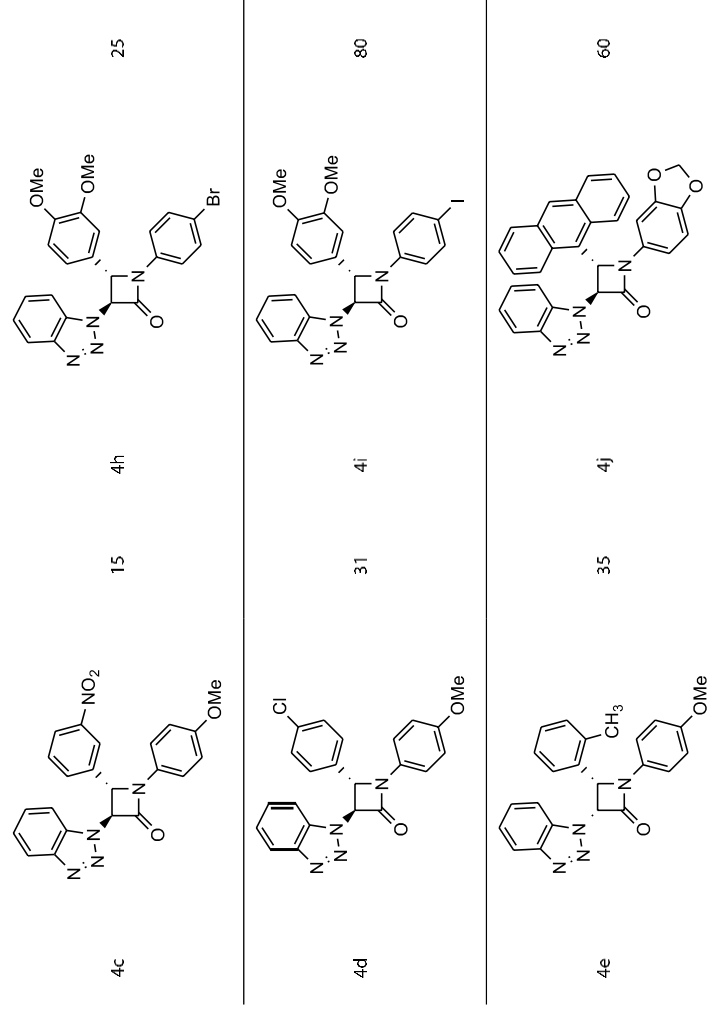


Figure 3. ORTEP Diagram of **4a**.

Biological activity

In a subsequent assessment, the biological properties of synthesized β -lactams were examined, first against Gram-positive and Gram-negative bacteria. Nevertheless, no antimicrobial activity was observed even at high concentrations (up to 400 μM). However, the compounds displayed moderate to good antimalarial activities against chloroquine-resistant *P. falciparum* (K1 strain) with IC_{50} values from 5.56 μM to 25.65 μM (Table 2). Since the β -lactams **4a-j** possess a benzotriazole group at the C-3 position of the ring, the preliminary structure-activity relationship (SAR) study indicates that the substituents at N-1 and C-4 of the β -lactam ring are responsible for these activity differences. There is no clear SAR dealing with our antimalarial activity results. Nevertheless, the most potent derivatives were **4j** (IC_{50} = 5.56 μM) and **4a** (IC_{50} = 9.73 μM) with 9-antracryl and 2-

naphthyl at C-4 position of the β -lactam ring, respectively. This suggests that the presence of naphthyl and anthracenyl substituents can be critical to the geometric orientation of compounds in the active site of the enzyme. The SAR study indicates that the β -lactam nitrogen moiety (R^1) plays a more significant role than the C-4 substituent R^2 , where R^2 is substituted phenyl ring. Comparison of lactams **4h** bearing 4-bromophenyl group at N-1 position, with an IC_{50} of 14.40 μM , to the 4-iodo analog **4i** (IC_{50} of 25.65 μM) suggests that the size of the *N*-aryl group might be a factor. Additionally, there is not significantly difference in activity between **4c,4d**, and **4e** with *p*-methoxy group on the N-1 of the ring. The presence of a chlorine atom might have positive effect on the mechanism of action involved against the *P. falciparum* K1 strain.

Furthermore, cytotoxicity effect of all compounds was determined against HepG2 cell line. As presented in Table 3, the desired compounds were non-cytotoxic on mammalian cells at 50 μM concentration even after 72 h incubation. The biological analyses of the synthesized compounds demonstrated their minor harmful effects on eukaryotes such as humans, supporting their potential therapeutic and environmental uses. The very low cytotoxicity effect on HepG2 cell line as hepatocytes at 200 μM final concentration revealed suitable selectivity of the compounds for malaria treatment.

Table 2. Antimalarial activities of benzotriazole- β -lactams 4a-j.

Compound	IC ₅₀ (μ M)	
	<i>P. falciparum</i> K1	<i>P. falciparum</i> K1
4a	9.73	4g 10.68
4b	11.44	4h 14.40
4c	10.71	4i 25.65
4d	10.24	4j 5.56
4e	10.79	Chloroquine 1.02
4f	14.64	

Table 3. Cell viability of HepG2 cells treated with the compounds of interest in two different concentrations, 50 and 200 μ M.

Compounds	Cell viability (%) ^a	
	50 μ M	200 μ M
4a	74.4 \pm 11.7	68.0 \pm 5.2
4b	83.7 \pm 7.0	56.2 \pm 7.5
4c	89.5 \pm 4.5	60.8 \pm 6.2
4d	90.0 \pm 2.5	69.8 \pm 3.0
4e	90.0 \pm 1.0	77.4 \pm 3.7
4f	77.7 \pm 4.0	65.5 \pm 9.6
4g	77.5 \pm 3.0	56.9 \pm 4.1
4h	81.1 \pm 3.7	68.0 \pm 8.7
4i	80.3 \pm 3.4	66.2 \pm 11.3
4j	82.7 \pm 2.7	75.5 \pm 8.8

^aThe cell survival assessments were conducted after 72 h incubation of the compounds.

