

(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
9 April 2009 (09.04.2009)

PCT

(10) International Publication Number
WO 2009/043902 A1

(51) International Patent Classification:
A61K 31/4184 (2006.01) A61K 45/06 (2006.01)
A61P 33/06 (2006.01)

(21) International Application Number:
PCT/EP2008/063224

(22) International Filing Date: 2 October 2008 (02.10.2008)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
07117987.3 5 October 2007 (05.10.2007) EP

(71) Applicant (for all designated States except US): **UNIVERSITE DE MONS HAINAUT** [BE/BE]; Place du Parc 20, B-7000 Mons (BE).

(72) Inventors; and

(75) Inventors/Applicants (for US only): **VANDEN EYNDE, Jean Jacques** [BE/BE]; 53, bd. Albert-Elisabeth, B-7000 Mons (BE). **PIETKA, Aurélie** [BE/BE]; Chemin Royal 18 B, B-7321 Blaton (BE). **HUANG, Tien** [US/US]; 573 Aris avenue, Metairie, LA 70005 (US). **MAYENCE, Annie** [BE/BE]; 53, bd. Albert-Elisabeth, B-7000 Mons (BE).

(74) Agents: **BRANTS, Johan, Philippe, Emile** et al.; De Clercq, Brants & Partners, E. Gevaertdreef 10a, B-9830 Sint-Martens-Latem (BE).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, NO, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Declaration under Rule 4.17:

— of inventorship (Rule 4.17(iv))

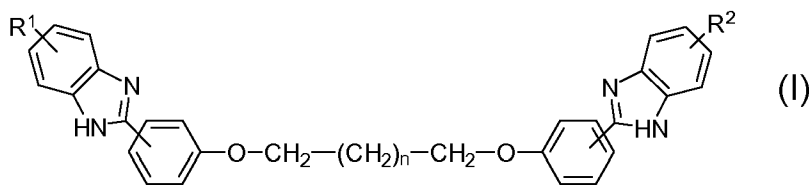
Published:

— with international search report
— before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments



WO 2009/043902 A1

(54) Title: BISBENZIMIDAZOLES AS ANTIMALARIAL AGENTS



(57) Abstract: The present invention relates to antimalarial agents. In particular, the present invention relates to the use of bisbenzimidazoles of Formula (I) for the prevention and/or treatment of malaria, wherein n is an integer selected from 0 to 10, and R¹ and R² are each independently selected from hydrogen, halogen, hydroxyl,

alkyl, alkoxy, amino, alkylamino, alkylcarbonylamino, alkylcarbonyloxy, formyl, alkylcarbonyl, alkyloxycarbonyl, halocarbonyl, haloalkyl, haloalkoxy, carbamoyl, cyano, nitro, sulfo, or a carboxyl group.

Bisbenzimidazoles as antimalarial agents

Field of the invention

The present invention relates to antimalarial agents. In particular, the present invention relates to bisbenzimidazoles for use in the treatment of malaria.

5 Background of the invention

Malaria is a parasitic disease which is caused by various species of Plasmodium protozoa. Together with AIDS and TB, malaria is responsible for largest number of deaths annually. The high rate of mortality associated with malaria can be attributed to the increasing cases of resistance of *Plasmodium falciparum*, the most deadly of the four
10 human infecting malarial parasites, to the contemporary antimalarial drugs. All attempts to develop vaccines against P. falciparum have failed so far. Therefore, therapies and preventive measures against malaria are confined to drugs. Various classes of antimalarial drugs exist. The most widely used are the quinoline antimalarials, e.g. chloroquine which has been an especially effective drug for both prophylaxis and therapy.
15 However, resistance to many of the currently available antimalarial drugs is spreading rapidly, threatening people in areas where malaria is endemic. Reports of multi-drug resistant strains of malaria parasites render the search for new antimalarial agents especially urgent.

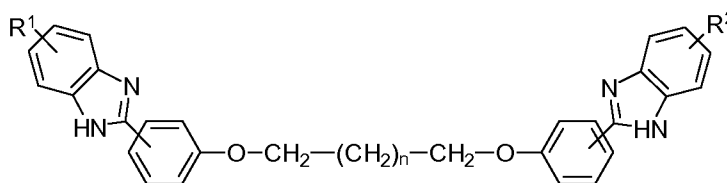
The limitations of the current antimalarial chemotherapeutic arsenal underscore the need
20 for new drugs in this therapeutic area.

It is the aim of the present invention to provide compounds effective in the treatment of malaria to overcome some of the above-mentioned drawbacks.

Summary of the invention

The present invention relates to bisbenzimidazoles which are useful in the treatment
25 and/or prevention of malaria, in particular Plasmodium falciparum malaria.

Accordingly, the present invention concerns a compound of formula I for use in the treatment of malaria. In particular, the invention concerns the use of at least one compound of Formula I or a pharmaceutically acceptable salt or solvate thereof for the preparation of a medicament for the prevention and/or the treatment of malaria,



I

wherein n is an integer selected from 0 to 10, and

R^1 and R^2 are each independently selected from hydrogen, halogen, hydroxyl, alkyl, alkoxy, amino, alkylamino, alkylcarbonylamino, alkylcarbonyloxy, formyl, alkylcarbonyl, alkyloxycarbonyl, halocarbonyl, haloalkyl, haloalkoxy, carbamoyl, cyano, nitro, sulfo, or a carboxyl group.

The present invention also relates to a method for the therapeutic and/or prophylactic treatment of malaria in a subject in need of such treatment comprising administering to the subject a therapeutically effective amount of a compound of formula I or a pharmaceutically acceptable salt or solvate thereof.

The present invention will now be further described. In the following passages, different aspects of the invention are defined in more detail. Each aspect so defined may be combined with any other aspect or aspects unless clearly indicated to the contrary. In particular, any feature indicated as being preferred or advantageous may be combined with any other feature or features indicated as being preferred or advantageous.

Detailed description

When describing the compounds for use in the invention, the terms used are to be construed in accordance with the following definitions, unless a context dictates otherwise.

The term "halo" or "halogen" as a group or part of a group is generic for fluoro, chloro, bromo, iodo.

The term "nitro" as used herein refers to the group $-NO_2$.

The term "cyano" as used herein refers to the group $-CN$.

The term "alkyl" by itself or as part of another substituent refers to a hydrocarbyl radical of Formula C_nH_{2n+1} wherein n is a number greater than or equal to 1. Generally, alkyl groups of this invention comprise from 1 to 24 carbon atoms, for example 1 to 20 carbon atoms, for example 1 to 18 carbon atoms, for example 1 to 12 carbon atoms, for example 1 to 8 carbon atoms, for example 1 to 6 carbon atoms, preferably from 1 to 4 carbon atoms, more preferably from 1 to 3 carbon atoms, still more preferably 1 to 2 carbon atoms. Alkyl

groups may be linear or branched and may be substituted as indicated herein. When a subscript is used herein following a carbon atom, the subscript refers to the number of carbon atoms that the named group may contain. Thus, for example, C₁₋₄alkyl means an alkyl of one to four carbon atoms. C₁₋₆alkyl includes all linear, or branched alkyl groups with between 1 and 6 carbon atoms, and thus includes methyl, ethyl, n-propyl, i-propyl, butyl and its isomers (e.g. n-butyl, i-butyl and t-butyl); pentyl and its isomers, hexyl and its isomers.

The term "haloalkyl" alone or in combination, refers to an alkyl radical having the meaning as defined above wherein one or more hydrogens are replaced with a halogen as defined above. Non-limiting examples of such haloalkyl radicals include chloromethyl, 1-bromoethyl, fluoromethyl, difluoromethyl, trifluoromethyl, 2,2,2-trifluoroethyl and the like.

The term "hydroxyl" or "hydroxy" as used herein refers to the group -OH.

The term "alkoxy" or "alkyloxy" as used herein refers to a radical having the Formula -OR^a wherein R^a is alkyl. Preferably, alkoxy is C₁₋₆ alkoxy, more preferably alkoxy is C₁₋₄ alkoxy.

Non-limiting examples of suitable alkoxy include methoxy, ethoxy, propoxy, isopropoxy, butoxy, isobutoxy, sec-butoxy, tert-butoxy, pentyloxy and hexyloxy. Where the oxygen atom in an alkoxy group is substituted with sulfur, the resultant radical is referred to as thioalkoxy.