Molecular docking studies

For a better understanding of the antimalarial activity of these benzotriazole- β -lactams, and to examine possible enzyme binding modes, each of the new β -lactam compounds **4a-j** were individually docked into the active site of *P. falciparum* dihydrofolate reductase enzyme using AutoDock. This reductase protein in *Plasmodium* has been investigated as a putative target for other antimalarial compounds having similar structural and activity profiles to our β -lactams.^[46] The published X-ray structure

(PfDHFR PDB ID: 1J3I) of the reductase was used for docking with the energy-minimized structure of each test compound. The predicted binding interactions of compounds **4a-j** against PfDHFR are shown in Table 4. We will present the docking results for two of the β -lactams, **4a** and **4j**, whose preferred binding orientations in the reductase active site are depicted in Figure 4. The naphthyl and anthracenyl substituents in compounds **4a** (IC₅₀ = 9.73 μ M) and **4j** (IC₅₀ = 5.56 μ M) are each directed toward the same open pocket and interact with Leu46 and Ile112 via π - π interactions. The triazole functionality within the benzotriazole- β -lactam core in both compounds is predicted to experience strong H-bond interactions with Ser108—the aryl groups on the ring help to align the orientation of the benzotriazole group to enhance this H-bond interaction. These docked conformations therefore suggest that the orientation of the

benzotriazole moiety relative to the pertinent amino acid active site residues are highly dependent on the aryl ring substituents.

Table 4. Interactions of benzotriazole- β -lactams 4a-j against PfDHFR.

Compd.	Polar interaction	π - interaction
4a	Ser108 to N3	Val195 to ring A, Leu46 to ring (B, C and D), Ile112 to ring D
4b	Ser111 to N3	No interaction
4c	Ser111 to N3	No interaction
4d	Ser111 to N3	No interaction
4e	No interaction	No interaction
4f	No interaction	No interaction
4g	No interaction	No interaction
4h	Ser111 to N3	Ala16 to ring D
4i	Ser111 to N3	Ala16 to ring D
4j	Ser108 to N3	Ile112 to ring D, Ala16 to ring D

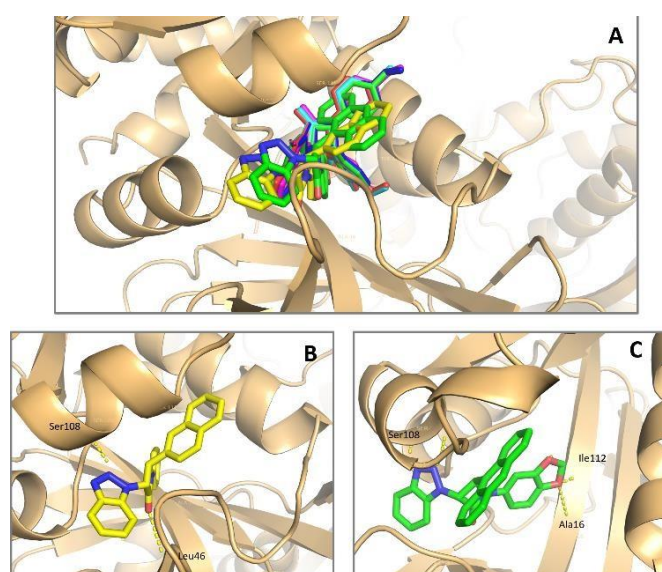


Figure 4. A) Comparative binding poses of all designed compounds in *P. falciparum* dihydrofolate reductase enzyme (PfDHFR) structure active site. B) The backbone of the compound **4a** (yellow sticks) and, C) **4j** (green sticks) are shown in active site (PDB ID: 1J3I). Important residues of the binding site are labeled, while colored dashed lines indicate different interactions.

Conclusions

Globally, malaria is still one of the most serious threats to public health that must be addressed. Herein, a series of novel β -lactam derivatives containing the benzotriazole moiety at C-3 has been synthesized, and their biological activities were evaluated. Some derivatives have good *in vitro* antimalarial activity. The modification to the aromatic ring on the N-1 and C-4 positions has the potential to modulate the activity against *P. falciparum* dihydrofolate reductase enzyme. Currently, further studies are being conducted to improve the design and biological activities of additional derivatives. A computer model of the interaction of compounds **4a** and **4j** with the *P. falciparum* dihydrofolate reductase enzyme was developed. The model involved a favorable H-bond between the Ser108 residue and the benzotriazole moiety.

Experimental Section

General

Our chemicals were obtained from Merck, Fluka, and Acros chemical companies without further purification. Before use, standard methods were followed to dry reagents and solvents, such as CH₂Cl₂. The HepG2 cell line was purchased from department of cell bank, Pasteur institute, Tehran, Iran. Spectral data was used to identify all compounds. A Shimadzu FT-IR 8300 spectrophotometer was used to measure the FT-IR spectra of potassium bromide pellets (λ in cm⁻¹). A Bruker Avance DPX instrument was used to measure ¹H NMR and ¹³C NMR spectra in dimethylsulfoxide-*d*₆ (DMSO-*d*₆) and chloroform-*d* (CDCl₃-*d*) (¹H NMR 250 MHz, ¹³C NMR 62.5 MHz). Chemical shift (δ) and *J* values are expressed in parts per million (ppm) and Hertz (Hz), respectively. The following splitting patterns are indicated: s: singlet, d: doublet, t: triplet, m: multiplet, dd: doublet of doublet, and br: broad peak. On a Shimadzu GC-MS QP 1000 EX, mass spectra were recorded. We performed elemental analyses on a Thermo Finnigan Flash EA-1112 series. Using a Buchi 510 melting point apparatus, melting points were measured. We used silica gel F254 analytical sheets (Fluka) and Kieselgel (230–270 mesh from Merck) to perform thin-layer chromatography and column chromatography, respectively. Moreover, X-ray data were collected on a Stoe IPDS 2 diffractometer.

Preparation of benzotriazole-1-yl acetic acid (3)

A mixture of benzotriazole (1.00 mmol), bromoacetic acid (1.00 mmol), sodium hydroxide (2.00 mmol) and 10.00 mL water was refluxed slowly for 3 h, and filtered immediately while still hot. At room temperature, the filtrate was cooled and acidified with dilute hydrochloric acid (HCl) until no deposits appeared. The mixture was filtered, washed thoroughly with cold water, dried, and recrystallized from ethanol (Yield 76%, mp 216–218 °C).

General procedure for the preparation of 1H-benzotriazole-1-yl-2,3-dimethyl-4-oxo-1,2,3,4-tetrahydro-1H-benzotriazin-4-ylidene-2-one (4a-j)

To a solution of Schiff base (1.00 mmol) in dry CH₂Cl₂ were added the benzotriazole-1-ylacetic acid (1.50 mmol), *p*-toluenesulfonyl chloride (1.50 mmol), triethylamine (4.00–5.00 mmol) and let to stirred for more than 30 h at room temperature. Afterward, the resultant mixture was washed with HCl (1 N), saturated sodium bicarbonate (NaHCO₃), and brine. Anhydrous Na₂SO₄ was used to dry the organic layer. Finally, the solvent was evaporated, and the crude product thus obtained was purified by recrystallization or column chromatography (1:19 ethyl acetate and *n*-hexane as eluent).

3-(1H-Benzotriazol[1,2,3-triazol-1-yl]-4-(naphthalen-2-yl))-1-phenylazetidion-2-one (4a). Purified by column chromatography to give a white solid (45%); mp: 172–174 °C; ¹H NMR (250 MHz, CDCl₃): δ =5.68 (d,

J=1.75 Hz, 1H; H-4), 6.01 (d, *J*=1.75 Hz, 1H; H-3), 7.04 (t, *J*=7.50 Hz, 1H; ArH), 7.18 (s, 1H; ArH), 7.23 (d, *J*=7.50 Hz, 2H; ArH), 7.33–7.47 (m, 6H; ArH), 7.72–7.86 ppm (m, 6H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ 64.1 (C-4), 77.4 (C-3), 117.8, 118.8, 122.6, 125.0, 126.4, 126.9, 127.2, 127.9, 128.0, 129.2, 129.8, 132.2, 133.7, 136.8, 144.9 (aromatic carbons), 159.1 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1758 cm⁻¹ (CO β -lactam); GC-MS: m/z: 390 [M⁺]; Anal. calcd for C₂₅H₁₈N₆O: C 76.91, H 4.65, N 14.35, Found: C 76.89, H 4.60, N 14.46.

3-(1H-Benzotriazol[1,2,3-triazol-1-yl]-4-(3-nitrophenyl))-1-(*p*-tolyl)azetidion-2-one (4b). Recrystallized from CH₂Cl₂/*n*-hexane (1:1) to give a light yellow solid (23%); mp: 173–175 °C; ¹H NMR (250 MHz, CDCl₃): δ 2.19 (s, 3H; CH₃), 5.66 (d, *J*=2.25 Hz, 1H; H-4), 5.88 (d, *J*=2.25 Hz, 1H; H-3), 6.80 (d, *J*=9.00 Hz, 2H; ArH), 7.21 (d, *J*=9.50 Hz, 2H; ArH), 7.38–7.48 (m, 3H; ArH), 7.59 (t, *J*=8.75 Hz, 1H; ArH), 7.71 (d, *J*=7.75 Hz, 1H; ArH), 8.05 (d, *J*=8.25 Hz, 1H; ArH), 8.19–8.23 ppm (m, 2H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ 29.6 (CH₃), 61.7 (C-4), 71.2 (C-3), 109.2, 114.8, 119.1, 120.6, 121.4, 124.5, 124.8, 128.7, 129.2, 130.8, 132.0, 137.3, 146.2, 157.2 (aromatic carbons), 158.4 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1766 cm⁻¹ (CO β -lactam); GC-MS: m/z: 399 [M⁺]; Anal. calcd for C₂₂H₁₇N₅O₃: C 66.16, H 4.29, N 17.53, Found: C 66.10, H 4.12, N 17.62.

3-(1H-Benzotriazol[1,2,3-triazol-1-yl]-1-(4-methoxyphenyl))-4-(3-nitrophenyl)azetidion-2-one (4c).

Purified by column chromatography to give a yellow solid (15%); mp: 171–173 °C; ¹H NMR (250 MHz, CDCl₃): δ =3.71 (s, 3H; OCH₃), 5.67 (d, *J*=1.75 Hz, 1H; H-4), 5.87 (d, *J*=1.75 Hz, 1H; H-3), 6.78 (d, *J*=9.00 Hz, 2H; ArH), 7.19–7.48 (m, 5H; ArH), 7.55 (t, *J*=7.50 Hz, 1H; ArH), 7.71 (d, *J*=7.25 Hz, 1H; ArH), 8.06 (d, *J*=8.25 Hz, 1H; ArH), 8.23 ppm (s, 2H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ =55.5 (OCH₃), 61.7 (C-4), 71.2 (C-3), 109.2, 114.8, 119.1, 120.6, 121.4, 124.5, 124.8, 128.7, 130.8, 132.0, 137.3, 146.2, 157.2, 158.4 (aromatic carbons), 162.2 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1705 cm⁻¹ (CO β -lactam); GC-MS: m/z: 415 [M⁺]; Anal. calcd for C₂₂H₁₇N₅O₄: C 63.61, H 4.13, N 16.86, Found: C 63.75, H 4.09, N 16.93.