The term "haloalkoxy" alone or in combination refers to a group of Formula -O-R^b wherein R^b is haloalkyl as defined herein. Non-limiting examples of suitable haloalkoxy include fluoromethoxy, difluoromethoxy, trifluoromethoxy, 2,2,2-trifluoroethoxy, 1,1,2,2-tetrafluoroethoxy, 2-fluoroethoxy, 2-chloroethoxy, 2,2-difluoroethoxy, 2,2,2-trichloroethoxy, trichloromethoxy, 2-bromoethoxy, pentafluoroethyl, 3,3,3-trichloropropoxy, 4,4,4-trichlorobutoxy.

The term "alkylcarbonyl" refers to a group of formula -COR^c, wherein R^c is C₁₋₆alkyl. Said alkylcarbonyl can be exemplified by acetyl, propionyl, butyryl, valeryl and pivaloyl.

The term "amino" refers to the group -NH₂.

The term "alkylamino" by itself or as part of another substituent refers to a group consisting of an amino group attached to one or two independently selected and optionally substituted alkyl groups, i.e., alkyl amino refers to -N(R^h)(Rⁱ) wherein R^h and Rⁱ are each independently selected from hydrogen or alkyl. Alkylamino includes mono-lower alkyl amino group (e.g. mono-C₁₋₆alkylamino group such as methylamino and ethylamino), di-lower alkylamino group (e.g. di-C₁₋₆alkylamino group such as dimethylamino and diethylamino). Non-limiting examples of suitable alkylamino groups also include n-

propylamino, isopropylamino, n-butylamino, isobutylamino, sec-butylamino, tert-butylamino, pentylamino, n-hexylamino, di-n-propylamino, diisopropylamino, ethylmethylamino, methyl-n-propylamino, methyl-i-propylamino, n-butylmethylamino, i-butylmethylamino, t-butylmethylamino, ethyl-n-propylamino, ethyl-i-propylamino, n-butylethylamino, i-butylethylamino, t-butylethylamino, di-n-butylamino, di-i-butylamino, methylpentylamino, methylhexylamino, ethylpentylamino, ethylhexylamino, propylpentylamino, propylhexylamino, and the like.

The term "carboxy" or "carboxyl" refers to the group $-\text{CO}_2\text{H}$. Thus, a carboxyalkyl is an alkyl group as defined above having at least one substituent that is $-\text{CO}_2\text{H}$.

10 The term "alkoxycarbonyl" or "alkyloxycarbonyl" refers to a carboxy group linked to an alkyl radical i.e. to form $-\text{C}(=\text{O})\text{OR}^d$, wherein R^d is alkyl as defined above.

The term "alkylcarbonyloxy" refers to a group of Formula $-\text{O}-\text{C}(=\text{O})\text{R}^d$ wherein R^d is alkyl as defined above.

15 The term "alkylcarbonylamino" refers to a group of Formula $-\text{NH}(\text{C}=\text{O})\text{R}^m$ or $-\text{NR}^o(\text{C}=\text{O})\text{R}^m$, wherein R^m and R^o are each independently alkyl or substituted alkyl.

The term "carbamoyl" (aminocarbonyl) refers to the group $-(\text{C}=\text{O})-\text{NH}_2$.

20 The term "alkylcarbamoyl" refers to a group of Formula $-(\text{C}=\text{O})-\text{NR}^k\text{R}^m$ wherein R^k is hydrogen or alkyl or substituted alkyl as defined herein, and R^m is alkyl or substituted alkyl as defined herein. The term encompasses "N-alkylcarbamoyl" wherein R^k is hydrogen and R^m is alkyl or substituted alkyl, and "N,N-dialkylcarbamoyl" wherein R^k and R^m are each independently alkyl or substituted alkyl.

The term "sulfo" or "sulfonic acid" refers to a group $-\text{SO}_3\text{H}$.

25 The term "optionally substituted alkyl" refers to an alkyl group optionally substituted with one or more substituent(s) (for example 1, 2, 3 or 4 substituent(s) at any available point of attachment). Unless stated otherwise, non-limiting examples of suitable substituents include halogen, hydroxyl, oxo, nitro, amido, carboxy, amino, cyano, haloalkoxy, and haloalkyl.

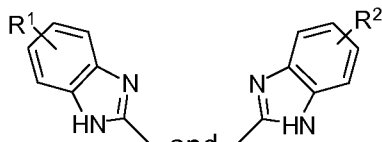
30 The present invention includes a method of treating or preventing malaria comprising administering to a subject in need thereof an antimalarial effective amount of a compound of Formula I or a pharmaceutically acceptable salt, or solvate thereof.

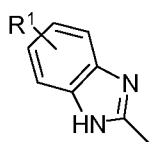
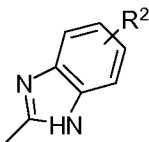
In a particular embodiment, the invention concerns the use of at least one compound of Formula I or a pharmaceutically acceptable salt or solvate thereof for the preparation of a medicament for the prevention and/or the treatment of malaria, wherein n is an integer

selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10, and R¹ and R² are each independently selected from hydrogen, halogen, hydroxyl, C₁₋₆alkyl, C₁₋₂₄alkoxy, amino, C₁₋₂₄alkylamino, C₁₋₂₄alkylcarbonylamino, C₁₋₂₄alkylcarbonyloxy, formyl, C₁₋₂₄alkylcarbonyl, C₁₋₂₄alkyloxycarbonyl, halocarbonyl, haloC₁₋₆alkyl, carbamoyl, C₁₋₂₄alkylcarbamoyl, cyano, nitro, sulfo, or a carboxyl group. In an embodiment, n is 0, 1, 2, 3, 4, 5, 6 or 7.

Preferably, the invention concerns the use of at least one compound of Formula I or a pharmaceutically acceptable salt or solvate thereof for the prevention and/or the treatment of malaria, wherein n is an integer selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10, and R¹ and R² are each independently selected from hydrogen, halogen, hydroxyl, C₁₋₄alkyl, C₁₋₁₈alkoxy, amino, C₁₋₁₈alkylamino, C₁₋₁₈alkylcarbonylamino, C₁₋₁₈alkylcarbonyloxy, formyl, C₁₋₁₈alkylcarbonyl, C₁₋₁₈alkyloxycarbonyl, halocarbonyl, haloC₁₋₄alkyl, carbamoyl, C₁₋₁₈alkylcarbamoyl, cyano, nitro, sulfo, or a carboxyl group, preferably, R¹ and R² are each independently selected from hydrogen, halogen, hydroxyl, C₁₋₄alkyl, C₁₋₁₂alkoxy, amino, C₁₋₁₂alkylamino, C₁₋₁₂alkylcarbonylamino, C₁₋₁₂alkylcarbonyloxy, formyl, C₁₋₁₂alkylcarbonyl, C₁₋₁₂alkyloxycarbonyl, halocarbonyl, haloC₁₋₄alkyl, carbamoyl, C₁₋₁₂alkylcarbamoyl, cyano, nitro, sulfo, or a carboxyl group and n is 0, 1, 2, 3, 4, 5, 6 or 7.

In another embodiment, R¹ and R² are each independently selected from hydrogen, hydroxyl, halogen, alkyl, alkoxy, haloalkyl, haloalkoxy, alkyloxycarbonyl, alkylcarbonyl, amino, alkylamino, cyano, nitro or a carboxyl group.



The groups of formula  and  can be independently linked in para, ortho or meta position on the phenyl moiety with respect to the ether bond. In an embodiment, both groups are in para. In another embodiment, both groups are in ortho. In yet another embodiment, both groups are in meta. Preferably both groups are in para, and n is selected from 1, 2, 3, 4, 5, 6 or 7.

Non-limiting examples of suitable compounds for use in the present invention can be selected from the group comprising 2,2' - [1,3-propanediylbis(oxy-1,4-phenylene)]bis-1H-benzimidazole, 2,2' - [1,4-butanediylbis(oxy-1,4-phenylene)]bis-1H-benzimidazole, 2,2' - [1,5-pentanediybis(oxy-1,4-phenylene)]bis-1H-benzimidazole, 2,2' - [1,6-hexanediybis(oxy-1,4-phenylene)]bis-1H-benzimidazole, 2,2' - [1,7-heptanediybis(oxy-1,4-phenylene)]bis-1H-benzimidazole, 2,2' - [1,5-pentanediybis(oxy-1,3-phenylene)]bis-1H-benzimidazole, 2,2' - [1,5-pentanediybis(oxy-1,2-phenylene)]bis-1H-benzimidazole, 2,2'-[1,5-pentanediybis(oxy-1,4-phenylene)]-1H-benzimidazole-5-carboxylic acid; 2,2'-

	[1,2-ethanediylbis(oxy-1,4-phenylene)]bis-5-nitro-1H-benzimidazole;	2,2'-[1,2-
	ethanediylbis(oxy-1,4-phenylene)]bis-5-chloro-1H-benzimidazole;	2,2'-[1,3-
	propanediylbis(oxy-1,4-phenylene)]bis-5-hydroxy-1H-benzimidazole;	2,2'-[1,7-
	heptanediylbis(oxy-1,4-phenylene)]bis-5-methoxy-1H-benzimidazole;	2,2'-[1,7-
5	heptanediylbis(oxy-1,3-phenylene)]bis-5-methyl-1H-benzimidazole;	2,2'-[1,5-
	pentanediylbis(oxy-1,3-phenylene)]bis-5-amino-1H-benzimidazole;	2,2'-[1,5-
	pentanediylbis(oxy-1,3-phenylene)]bis-5-cyano-1H-benzimidazole;	2,2' – [1,5-
	pentanediylbis(oxy-1,4-phenylene)]bis-5-trifluoromethoxy-1H-benzimidazole;	2,2' – [1,3-
	propanediylbis(oxy-1,2-phenylene)]bis-5-tertbutyl-1H-benzimidazole;	2,2' – [1,3-
10	pentanediylbis(oxy-1,3-phenylene)]bis-5-tertbutyl-1H-benzimidazole,	2,2'-[1,3-
	propanediylbis(oxy-1,2-phenylene)]bis-5-bromo-1H-benzimidazole,	2,2'-[1,4-
	butanediylbis(oxy-1,3-phenylene)]bis-5-bromo-1H-benzimidazole,	2,2'-[1,4-
	butanediylbis(oxy-1,3-phenylene)]bis-5-diethylamino-1H-benzimidazole,	2,2'-[1,2-
	ethanediylbis(oxy-1,4-phenylene)]bis-5-propoxy-1H-benzimidazole,	diethyl 2,2'-[1,2-
15	ethanediylbis(oxy-1,4-phenylene)]bis-1H-benzimidazole-5-carboxylate,	2,2'-[1,7-
	heptanediylbis(oxy-1,3-phenylene)]bis-5-dodecoxy-1H-benzimidazole,	dioctyl 2,2'-[1,5-
	pentanediylbis(oxy-1,2-phenylene)]bis-1H-benzimidazole-5-carboxylate.	

In an embodiment, the compound of formula I can be used in combination with at least one additional drug selected from the group consisting of chloroquine, artesinin, qinghaosu, 8-aminoquinoline, amodiaquine, arteether, artemether, artemisinin, artesunate, artesunic acid, artelinic acid, atovoquone, azithromycine, biguanide, chloroquine phosphate, chlorproguanil, cycloguanil, dapsone, desbutyl halofantrine, desipramine, doxycycline, dihydrofolate reductase inhibitors, dipyrindamole, halofantrine, haloperidol, hydroxychloroquine sulfate, imipramine, mefloquine, penfluridol, phospholipid inhibitors, primaquine, proguanil, pyrimethamine, pyronaridine, quinine, quinidine, quinacrineartemisinin, sulfonamides, sulfones, sulfadoxine, sulfalene, tafenoquine, tetracycline, tetrandine, triazine, salts or mixture thereof.

In an embodiment, the present invention concerns the use of compounds of formula I, for the treatment of drug-resistant malaria.

30 Preferably, said malaria is selected from *Plasmodium falciparum* malaria, *P. vivax* malaria, *P. ovale* malaria, or *P. malariae* malaria.

It was found by the invention that the compounds as defined above are particularly useful to prevent or treat an infection of a malarial parasite in a subject and/or for preventing, treating and/or alleviating complications and/or symptoms associated therewith and can

then be used in the preparation of a medicament for the treatment and/or prevention of such disease.

The method of the invention comprises administering to a subject in need of such treatment a therapeutically effective amount of a compound of formula I or a
5 pharmaceutically acceptable salt or solvate thereof.

The term "malaria" as used herein refers to an infectious disease, also known as ague or marsh fever, typically caused by a protistan parasite of the genus *Plasmodium*, suitably, *P. falciparum*, *P. vivax*, *P. ovale* or *P. malariae*. This parasite is transmitted primarily by female *Anopheles* mosquitoes. *Plasmodium* invades and consumes the red blood cells of
10 its hosts, which leads to symptoms including fever, anemia, and in severe cases, a coma potentially leading to death.

In an embodiment, the method comprises treating a subject that has been infected with *Plasmodium falciparum*. In an embodiment, the method comprises treating a subject that has been infected with *P. vivax*. In an embodiment, the method comprises treating a
15 subject that has been infected with *P. ovale*. In an embodiment, the method comprises treating a subject that has been infected with *P. malariae*.

In an embodiment, the compound of Formula I is administered after the subject has been exposed to the malaria parasite. In another embodiment, the compound of Formula I is administered before the subject travels to a country where malaria is endemic.

20 In another embodiment, the malaria parasite is a drug-resistant malarial strain.

The term a "therapeutically effective amount", "effective amount" or a "sufficient amount" of a compound of Formula I is a quantity sufficient to, when administered to the subject, including a mammal, for example a human, effect beneficial or desired results, including clinical results, and, as such, an "effective amount" or synonym thereto depends upon the
25 context in which it is being applied.

In the context of disease, therapeutically effective amounts of the compounds of Formula I are used to treat, modulate, attenuate, reverse, or effect malaria in a mammal. An "effective amount" is intended to mean that amount of a compound that is sufficient to treat, prevent or inhibit malaria or a disease associated with malaria. In some suitable
30 embodiments, malaria or the disease or disorder associated with malaria is caused by a *Plasmodium* parasite, suitably, *P. falciparum*, *P. vivax*, *P. ovale* or *P. malariae*, thus it is the amount sufficient to, when administered to the subject, including a mammal, e.g., a human, to treat, prevent or inhibit malaria or a disease or a disorder associated with malaria or a malarial parasite, e.g. *Plasmodium* parasite. The amount of a given

compound that will correspond to such an amount will vary depending upon various factors, such as the given drug or compound, the pharmaceutical formulation, the route of administration, the type of disease or disorder, the identity of the subject or host being treated, and the like, but can nevertheless be routinely determined by one skilled in the art. Also, as used herein, a "therapeutically effective amount" of a compound of Formula I is an amount which prevents, inhibits, suppresses or reduces malaria (e.g., as determined by clinical symptoms or the amount of malarial parasites, e.g., Plasmodium organisms) in a subject as compared to a control. As defined herein, a therapeutically effective amount of a compound of Formula I may be readily determined by one of ordinary skill by routine methods known in the art. The skilled artisan will appreciate that certain factors may influence the dosage required to effectively treat a subject, or prevent a subject, from being afflicted with malaria and these factors include, but are not limited to, the severity of the disease or disorder, previous treatments, the general health and/or age of the subject and other diseases present.

Moreover, a "treatment" or "prevention" regime of a subject with a therapeutically effective amount of the compound of Formula I may consist of a single administration, or alternatively comprise a series of applications. For example, the compound of Formula I may be administered at least once a week. However, in another embodiment, the compound of Formula I may be administered to the patient from about one time per week to about once daily for a given treatment. The length of the treatment period depends on a variety of factors, such as the severity of the disease, the age of the patient, the concentration and the activity of the compounds of Formula I, or a combination thereof. It will also be appreciated that the effective dosage of the compound used for the treatment or prophylaxis may increase or decrease over the course of a particular treatment or prophylaxis regime. Changes in dosage may result and become apparent by standard diagnostic assays known in the art. In some instances, chronic administration may be required. The compounds of Formula I may be administered before, during or after exposure to malaria or malarial parasite, e.g. Plasmodium parasite.

As used herein, and as well understood in the art, "treatment" is an approach for obtaining beneficial or desired results, including clinical results. Beneficial or desired clinical results can include, but are not limited to, alleviation or amelioration of one or more symptoms or conditions, diminishment of extent of disease, stabilized (i.e. not worsening) state of disease, preventing spread of disease, delay or slowing of disease progression, amelioration or palliation of the disease state, and remission (whether partial or total),

whether detectable or undetectable. "Treatment" can also mean prolonging survival as compared to expected survival if not receiving treatment.

The term "prevention" or "prophylaxis", or synonym thereto, as used herein refers to a reduction in the risk or probability of a patient becoming afflicted with malaria or manifesting a symptom associated with malaria.

The term "subject" as used herein refers to a mammal. The individual will preferably be a human, but may also be a domestic livestock, laboratory or pet animal.

As used in the specification and the appended claims, the singular forms "a", "an," and "the" include plural referents unless the context clearly dictates otherwise. By way of example, "a compound" means one compound or more than one compound.