3-(1H-Benzotriazol[1,2,3-triazol-1-yl]-4-(4-chlorophenyl))-1-(4-methoxyphenyl)azetidion-2-one (4d).

Purified by column chromatography to give a yellow solid (31%); mp: 143–145 °C; ¹H NMR (250 MHz, CDCl₃): δ =3.69 (s, 3H; OCH₃), 5.47 (d, *J*=2.25 Hz, 1H; H-4), 5.80 (d, *J*=2.25 Hz, 1H; H-3), 6.77 (d, *J*=8.75 Hz, 2H; ArH), 7.18–7.44 (m, 9H; ArH), 8.02 ppm (d, *J*=8.75 Hz, 1H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ =55.4 (OCH₃), 62.2 (C-4), 71.3 (C-3), 109.2, 114.6, 119.1, 120.4, 124.6, 127.5, 128.5, 129.6, 129.8, 133.4, 135.5, 146.2, 157.0 (aromatic carbons), 158.7 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1743 cm⁻¹ (CO β -lactam); GC-MS: m/z: 404 [M⁺]; Anal. calcd for C₂₂H₁₇ClN₅O₂: C 65.27, H 4.23, N 13.84, Found: C 65.17, H 4.29, N 13.91.

3-(1H-Benzotriazol[1,2,3-triazol-1-yl]-1-(4-methoxyphenyl))-4-(*o*-tolyl)azetidion-2-one (4e).

Recrystallized from CHCl₃/*n*-hexane to give a light yellow solid (35%); mp: 196–198 °C; ¹H NMR (250 MHz, CDCl₃): δ =2.22 (s, 3H; CH₃), 3.73 (s, 3H; OCH₃), 5.77 (d, *J*=5.25 Hz, 1H; H-4), 6.63 (d, *J*=5.25 Hz, 1H; H-3), 6.68–6.82 (m, 5H; ArH), 7.09–7.48 (m, 6H; ArH), 7.74 ppm (d,

60.0 (C-4), 66.6 (C-3), 110.3, 114.5, 118.9, 119.8, 123.8, 125.6, 126.6, 127.4, 128.5, 128.9, 130.3, 130.4, 135.5, 145.5, 156.9 (aromatic carbons), 158.6 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1751 cm^{-1} (CO β -lactam); GC-MS: m/z: 384 [M⁺]; Anal. calcd for C₂₃H₂₀N₄O₂: C 71.86, H 5.24, N 14.57, Found: C 71.73, H 5.22, N 14.61.

3-(1*H*-Benzotriazol-1-yl)-4-(4-chlorophenyl)-1-(2-methoxyphenyl)azetidin-2-one (4f). Recrystallized from EtOAc/*n*-hexane to give a light yellow solid (65%): mp: 127–129 °C; ¹H NMR (250 MHz, CDCl₃): δ =3.63 (s, 3H; OCH₃), 5.88 (d, *J*=2.00 Hz, 1H; H-4), 6.01 (d, *J*=2.00 Hz, 1H; H-4), 6.82 (d, *J*=9.00 Hz, 1H; ArH), 6.96 (t, *J* = 7.5 Hz, 1H; ArH), 7.15 (t, *J*=8.25 Hz, 1H; ArH), 7.28–7.50 (m, 6H; ArH), 7.67 (d, *J*=8.00 Hz, 1H; ArH), 7.84 (d, *J*=8.00 Hz, 1H; ArH), 8.06 ppm (d, *J*=8.25 Hz, 1H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ =55.4 (OCH₃), 65.4 (C-4), 71.8 (C-3), 109.6, 112.1, 120.2, 121.1, 123.5, 124.1, 124.5, 127.5, 127.6, 128.3, 129.3, 132.3, 134.9, 151.5 (aromatic carbons), 160.5 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1766 cm^{-1} (CO β -lactam); GC-MS: m/z: 404 [M⁺]; Anal. calcd for C₂₂H₁₇ClN₄O₂: C 65.27, H 4.23, N 13.84, Found: C 65.06, H 4.25, N 13.89.

3-(1*H*-Benzotriazol-1-yl)-4-(4-chlorophenyl)-1-(3-nitrophenyl)azetidin-2-one (4g). Recrystallized from EtOAc/*n*-hexane to give a white solid (17%): mp: 231–233 °C; ¹H NMR (250 MHz, CDCl₃): δ =5.71 (d, *J*=5.25 Hz, 1H; H-4), 6.53 (d, *J*=5.25 Hz, 1H; H-3), 6.95 (dd, *J*=8.50 Hz, 3H; ArH), 7.19 (s, 1H; ArH), 7.25–7.33 (m, 3H; ArH), 7.50 (t, *J*=8.25 Hz, 1H; ArH), 7.83 (t, *J*=8.25 Hz, 2H; ArH), 7.97 (d, *J*=8.25 Hz, 1H; ArH), 8.14 ppm (s, 1H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ =61.5 (C-4), 66.8 (C-3), 108.9, 112.1, 119.9, 120.4, 123.2, 124.3, 128.0, 128.1, 128.9, 130.6, 132.5, 141.5 (aromatic carbons), 159.1 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1766 cm^{-1} (CO β -lactam); GC-MS: m/z: 419 [M⁺]; Anal. calcd for C₂₁H₁₄ClN₄O₂: C 60.08, H 3.36, N 16.68, Found: C 59.93, H 3.34, N 16.74.