Whenever used in the present invention the term "compounds for use in the invention" or a similar term is meant to include the compounds of general Formula I, N-oxides, salts, solvates, hydrates, tautomeric forms, analogues, pro-drugs, esters and metabolites, as well as their quaternized nitrogen analogues. The N-oxide forms of said compounds are meant to comprise compounds wherein one or several nitrogen atoms are oxidized to the so-called N-oxide.

The compounds of the invention may be in the form of pharmaceutically acceptable salts, as generally described below. Some preferred, but non-limiting examples of suitable pharmaceutically acceptable organic and/or inorganic acids are as hydrochloric acid, hydrobromic acid, sulfuric acid, nitric acid, acetic acid and citric acid, as well as other pharmaceutically acceptable acids known per se (for which reference is made to the prior art referred to below).

When the compounds of the invention contain an acidic group as well as a basic group the compounds of the invention may also form internal salts, and such compounds are within the scope of the invention. When the compounds of the invention contain a hydrogen-donating heteroatom (e.g. NH), the invention also covers salts and/or isomers formed by transfer of said hydrogen atom to a basic group or atom within the molecule.

Pharmaceutically acceptable salts of the compounds of formula I include the acid addition and base salts thereof. Suitable acid addition salts are formed from acids which form non-toxic salts. Examples include the acetate, adipate, aspartate, benzoate, besylate, bicarbonate/carbonate, bisulphate/sulphate, borate, camsylate, citrate, cyclamate, edisylate, esylate, formate, fumarate, gluceptate, gluconate, glucuronate, hexafluorophosphate, hibenzate, hydrochloride/chloride, hydrobromide/bromide, hydroiodide/iodide, isethionate, lactate, malate, maleate, malonate, mesylate,

methysulphate, naphthylate, 2-napsylate, nicotinate, nitrate, orotate, oxalate, palmitate, pamoate, phosphate/hydrogen phosphate/dihydrogen phosphate, pyroglutamate, saccharate, stearate, succinate, tannate, tartrate, tosylate, trifluoroacetate and xinofoate salts. Suitable base salts are formed from bases which form non-toxic salts. Examples
5 include the aluminium, arginine, benzathine, calcium, choline, diethylamine, diolamine, glycine, lysine, magnesium, meglumine, olamine, potassium, sodium, tromethamine and zinc salts. Hemisalts of acids and bases may also be formed, for example, hemisulphate and hemicalcium salts. For a review on suitable salts, see Handbook of Pharmaceutical
10 Salts: Properties, Selection, and Use by Stahl and Wermuth (Wiley-VCH, 2002), incorporated herein by reference.

Pharmaceutical composition for use in the present invention preferably comprises an effective amount of a compound of formula I and a suitable pharmaceutical acceptable carrier.

The preparations may be prepared in a manner known per se, which usually involves
15 mixing the at least one compound according to the invention with the one or more pharmaceutically acceptable carriers, and, if desired, in combination with other pharmaceutical active compounds, when necessary under aseptic conditions. Reference is again made to US-A-6,372,778, US-A-6,369,086, US-A-6,369,087 and US-A-6,372,733 and the further prior art mentioned above, as well as to the standard handbooks, such as
20 the latest edition of Remington's Pharmaceutical Sciences.

Generally, for pharmaceutical use, the compounds may be formulated as a pharmaceutical preparation comprising at least one compound of formula I and at least one pharmaceutically acceptable carrier, diluent or excipient and/or adjuvant, and optionally one or more further pharmaceutically active compounds.

25 The pharmaceutical preparations of the invention are preferably in a unit dosage form, and may be suitably packaged, for example in a box, blister, vial, bottle, sachet, ampoule or in any other suitable single-dose or multi-dose holder or container (which may be properly labeled); optionally with one or more leaflets containing product information and/or instructions for use. Generally, such unit dosages will contain between 1 and 1000
30 mg, and usually between 5 and 500 mg, of the at least one compound of the invention, e.g. about 10, 25, 50, 100, 200, 300 or 400 mg per unit dosage.

The compounds can be administered by a variety of routes including the oral, ocular, rectal, transdermal, subcutaneous, intravenous, intramuscular or intranasal routes, depending mainly on the specific preparation used. The compound of formula I will

generally be administered in an "effective amount", by which is meant any amount of a compound of the Formula I that, upon suitable administration, is sufficient to achieve the desired therapeutic or prophylactic effect in the subject to which it is administered. Usually, depending on the condition to be prevented or treated and the route of administration, such an effective amount will usually be between 0.01 to 1000 mg per kilogram body weight of the patient per day, more often between 0.1 and 500 mg, such as between 1 and 250 mg, for example about 5, 10, 20, 50, 100, 150, 200 or 250 mg, per kilogram body weight of the patient per day, which may be administered as a single daily dose, divided over one or more daily doses. The amount(s) to be administered, the route of administration and the further treatment regimen may be determined by the treating clinician, depending on factors such as the age, gender and general condition of the patient and the nature and severity of the disease/symptoms to be treated. Reference is again made to US-A-6,372,778, US-A-6,369,086, US-A-6,369,087 and US-A-6,372,733 and the further prior art mentioned above, as well as to the standard handbooks, such as the latest edition of Remington's Pharmaceutical Sciences.

For an oral administration form, the compound of formula I can be mixed with suitable additives, such as excipients, stabilizers or inert diluents, and brought by means of the customary methods into the suitable administration forms, such as tablets, coated tablets, hard capsules, aqueous, alcoholic, or oily solutions. Examples of suitable inert carriers are gum arabic, magnesia, magnesium carbonate, potassium phosphate, lactose, glucose, or starch, in particular, corn starch. In this case, the preparation can be carried out both as dry and as moist granules. Suitable oily excipients or solvents are vegetable or animal oils, such as sunflower oil or cod liver oil. Suitable solvents for aqueous or alcoholic solutions are water, ethanol, sugar solutions, or mixtures thereof. Polyethylene glycols and polypropylene glycols are also useful as further auxiliaries for other administration forms. As immediate release tablets, these compositions may contain microcrystalline cellulose, dicalcium phosphate, starch, magnesium stearate and lactose and/or other excipients, binders, extenders, disintegrants, diluents and lubricants known in the art.

When administered by nasal aerosol or inhalation, the compositions may be prepared according to techniques well-known in the art of pharmaceutical formulation and may be prepared as solutions in saline, employing benzyl alcohol or other suitable preservatives, absorption promoters to enhance bioavailability, fluorocarbons, and/or other solubilizing or dispersing agents known in the art. Suitable pharmaceutical formulations for administration in the form of aerosols or sprays are, for example, solutions, suspensions

or emulsions of the compounds of the invention or their physiologically tolerable salts in a pharmaceutically acceptable solvent, such as ethanol or water, or a mixture of such solvents. If required, the formulation can also additionally contain other pharmaceutical auxiliaries such as surfactants, emulsifiers and stabilizers as well as a propellant.

5 For subcutaneous or intravenous administration, the compounds of formula I, if desired with the substances customary therefore such as solubilizers, emulsifiers or further auxiliaries are brought into solution, suspension, or emulsion. The compounds of formula I can also be lyophilized and the lyophilizates obtained used, for example, for the production of injection or infusion preparations. Suitable solvents are, for example, water,
10 physiological saline solution or alcohols, e.g. ethanol, propanol, glycerol, in addition also sugar solutions such as glucose or mannitol solutions, or alternatively mixtures of the various solvents mentioned. The injectable solutions or suspensions may be formulated according to known art, using suitable non-toxic, parenterally-acceptable diluents or solvents, such as mannitol, 1,3-butanediol, water, Ringer's solution or isotonic sodium
15 chloride solution, or suitable dispersing or wetting and suspending agents, such as sterile, bland, fixed oils, including synthetic mono- or diglycerides, and fatty acids, including oleic acid.

When rectally administered in the form of suppositories, the formulations may be prepared by mixing the compounds of formula I with a suitable non-irritating excipient,
20 such as cocoa butter, synthetic glyceride esters or polyethylene glycols, which are solid at ordinary temperatures, but liquefy and/or dissolve in the rectal cavity to release the drug.

The compounds may be formulated for local effect, for instance topical or non-adsorbent applications.

The compounds of formula I or the above-mentioned pharmaceutical compositions may
25 also be used in combination with one or more other therapeutically useful substances selected from the group comprising antimalarials like quinolines (quinine, chloroquine, amodiaquine, mefloquine, primaquine, tafenoquine); peroxide antimalarials (artemisinin, artemether, artesunate); pyrimethamine-sulfadoxine antimalarials (e.g. Fansidar); hydroxynaphtoquinones (e.g. atovaquone); acroline-type antimalarials (e.g. pyronaridine);
30 and antiprotozoal agents such as ethylstilbamidine, hydroxystilbamidine, pentamidine, stilbamidine, quinapyramine, puromycine, propamidine, nifurtimox, melarsoprol, nimorazole, nifuroxime, aminitroazole and the like.

In particular, the present invention also concerns a method of treating or preventing malaria comprising administering to a subject in need thereof an anti-malarial effective

amount compound selected from a compound of Formula I, pharmaceutically acceptable salts, solvates thereof, in combination with at least one additional drug selected from the group consisting of chloroquine, artemesin, qinghaosu, 8-aminoquinoline, amodiaquine, arteether, artemether, artemisinin, artesunate, artesunic acid, artelinic acid, atovoquone, azithromycine, biguanide, chloroquine phosphate, chlorproguanil, cycloguanil, dapson, 5 desbutyl halofantrine, desipramine, doxycycline, dihydrofolate reductase inhibitors, dipyridamole, halofantrine, haloperidol, hydroxychloroquine sulfate, imipramine, mefloquine, penfluridol, phospholipid inhibitors, primaquine, proguanil, pyrimethamine, pyronaridine, quinine, quinidine, quinacrineartemisinin, sulfonamides, sulfones, 10 sulfadoxine, sulfalene, tafenoquine, tetracycline, tetrandine, triazine or salts thereof.

Suitable method comprises the step of exposing malaria cells to an effective concentration of a compound of the formula I, in combination with at least one additional drug selected from the group described above.

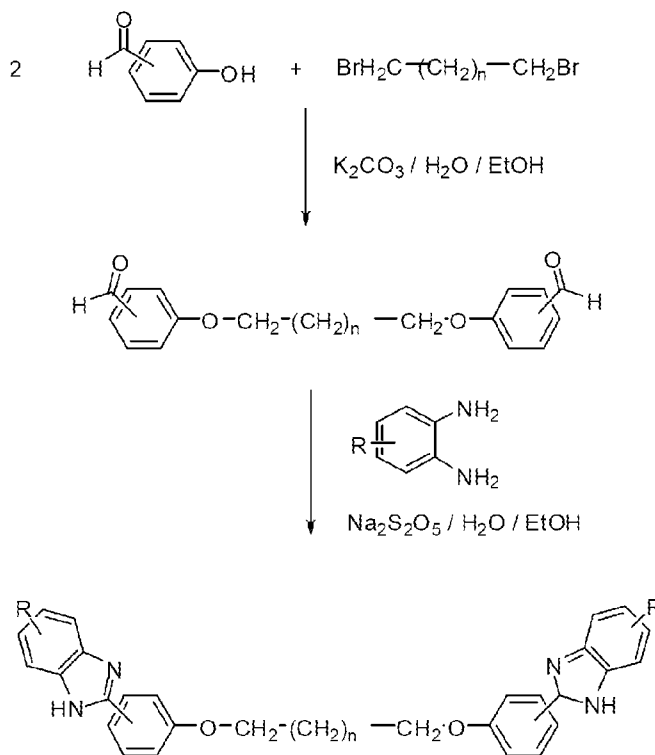
The present invention also concerns a composition for preventing or treating malaria 15 comprising: a compound of the formula I in combination with at least one additional drug selected from the group consisting of chloroquine, artemesin, qinghaosu, 8-aminoquinoline, amodiaquine, arteether, artemether, artemisinin, artesunate, artesunic acid, artelinic acid, atovoquone, azithromycine, biguanide, chloroquine, chloroquine phosphate, chlorproguanil, cycloguanil, dapson, desbutyl halofantrine, desipramine, 20 doxycycline, dihydrofolate reductase inhibitors, dipyridamole, halofantrine, haloperidol, hydroxychloroquine sulfate, imipramine, mefloquine, penfluridol, phospholipid inhibitors, primaquine, proguanil, pyrimethamine, pyronaridine, quinine, quinidine, quinacrineartemisinin, sulfonamides, sulfones, sulfadoxine, sulfalene, tafenoquine, tetracycline, tetrandine, triazine or derivatives or mixture thereof.

25 It was found by the invention that the compounds as defined above are particularly useful to prevent or treat an infection of a malarial parasite in a subject and/or for preventing, treating and/or alleviating complications and/or symptoms associated therewith and can then be used in the preparation of a medicament for the treatment and/or prevention of such disease.

30 The invention will now be illustrated by means of the following synthetic and biological examples, which do not limit the scope of the invention in any way.

Examples

The following general procedures and intermediates were used to prepare the compounds described herein. Compounds for use in the present invention can be prepared as described in Scheme 1.



5

Scheme 1

with $n = 0$ to 10 , and each R is independently selected from hydrogen, halogen, hydroxyl, alkyl, alkoxy, amino, alkylamino, alkylcarbonylamino, alkylcarbonyloxy, formyl, alkylcarbonyl, alkyloxycarbonyl, halocarbonyl, haloalkyl, haloalkoxy, carbamoyl, cyano, nitro, sulfo, or a carboxyl group.

10

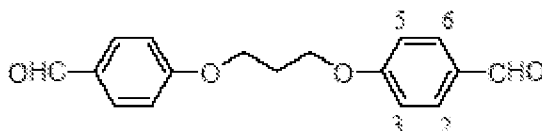
For example, each R is independently selected from H , $-\text{CO}_2\text{H}$, Cl , F , Br , $-\text{OH}$, $-\text{Me}$, $-\text{Et}$, $-\text{nPr}$, $-\text{iPr}$, tBu , $-\text{OMe}$, $-\text{OEt}$, $-\text{OnPr}$, $-\text{OiPr}$, $-\text{OnBu}$, $-\text{OiBu}$, $-\text{OtBu}$, $-\text{O}-\text{C}_5\text{H}_{11}$, $-\text{O}-\text{C}_6\text{H}_{13}$, $-\text{O}-\text{C}_7\text{H}_{15}$, $-\text{O}-\text{C}_8\text{H}_{17}$, $-\text{O}-\text{C}_9\text{H}_{19}$, $-\text{O}-\text{C}_{10}\text{H}_{21}$, $-\text{O}-\text{C}_{11}\text{H}_{23}$, $-\text{O}-\text{C}_{12}\text{H}_{25}$, $-\text{O}-\text{C}_{13}\text{H}_{27}$, $-\text{O}-\text{C}_{14}\text{H}_{29}$, ..., $-\text{OC}_{18}\text{H}_{37}$, ... $-\text{OC}_{24}\text{H}_{49}$, $-\text{NH}_2$, $-\text{NH}(\text{Me})$, $-\text{N}(\text{Me})_2$, $-\text{N}(\text{Et})_2$, $-\text{NH}-\text{CO}-\text{Me}$, $-\text{NH}-\text{CO}-\text{Et}$, $-\text{OCOMe}$, $-\text{OCOEt}$, $-\text{OCOnPr}$, $-\text{OCOiPr}$, $-\text{OCOnBu}$, $-\text{OCOiBu}$, $-\text{OCOtBu}$, $-\text{OCO}-\text{C}_5\text{H}_{11}$, $-\text{OCO}-\text{C}_6\text{H}_{13}$, $-\text{OCO}-\text{C}_7\text{H}_{15}$, $-\text{OCO}-\text{C}_8\text{H}_{17}$, $-\text{OCO}-\text{C}_9\text{H}_{19}$, $-\text{OCO}-\text{C}_{10}\text{H}_{21}$, $-\text{OCO}-\text{C}_{11}\text{H}_{23}$, $-\text{OCO}-\text{C}_{12}\text{H}_{25}$, $-\text{OCO}-\text{C}_{13}\text{H}_{27}$, $-\text{OCO}-\text{C}_{14}\text{H}_{29}$, ..., $-\text{OCOC}_{18}\text{H}_{37}$, ... $-\text{OCOC}_{24}\text{H}_{49}$, $-\text{CHO}$, $-\text{COMe}$, $-\text{COEt}$, $-\text{CO}_2\text{Me}$, $-\text{CO}_2\text{Et}$, $-\text{COCl}$, $-\text{CF}_3$, $-\text{CCl}_3$, $-\text{CHF}_2$, $-\text{OCF}_3$, $-\text{CONH}_2$, $-\text{CN}$, $-\text{NO}_2$, $-\text{SO}_3\text{H}$.