3-(1*H*-Benzotriazol-1-yl)-1-(4-bromophenyl)-4-(3,4-dimethoxyphenyl)azetidin-2-one (4h). Recrystallized from CHCl₃ to give a yellow solid (25%): mp: 121–124 °C; ¹H NMR (250 MHz, CDCl₃): δ =3.83 (s, 3H; OCH₃), 3.89 (s, 3H; OCH₃), 5.84 (d, *J*=2.50 Hz, 1H; H-4), 5.91 (d, *J*=2.50 Hz, 1H; H-3), 6.85–6.95 (m, 3H; ArH), 7.26–7.52 (m, 7H; ArH), 8.12 ppm (d, *J*=8.00 Hz, 1H; ArH); ¹³C NMR (62.5 MHz, CDCl₃): δ =56.0 (OCH₃), 56.1 (OCH₃), 63.0 (C-4), 71.5 (C-3), 108.5, 109.1, 111.8, 118.0, 118.8, 119.2, 120.5, 124.6, 126.6, 128.4, 132.4, 132.5, 135.5, 146.2, 150.0, 150.1 (aromatic carbons), 159.8 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1766 cm^{-1} (CO β -lactam); GC-MS: m/z: 479 [M⁺]; Anal. calcd for C₂₃H₁₉BrN₄O₂: C 57.63, H 4.00, N 11.69, Found: C 57.52, H 4.03, N 11.7.

3-(1*H*-Benzotriazol-1-yl)-4-(3,4-dimethoxyphenyl)-1-(4-

iodophenyl)azetidin-2-one (4i). Recrystallized from EtOAc/*n*-hexane to give a yellow solid (80%): mp: 126–128 °C; ¹H NMR (250 MHz, CDCl₃): δ =3.83 (s, 3H; OCH₃), 3.89 (s, 3H; OCH₃), 5.53 (d, 1H, *J*=2.25 Hz; H-4), 5.90 (d, *J*=2.25 Hz, 1H; H-3), 6.85–6.97 (m, 3H; ArH), 7.18 (d, *J*=8.75 Hz, 2H; ArH), 7.39–

ArH); ¹³C NMR (62.5 MHz, DMSO-*d*₆): δ =55.4 (OCH₃), 62.6 (C-4), 69.5 (C-3), 89.0, 110.1, 110.4, 111.7, 119.5, 119.7, 124.7, 126.6, 128.4, 132.4, 135.8, 137.9, 145.0, 149.0, 149.3 (aromatic carbons), 160.7 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1766 cm^{-1} (CO β -lactam); GC-MS: m/z: 526 [M⁺]; Anal. calcd for C₂₃H₁₉I₄N₄O₂: C 52.49, H 3.64, N 10.64, Found: C 52.51, H 3.68, N 10.69.

4-(Anthracen-9-yl)-3-(1*H*-benzotriazol-1-yl)-1-(benzotriazol-1,2,3-dioxol-5-yl)azetidin-2-one (4j). Recrystallized from EtOAc/*n*-hexane to give a yellow solid (60%): mp: 133–135 °C; ¹H NMR (250 MHz, DMSO-*d*₆): δ =6.04 (d, *J*=2.50 Hz, 1H; H-3), 6.06 (d, *J*=2.50 Hz, 1H; H-4), 6.74–6.91 (m, 2H; CH₂), 7.14 (t, *J*=7.50 Hz, 1H; ArH), 7.23–7.40 (m, 5H; ArH), 7.48 (t, *J*=7.50 Hz, 1H; ArH), 7.60 (d, *J*=7.50 Hz, 1H; ArH), 7.64 (d, *J*=7.50 Hz, 2H; ArH), 7.77 (d, *J*=7.50 Hz, 1H; ArH), 7.88–7.96 (m, 2H; ArH), 8.42 (s, 1H; ArH), 8.71 (d, *J*=10.00 Hz, 1H; ArH), 8.82 ppm (d, *J*=10.00 Hz, 1H; ArH); ¹³C NMR (62.5 MHz, DMSO-*d*₆): δ =61.4 (C-4), 66.5 (C-3), 98.9 (CH₂), 101.4, 108.7, 109.7, 109.9, 118.7, 120.7, 122.9, 123.9, 124.4, 124.6, 124.9, 126.8, 127.2, 128.9, 129.1, 129.3, 129.7, 129.9, 130.7, 132.2, 132.3, 143.7, 144.2, 147.6 (aromatic carbons), 159.7 ppm (CO, β -lactam); IR (KBr): $\bar{\nu}$ =1751 cm^{-1} (CO β -lactam); GC-MS: m/z: 484 [M⁺]; Anal. calcd for C₃₀H₂₀N₄O₂: C 74.37, H 4.16, N 11.56, Found: C 74.25, H 4.13, N 11.61.

Determination of minimal inhibitory concentrations against bacteria

All compounds were evaluated for their antimicrobial activity by determining minimal inhibitory concentrations (MICs). Microbroth dilution methods were used to determine the MICs of the compounds according to NCCLS guidelines M7-A2. Trypticase soy agar (Becton Dickinson) was used to grow the bacteria at 37 °C for 24 h (*E. coli* ATCC28922, *P. aeruginosa* PAO1, *S. aureus* ATCC25923) in LB broth. Inocula were prepared in TCE (tryptone 0.1%, NaCl 8%, wt/vol) by adjusting the turbidity at 623 nm to obtain 1–3 × 10⁶ CFU/mL. In sterile 96-well microplates, broth microdilution assays were performed to assess antimicrobial activity. The synthesized β -lactam was solubilized in DMSO at a concentration of 5 mg/mL and then transferred to each microplate well to obtain a two-fold serial dilution in 100 μ L of broth and 100 μ L of inocula containing 2–6 × 10⁵ CFU of each bacterium and yeast. In all cases, concentrations of the DMSO did not exceed 2% of the total proportion. Several wells were reserved for the controls, inoculum viability and solvent effect. Growth was assessed at 623 nm using an IEMS Labsystem automatic plate reader after 24 or 48 hours of incubation. For each agent, the MIC was determined from duplicate observations at the lowest concentration at which no visible growth was observed.