15

General procedure

A mixture of an hydroxybenzaldehyde (5.12 g; 42 mmol), a dibromoalkane (20 mmol), and potassium carbonate (2.76 g; 20 mmol) in ethanol (10 ml) was heated under reflux for 8 hours. After cooling, the precipitate was filtered and successively washed with water,
5 ethanol and ether. The bisbenzaldehyde intermediate was pure enough to be engaged in the second step.

A mixture of the bisbenzaldehyde intermediate (3 mmol), sodium pyrosulfite (0.76 g; 4 mmol), an ortho- phenylenediamine (6.5 mmol), and water (3 ml) in ethanol (9 ml) was irradiated in a professional microwave oven for 15 min at 140 °C. After cooling the
10 precipitate was filtered and thoroughly washed with water, ethanol, and ether and dried to obtain the compound for use in the invention.

Intermediates**Intermediate 1: 4,4'-[1,3-propanediyl(oxy)]bisbenzaldehyde**

15 IR (KBr, cm⁻¹): 2952, 1683, 1606, 1578, 1510.

¹H NMR (DMSO-d₆, ppm): 9.9 (s, 2H, CHO); 7.8 (d, J=7Hz, 4H, C₆H₄:H₂,H₆); 7.2 (d, 4H, J=7Hz, C₆H₄:H₃,H₅); 4.3 (t, 4H, J=6Hz, O-CH₂-CH₂); 2.3 (m, 2H, J=6Hz, O-CH₂-CH₂).

Mp (°C): 193.2- 196.3 °C. Yield: 78% MW: 284.31.

Intermediate 2: 4,4'-[1,4-butanediyl(oxy)]bisbenzaldehyde

20

IR (KBr, cm⁻¹): 2943, 1697, 1601, 1580, 1510.

¹H NMR (DMSO-d₆, ppm): 9.9 (s, 2H, CHO); 7.9 (d, 4H, J=6Hz, C₆H₄: H₂,H₆); 7.1 (d, 4H, J=6Hz, C₆H₄:H₃,H₅); 4.2 (t, 4H, J=6Hz, O-CH₂-CH₂); 1.9 (m, 4H, O-CH₂-CH₂).

Mp (°C): 153- 158°C. Yield: 70% MW: 298.33.

25 **Intermediate 3: 4,4'-[1,5-pentanediyloxy]bisbenzaldehyde**



IR (KBr, cm⁻¹): 2945, 1692, 1604, 1576, 1510.

¹H NMR (DMSO-d₆, ppm): 9.9 (s, 2H, CHO); 7.9 (d, 4H, J=8Hz, C₆H₄:H₂,H₆); 7.1 (d, 4H, J= 8Hz, C₆H₄:H₃,H₅); 4.2 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.5 (m, 2H, J= 6Hz, O-CH₂-CH₂-CH₂).

5 Mp (°C): 90.7- 94.5°C. Yield: 90% MW : 312.36.

Intermediate 4: 4,4'-[1,6-hexanediyl(oxy)]bisbenzaldehyde



IR (KBr, cm⁻¹): 2946, 2876, 1686, 1599, 1511.

10 ¹H NMR (DMSO-d₆, ppm): 9.9 (s, 2H, CHO); 7.9 (d, 4H, J=9Hz, C₆H₄:H₂,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄:H₃,H₅); 4.1 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, O-CH₂-CH₂-CH₂); 1.5 (m, 4H, O-CH₂-CH₂-CH₂).

Mp (°C): 111- 116°C. Yield: 84% MW: 326.39.

Intermediate 5: 4,4'-[1,7-heptane(oxy)]bisbenzaldehyde

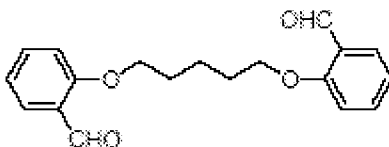


15 IR (KBr, cm⁻¹): 2945, 1686, 1599, 1573, 1510.

¹H NMR (DMSO-d₆, ppm): 9.9 (s, 2H, CHO); 7.9 (d, 4H, J=9Hz, C₆H₄:H₂,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄:H₃,H₅); 4.1 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, O-CH₂-CH₂-CH₂); 1.4 (m, 2H+4H, O-CH₂-CH₂-CH₂).

Mp (°C): 63.8- 65.3°C. Yield: 59% MW: 340.41.

20 **Intermediate 6: 2,2'-[1,5-pentanediyloxy]bisbenzaldehyde**



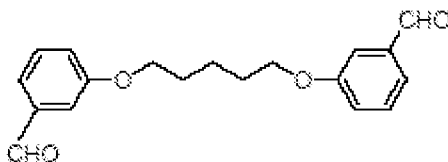
IR (KBr, cm⁻¹): 2947, 2874, 1678, 1601, 1459.

¹H NMR (DMSO-d₆, ppm): 10.4 (s, 2H, CHO); 7.7 (d, 2H, J=8Hz, C₆H₄: H₆); 7.6 (d, 2H, J=8Hz, C₆H₄: H₄); 7.2 (d, 2H, J=8Hz, C₆H₄: H₃); 7.1 (t, 2H, J=8Hz, C₆H₄: H₅); 4.2 (t,

4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.9 (m, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.7 (m, 2H, J=6Hz, O-CH₂-CH₂-CH₂).

Mp (°C): 65.8- 70°C. Yield: 79% MW: 312.36.

Intermediate 7: 3,3'-[1,5-pentanediyloxy]bisbenzaldehyde



5

IR (KBr, cm⁻¹): 2940, 2793, 1697, 1603, 1455.

¹H NMR (DMSO-d₆, ppm): 10 (s, 2H, CHO); 7.6-7.2 (m, 8H, Ph); 4.1 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, J=6Hz, 4H, O-CH₂-CH₂-CH₂); 1.6 (m, 2H, J=6Hz, O-CH₂-CH₂-CH₂).

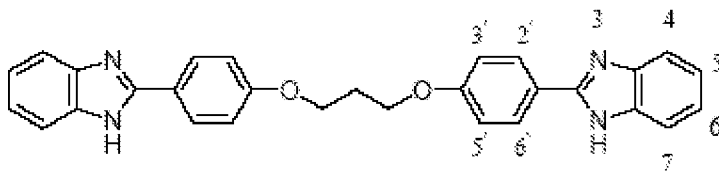
10 Mp (°C): 55.1- 56.9°C. Yield: 37% MW: 312.36.

Compounds

The compounds were obtained following the general procedure described herein using the corresponding intermediates described herein. The present invention encompasses the use of compounds 1 to 25 and their derivatives, N-oxides, salts, solvates, hydrates, tautomeric forms, analogues, pro-drugs, esters and metabolites, as well as their quaternized nitrogen analogues.

15

Compound 1: 2,2' – [1,3-propanediyloxy]bis(1,4-phenylene)]bis-1H-benzimidazole

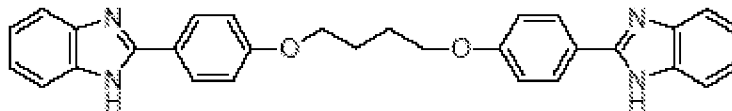


IR (KBr, cm⁻¹): 3631-2667(N-H bz), 1612, 1499, 1451, 1252.

20 ¹H NMR (DMSO-d₆, ppm): 8.1 (d, 4H, J=9Hz, C₆H₄: H₂',H₆'); 7.6 (AA'BB', 4H, benz: H₄,H₇); 7.2 (AA'BB', 4H, benz: H₅,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄: H₃',H₅'); 4.3 (t, 4H, J=6Hz, O-CH₂-CH₂); 2.2 (m, 2H, J=6Hz, O-CH₂-CH₂).

HRMS (ESI-ToF) MH⁺ C₂₉H₂₅N₄O₂ : MW experimental: m/z 461.1973. MW calculated: m/z 461.1978.

25 Mp (°C): > 300°C. Yield: 98%.

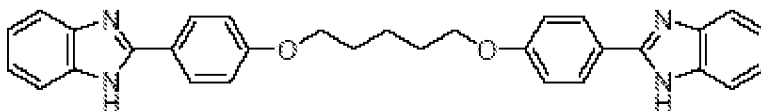
Compound 2: 2,2' – [1,4-butanediylbis(oxy-1,4-phenylene)]bis-1H-benzimidazole

IR (KBr, cm⁻¹): 3631-2364(N-H bz), 1612, 1500, 1439, 1253.

1H NMR (DMSO-d₆, ppm): 8.1 (d, 4H, J=9Hz, C₆H₄: H₂',H₆'); 7.6 (AA'BB', 4H, benz: H₄,H₇); 7.2 (AA'BB', 4H, benz: H₅,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄: H₃',H₅ '); 4.2 (t, 4H, J=6Hz, O-CH₂-CH₂); 2.0 (m, 4H, O-CH₂-CH₂).