Determination of *in vitro* antimalarial activity

The chloroquine-resistant *P. falciparum* K1 strain (MR4-ATCC) was cultured

supplemented with 10% human serum, 20 mg/L gentamycin, 25 mM NaHCO₃, and 50 mg/l hypoxanthine, at 37 °C under an environment of 85% N₂, 10% O₂ and 5% CO₂.^[57] Sorbitol was used to synchronize cultures.^[58] Stock solutions of compounds were prepared in sterile DMSO (10 mM) and later dilutions were with complete culture medium. Parasites were cultured (0.5% parasitemia; 1.5% hematocrit) in the presence of increasing concentrations of compounds (top concentration = 125 µM) in 96-well plates (final volume: 200 µL/well). Chloroquine was used as a reference drug. The negative control was DMSO (without compound) at a concentration equal to that of the drug-treated samples. An atmosphere of 85% N₂, 10% O₂, and 5% CO₂ was used to incubate the plates for 72 hours at 37°C, and then they were frozen at -80 °C. A homemade HRP2-based ELISA assay revealed parasite growth.^[59] The Sunrise™ spectrophotometer (Tecan, Austria) and Magellan™ data analysis software (Tecan, Austria) were used to measure absorbance at 450 nm. Non-linear regression analysis was used to calculate the 50% inhibitory concentrations (IC₅₀) from the dose response relationship fitted by softwareICEstimator 1.2.^{[60][61]}

Cell viability assay

The HepG2 cancerous cell line (liver adenocarcinoma cell line) was cultured in a humidified atmosphere containing 5% CO₂ at 37 °C in DMEM high medium supplemented with 100 units/mL penicillin, 100 µg/mL streptomycin and 10% fetal bovine serum. Cell toxicity of each compound was performed using the MTT method. This study was conducted by seeding cells in 96 well plates and allowing them to incubate for 24 hours at 37 °C in the presence of 5% CO₂. Afterward, a defined number of cells was exposed to the synthesized β-lactams at both final concentrations of 50 and 200 µM for 72 h. Then, 10 µM of 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) solution was added to each well to reach a final concentration of 0.45 mg/ml, and incubated at 37°C for 4 h. Finally, 100 µM solubilization solution (40% dimethylformamide (DMF) in 16% sodium dodecyl sulfate (SDS) and 2% glacial acetic acid, pH of about 4.7) was added to each well to dissolve the formazan crystals. The absorbances were recorded at 570 and 630 nm by SPECTROstar ^{nano}, BMG Labtech, Germany. Cell viability percentage was calculated as follows:

$$\text{Cell viability (\%)} = \left[\frac{A_{\text{T}} (\text{sample})}{A_{\text{T}} (\text{control})} \right] \times 100$$

$$A_{\text{T}} = A_{570} - A_{630}$$

Molecular docking studies

In this study, molecular docking was performed by Auto-Dock version (1.5.6). X-ray crystallographic structures of *P. falciparum* (1J3I.pdb) were obtained from the Brookhaven Protein Data Bank (<http://www.rcsb.org>). It was necessary to remove water molecules and the cognate ligand from the receptor. The ligand structure was constructed by Hyper Chem software version 8.0.10 and then energetically minimized by MM⁺ force field. The redocking process with the cognate ligand and the receptor resulted in a root mean square deviation (RMSD) of 1.87Å, which falls below the

acceptable threshold of 2Å. The active sites were defined based on the center (box x: 28.46 y: 5.73 z: 62.35) and radius of cognate ligand which are mentioned in X-ray crystal structure. The Lamarckian genetic algorithm with grid size 25x25x25 (grid spacing 0.375 Å) was used for docking computations, and in accordance with previous work, docking was carried out.^[62]

Supplementary Material (optional)

Supporting information for this article is available on the WWW under <http://dx.doi.org/10.1002/MS-number>.

Acknowledgements

The authors thank the Shiraz University Research Council for financial support (Grant No. 98-GR-SC-23).

Keywords: Antimalarial, 2-Azetidinone, Cycloaddition, *In silico*, Nitrogen heterocycles.

Author Contribution Statement

Malihe Aye: Conceptualization, Visualization, Methodology, Data curation and analysis, Writing-original draft; Aliasghar Jarrahpour: Conceptualization, Supervision, Visualization, Reviewing and editing; Zahra Haghhighjoo: Docking study, Writing-original draft; Roghayeh Heiran: Writing-original draft preparation; Data analysis; Preparing Figures, Reviewing and editing; Roya Pourmejati: Methodology, Writing-original draft; Hamid Reza Karbalaeei-Heidari: Methodology; Veronique Sinou: Methodology; Jean Michel Brunel: Methodology; Mehmet Akkurt: Crystallography study; Namik Özdemir: Crystallography study; Edward Turos: Reviewing and editing.

References

- [1] J. E. Hyde, *FEBS J.* **2007**, *274*, 4688–4698.
- [2] F. V. Rahmasari, P. B. S. Asih, F. K. Dewayanti, C. Rotejanaprasert, P. Charunwatthana, M. Imwong, D. Syafruddin, *Malar. J.* **2022**, *21*, 354.
- [3] World malaria report 2022, <https://www.who.int/publications/i/item/9789240064898>.
- [4] Talapko, Škrlec, Alebić, Jukić, Včev, *Microorganisms* **2019**, *7*, 179.
- [5] A. Jarrahpour, Z. Jowkar, Z. Haghhighjoo, R. Heiran, J. A. Rad, V. Sinou, F. Rouviev, C. Latour, J. M. Brunel, N. Özdemir, *Med. Chem. Res.* **2022**, *31*, 1026–1034.
- [6] A. Saneii-Dehkordi, R. Heiran, M. D. Moemenbellah-Fard, S. Sayah, M. Osanloo, *Bionanoscience* **2022**, *12*, 359–369.
- [7] A. Saneii-Dehkordi, R. Heiran, G. Roozitalab, N. Elahi, M. Osanloo, *Psyche A J. Entomol.* **2022**, *2022*, 1–8.
- [8] I. Seck, I. Ciss, A. Diéchiou, M. Baldé, S. Ka, L. A. Ba, S. F. Ndoye, B. Figadère, B. Seon-Meniel, G. Gomez, S. Cojean, S. Pomei, P. M. Loiseau, Y. Fall, M. Seck, *Med. Chem. Res.* **2023**, *32*, 158–164. P. D. Mehta, N. P. S. Sengar, A. K. Pathak, *Eur. J. Med. Chem.*