HRMS (ESI-ToF) MH⁺ C₃₀H₂₇N₄O₂: MW experimental: m/z 475.2155. MW calculated: m/z 475.2134.

Mp (°C): > 300°C. Yield: 93%.

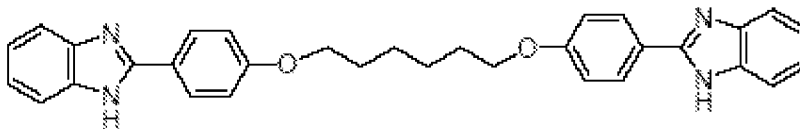
10 Compound 3: 2,2' – [1,5-pentanediybis(oxy-1,4-phenylene)]bis-1H-benzimidazole

IR (KBr, cm⁻¹): 3623-2400(N-H bz), 1762, 1611, 1500, 1254.

1H NMR (DMSO-d₆, ppm): 8.1 (d, 4H, J=9Hz, C₆H₄: H₂',H₆'); 7.6 (AA'BB', 4H, benz: H₄,H₇); 7.2 (AA'BB', 4H, benz: H₅,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄: H₃',H₅'); 4.1 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, O-CH₂-CH₂-CH₂); 1.6 (m, 2H, O-CH₂-CH₂-CH₂).

HRMS (ESI-ToF) MH⁺ C₃₁H₂₉N₄O₂: MW experimental: m/z 489.2275. MW calculated: m/z 489.2291.

Mp (°C): > 300°C. Yield: 94%.

Compound 4: 2,2' – [1,6-hexanediybis(oxy-1,4-phenylene)]bis-1H-benzimidazole

20

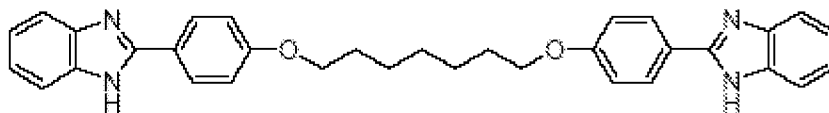
IR (KBr, cm⁻¹): 3570-2756(N-H bz), 1613, 1500, 1451, 1254.

1H NMR (DMSO-d₆, ppm): 8.1 (d, 4H, J=9Hz, C₆H₄: H₂',H₆'); 7.6 (AA'BB', 4H, benz: H₄,H₇); 7.2 (AA'BB', 4H, benz: H₅,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄: H₃',H₅'); 4.1 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, O-CH₂-CH₂-CH₂); 1.6 (m, 4H, O-CH₂-CH₂-CH₂).

HRMS (ESI-ToF) MH⁺ C₃₂H₃₁N₄O₂: MW experimental: m/z 503.2462. MW calculated: m/z 503.2447.

Mp (°C): > 300°C. Yield: 89%.

Compound 5: 2,2' – [1,7-heptanediylbis(oxy-1,4-phenylene)]bis-1H-benzimidazole



5

IR (KBr, cm⁻¹): 3632-2757(N-H bz), 1613, 1500, 1451, 1255.

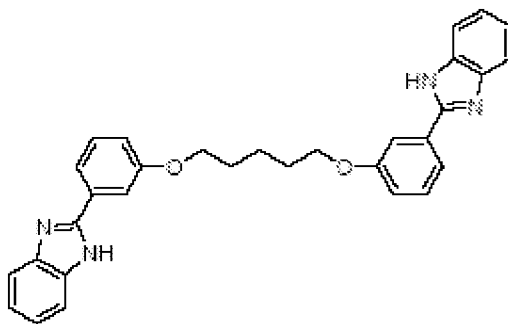
¹H NMR (DMSO-d₆, ppm): 8.1 (d, 4H, J=9Hz, C₆H₄: H_{2'},H_{6'}); 7.6 (AA'BB', 4H, benz: H₄,H₇), 7.2 (AA'BB', 4H, benz: H₅,H₆); 7.1 (d, 4H, J=9Hz, C₆H₄: H_{3'},H_{5'}); 4.0 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, O-CH₂-CH₂-CH₂); 1.4 (m, 2H+4H, O-CH₂-CH₂-CH₂).

10

HRMS (ESI-ToF) MH⁺ C₃₃H₃₃N₄O₂: MW experimental: m/z 517.2627; MW calculated: m/z 517.2604.

Mp (°C): 298- 301°C. Yield: 96%.

Compound 6: 2,2' – [1,5-pentanediybis(oxy-1,3-phenylene)]bis-1H-benzimidazole



15

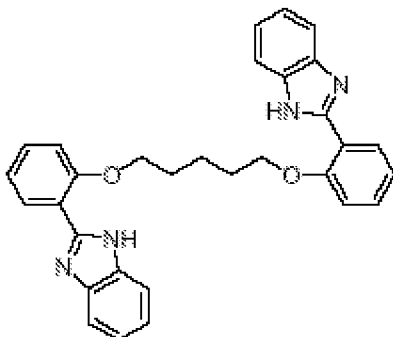
IR (KBr, cm⁻¹): 3705-2642(N-H bz), 1601, 1492, 1450, 1236.

¹H NMR (DMSO-d₆, ppm): 7.8 (d,s., J=7Hz, 2H+2H, C₆H₄: H_{4'},H_{2'}); 7.6 (AA'BB', 4H, benz: H₄,H₇); 7.4 (t, 2H, J=7Hz, C₆H₄: H_{5'}); 7.1 (d, 2H, J=7Hz, C₆H₄: H_{6'}); 7.2 (AA'BB',4H, benz: H₅,H₆); 4.1 (t, 4H, J= 6Hz, O-CH₂-CH₂-CH₂); 1.9 (m, 4H, J= 6Hz, O-CH₂-CH₂-CH₂); 1.5 (m, 2H, J= 6Hz, O-CH₂-CH₂-CH₂).

20

HRMS (ESI-ToF) MH⁺ C₃₁H₂₉N₄O₂ : MW experimental: m/z 489.2271. MW calculated: m/z 489.2291.

Mp (°C): 240- 243°C. Yield: 75%.

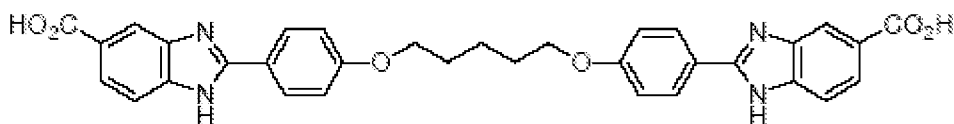
Compound 7: 2,2' – [1,5-pentanediyldis(oxy-1,2-phenylene)]bis-1H-benzimidazole

IR (KBr, cm⁻¹): 3652-2285(N-H bz), 1603, 1490, 1455, 1240.

1H NMR (DMSO-d₆, ppm): 8.2 (d, J=7Hz, 2H, C₆H₄: H_{6'}); 7.6 (AA'BB', 4H, benz: H₄,H₇);
 5 7.4 (t, 2H, J=7Hz, C₆H₄: H_{4'}); 7.2 (AA'BB', d, 2H+4H, C₆H₄: H_{3'} benz: H₅,H₆); 7.1 (t, 2H, J=7Hz, C₆H₄: H_{5'}); 4.2 (t, 4H, J=6Hz, O-CH₂-CH₂-CH₂); 1.9 (m, 4H, J= 6Hz, O-CH₂-CH₂-CH₂); 1.5 (m, 2H, J= 6Hz, O-CH₂-CH₂-CH₂).

HRMS (ESI-ToF) MH⁺ C₃₁H₂₉N₄O₂: MW experimental: m/z 489.2296. MW calculated: m/z 489.2291.

10 Mp (°C): 190- 193°C. Yield: 74%.

Compound 8: 2,2'-[1,5-pentanediyldis(oxy-1,4-phenylene)]-1H-benzimidazole-5-carboxylic acid

IR (KBr, cm⁻¹): 3691-2362(N-H bz), 1680, 1609, 1505, 1256.

15 1H NMR (DMSO-d₆, ppm): 13.0 (s, 1H, COOH); 8.2 (d,s.,4H+2H,J=8Hz, C₆H₄: H_{6'},H_{2'}, benz: H₄); 7.8 (d, 2H, J= 8Hz, benz: H₆); 7.6 (d, 2H, J= 8Hz, benz: H₇); 7.2 (d, 4H, J= 8Hz, C₆H₄: H_{5'}, H_{3'}); 4.1 (t, 4H, J= 6Hz, O-CH₂-CH₂-CH₂); 1.8 (m, 4H, J= 6Hz, O-CH₂-CH₂-CH₂); 1.6 (m, 2H, J=6Hz, O-CH₂-CH₂-CH₂).