- [10] **2010**, 45, 5541–5560.
N. S. Vatmurge, B. G. Hazra, V. S. Pore, F. Shirazi, P. S. Chavan, M. V. Deshpande, *Bioorg. Med. Chem. Lett.* **2008**, 18, 2043–2047.
- [11] S. B. Rosenblum, T. Huynh, A. Afonso, H. R. Davis, N. Yumibe, J. W. Clader, D. A. Burnett, *J. Med. Chem.* **1998**, 41, 973–980.
- [12] D. A. Burnett, M. A. Caplen, H. R. Davis, R. E. Burrier, J. W. Clader, *J. Med. Chem.* **1994**, 37, 1733–1736.
- [13] W. A. Slusarchyk, S. A. Bolton, K. S. Hartl, M.-H. Huang, G. Jacobs, W. Meng, M. L. Ogletree, Z. Pi, W. A. Schumacher, S. M. Seiler, J. C. Sutton, U. Treuner, R. Zahler, G. Zhao, G. S. Bisacchi, *Bioorg. Med. Chem. Lett.* **2002**, 12, 3235–3238.
- [14] W. T. Han, A. K. Trehan, J. J. Kim Wright, M. E. Federici, S. M. Seiler, N. A. Meanwell, *Bioorg. Med. Chem.* **1995**, 3, 1123–1143.
- [15] C. D. Guillon, G. A. Koppel, M. J. Brownstein, M. O. Chaney, C. F. Ferris, S. Lu, K. M. Fabio, M. J. Miller, N. D. Heindel, D. C. Hunden, R. D. G. Cooper, S. W. Kaldor, J. J. Skelton, B. A. Dressman, M. P. Clay, M. I. Steinberg, R. F. Bruns, N. G. Simon, *Bioorg. Med. Chem.* **2007**, 15, 2054–2080.
- [16] A. K. Halve, D. Bhadauria, R. Dubey, *Bioorg. Med. Chem. Lett.* **2007**, 17, 341–345.
- [17] R. Heiran, A. Jarrahpour, E. Riazimontazer, A. Gholami, A. Troudi, C. Digiorgio, J. M. Brunel, E. Turos, *ChemistrySelect* **2021**, 6, 5313–5319.
- [18] E. Riazimontazer, R. Heiran, A. Jarrahpour, A. Gholami, Z. Hashemi, A. Kazemi, *ChemistrySelect* **2022**, 7, e202203373.
- [19] S. Ranjbari, A. Jarrahpour, S. Kianpour, S. Sepehri, R. Heiran, Y. Ghasemi, E. Turos, *J. Mol. Struct.* **2023**, 1294, 136298.
- [20] M. Zarei, M. Mohamadzadeh, *Tetrahedron* **2011**, 67, 5832–5840.
- [21] A. Jarrahpour, M. Aye, V. Sinou, C. Latour, J. M. Brunel, *J. Iran. Chem. Soc.* **2015**, 12, 2083–2092.
- [22] R. Heiran, S. Sepehri, A. Jarrahpour, C. Digiorgio, H. Douafer, J. M. Brunel, A. Gholami, E. Riazimontazer, E. Turos, *Bioorg. Chem.* **2020**, 102, 104091.
- [23] P. Singh, S. Sachdeva, R. Raj, V. Kumar, M. P. Mahajan, S. Nasser, L. Vivas, J. Gut, P. J. Rosenthal, T.-S. Feng, K. Chibale, *Bioorg. Med. Chem. Lett.* **2011**, 21, 4561–4563.
- [24] P. Singh, R. Raj, V. Kumar, M. P. Mahajan, P. M. S. Bedi, T. Kaur, A. K. Saxena, *Eur. J. Med. Chem.* **2012**, 47, 594–600.
- [25] N. Borazjani, S. Sepehri, M. Behzadi, A. Jarrahpour, J. A. Rad, M. Sasani, M. Mohkam, Y. Ghasemi, A. R. Akbarizadeh, C. Digiorgio, J. M. Brunel, M. M. Ghanbari, G. Batta, E. Turos, *Eur. J. Med. Chem.* **2019**, 179, 389–403.
- [26] G. Dive, C. Bouillon, A. Sliwa, B. Valet, O. Verlainne, E. Sauvage, J. Marchand-Brynaert, *Eur. J. Med. Chem.* **2013**, 64, 365–376.
- [27] I. Briguglio, S. Piras, P. Corona, E. Gavini, M. Nieddu, G. Boatto, A. Carta, *Eur. J. Med. Chem.* **2015**, 97, 612–648.
- [28] A. Mermer, S. Demirci, *Eur. J. Med. Chem.* **2023**, 259, 115655.
- [29] H. K. Gulati, N. Kumar, A. Sharma, Jyoti, A. Khanna, S. Sharma, R. Salwan, P. M. S. Bedi, *J. Mol. Struct.* **2023**, 1284, 135354.
- [30] A. Singh, K. Singh, J. Kaur, R. Kaur, A. Sharma, J. Kaur, U. Kaur, R. Chadha, P. M. S. Bedi, *ACS Chem. Neurosci.* **2023**, 14, 3291–3317.
- [31] S. Zhao, J. Liu, Z. Lv, G. Zhang, Z. Xu, *Eur. J. Med. Chem.* **2023**, 251, 115254.
- [32] B. Rathod, K. Kumar, *Chem. Biodivers.* **2022**, 19, DOI 10.1002/cbdv.202200679.
- [33] N. Agouram, *Med. Chem. Res.* **2023**, 32, 2458–2472.
- [34] A. Singh, K. Singh, A. Sharma, K. Joshi, B. Singh, S. Sharma, K. Batra, K. Kaur, D. Singh, R. Chadha, P. Mohinder Singh Bedi, *Chem. Biodivers.* **2023**, 20, DOI 10.1002/cbdv.202300024.
- [35] G. Tian, Q. Song, Z. Liu, J. Guo, S. Cao, S. Long, *Eur. J. Med. Chem.* **2023**, 259, 115603.
- [36] A. Sharma, R. Dubey, R. Bhupal, P. Patel, S. K. Verma, S. Kaya, V. Asati, *Mol. Divers.* **2023**, DOI 10.1007/s11030-023-10728-1.
- [37] H. B. Bollikolla, S. N. M. Boddapati, S. Thangamani, B. R. Mutchu, M. M. Alam, M. Hussien, S. B. Jonnalagadda, *J. Heterocycl. Chem.* **2023**, 60, 705–742.
- [38] S. M. Abdul Rahman, J. S. Bhatti, S. Thareja, V. Monga, *Eur. J. Med. Chem.* **2023**, 259, 115699.
- [39] L. Ravindar, S. A. Hasbullah, K. P. Rakesh, N. I. Hassan, *Eur. J. Med. Chem.* **2023**, 259, 115694.
- [40] D.-J. Fu, S.-Y. Zhang, Y.-C. Liu, L. Zhang, J.-J. Liu, J. Song, R.-H. Zhao, F. Li, H.-H. Sun, H.-M. Liu, Y.-B. Zhang, *Bioorg. Med. Chem. Lett.* **2016**, 26, 3918–3922.
- [41] P. Shiri, *Mini-Reviews Med. Chem.* **2021**, 21, 536–553.
- [42] A.-C. Vilain, B. Pirotte, J. Vergely, N. Boggetto, B. Masereel, M. Schynts, J. Delarge, M. Reboud-Ravaux, *J. Pharm. Pharmacol.* **2011**, 45, 466–472.
- [43] S. Dhawan, P. Awolade, P. Kisten, N. Cele, A. S. Pillay, S. T. Saha, M. Kaur, S. B. Jonnalagadda, P. Singh, *Chem. Biodivers.* **2020**, 17, e1900462.
- [44] J. M. Dive, S. B. Mhaske, C. R. Charolkar, D. G. Sant, S. G. Tupe, M. V. Deshpande, V. M. Khedkar, L. U. Nawale, D. Sarkar, V. S. Pore, *New J. Chem.* **2017**, 41, 470–479.
- [45] K. Kumar, S. Carrère-Kremer, L. Kremer, Y. Guérardel, C. Biot, V. Kumar, *Dalt. Trans.* **2013**, 42, 1492–1500.
- [46] K. Kumar, B. Pradines, M. Madamet, R. Amalvict, V. Kumar, *Eur. J. Med. Chem.* **2014**, 86, 113–121.
- [47] P. Singh, P. Singh, M. Kumar, J. Gut, P. J. Rosenthal, K. Kumar, V. Kumar, M. P. Mahajan, K. Bisetty, *Bioorg. Med. Chem. Lett.* **2012**, 22, 57–61.
- [48] A. Jarrahpour, M. Aye, J. A. Rad, S. Yousefinejad, V. Sinou, C. Latour, J. M. Brunel, E. Turos, *J. Iran. Chem. Soc.* **2018**, 15, 1311–1326.
- [49] A. Jarrahpour, P. Shirvani, V. Sinou, C. Latour, J. M. Brunel, *Med. Chem. Res.* **2016**, 25, 149–162.
- [50] X. Chen, C. Liu, J. Wang, Y. Li, J. Heterocycl. Chem. **2010**, 47, 1225–1229.
- [51] M. Akkurt, S. Karaca, A. A. Jarrahpour, M. Zarei, O. Büyükgüngör, *Acta Crystallogr. Sect. E Struct. Reports Online* **2008**, 64, o924–

- o924.
- [52] M. Akkurt, S. Karaca, A. A. Jarrahpour, D. Khalili, O. Büyükgüngör, *Acta Crystallogr. Sect. E Struct. Reports Online* **2006**, *62*, o866–o868.
- [53] M. Akkurt, S. Türktekin, A. Jarrahpour, S. A. T. Badrabad, O. Büyükgüngör, *Acta Crystallogr. Sect. E Struct. Reports Online* **2011**, *67*, o183–o183.
- [54] B. Bananezhad, M. Islami, *Synlett* **2017**, *28*, 1453–1456.
- [55] M. Alborz, A. Jarrahpour, R. Pournejati, H. R. Karbalaei-Heidari, V. Sinou, C. Latour, J. M. Brunel, H. Sharghi, M. Aberi, E. Turos, L. Wojtas, *Eur. J. Med. Chem.* **2018**, *143*, 283–291.
- [56] F. Zigheimat, M. Islami, F. Nourmohammadian, *Synlett* **2013**, *25*,
W. Trager, J. B. Jensen, *Science* (80-.). **1976**, *193*, 673–675. [57]
C. Lambros, J. P. Vanderberg, *J. Parasitol.* **1979**, *65*, 418–20. [58]
R. Nakweti, V. Sinou, S. Ndiku, F. Sabot, C. Franche, *European J. Med. Plants* **2017**, *18*, 1–10. [59]
H. Le Nagard, C. Vincent, F. Mentré, J. Le Bras, *Comput. Methods Programs Biomed.* **2011**, *104*, 10–18. [60]
<http://www.antimalarialicestimator.net>.
Z. Haghighijoo, O. Firuzi, B. Hemmateenejad, S. Emami, N. Edraki, R. Miri, *Bioorg. Chem.* **2017**, *74*, 126–133. [61]
[62]