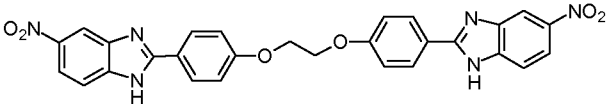
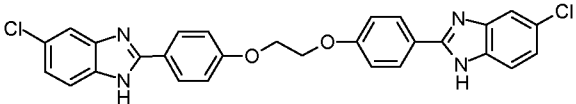
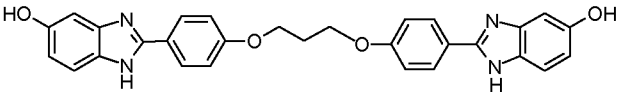
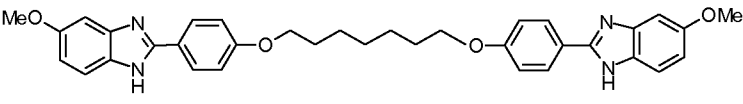
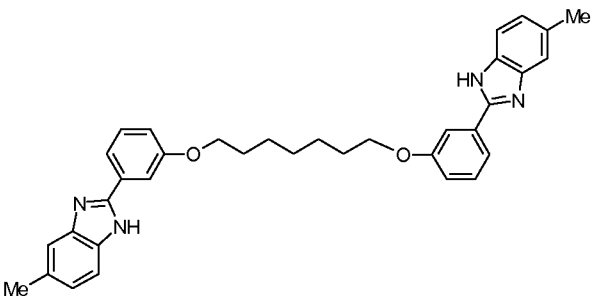
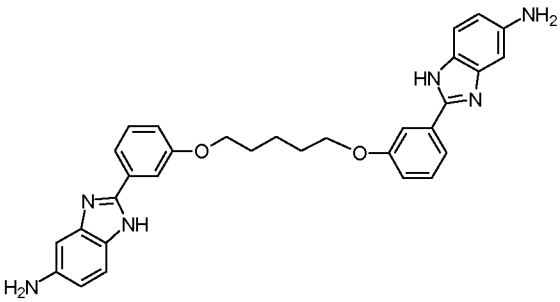
HRMS (ESI-ToF) MH⁺ C₃₃H₂₈N₄O₆: MW experimental: m/z 577.2091. MW calculated: m/z 577.2087.

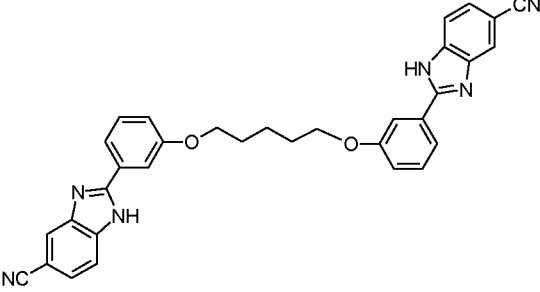
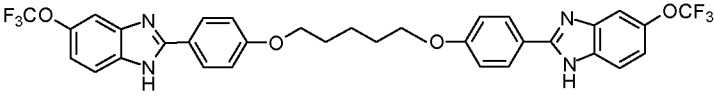
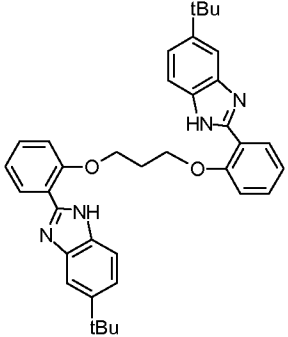
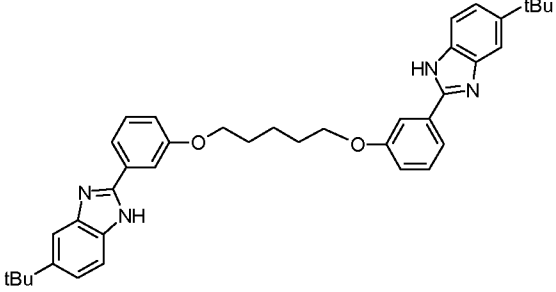
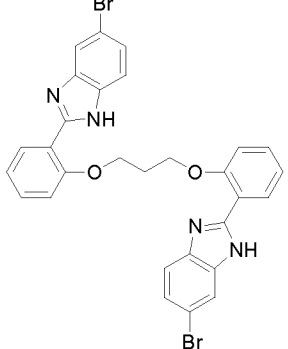
20

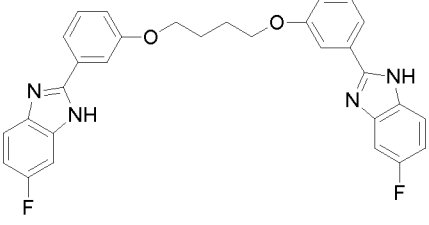
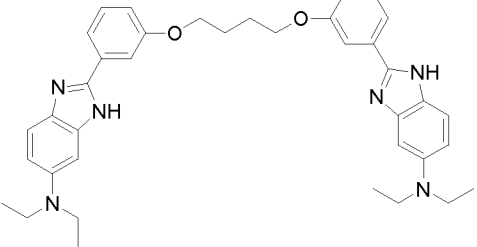
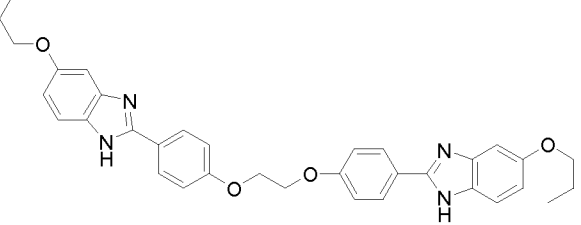
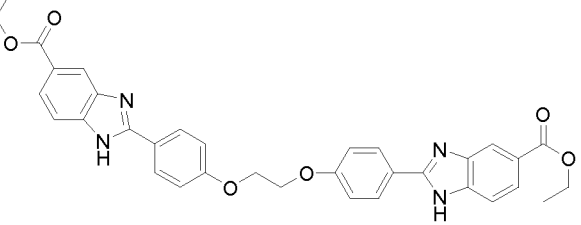
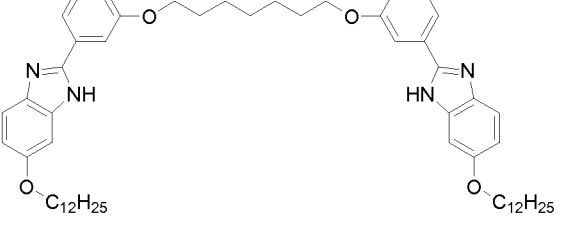
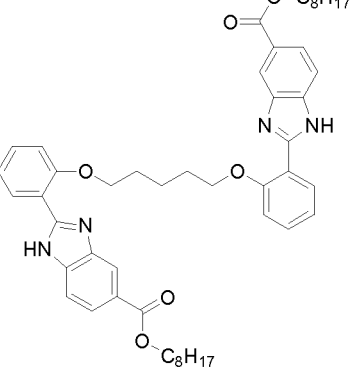
Mp (°C): 292- 296°C. Yield: 97%.

Compounds 9-25 set out in tabulated form in Table 1 are synthesized using similar protocols.

Table 1

Compound	Name	Structure
9	2,2'-[1,2-ethanediylbis(oxy-1,4-phenylene)]bis-5-nitro-1H-benzimidazole	
10	2,2'-[1,2-ethanediylbis(oxy-1,4-phenylene)]bis-5-chloro-1H-benzimidazole	
11	2,2'-[1,3-propanediylbis(oxy-1,4-phenylene)]bis-5-hydroxy-1H-benzimidazole	
12	2,2'-[1,7-heptanediylbis(oxy-1,4-phenylene)]bis-5-methoxy-1H-benzimidazole	
13	2,2'-[1,7-heptanediylbis(oxy-1,3-phenylene)]bis-5-methyl-1H-benzimidazole	
14	2,2'-[1,5-pentanediybis(oxy-1,3-phenylene)]bis-5-amino-1H-benzimidazole	

Compound	Name	Structure
15	2,2'-[1,5-pentanediy]bis(oxy-1,3-phenylene)]bis-5-cyano-1H-benzimidazole	
16	2,2' - [1,5-pentanediy]bis(oxy-1,4-phenylene)]bis-5-trifluoromethoxy-1H-benzimidazole	
17	2,2' - [1,3-propanediy]bis(oxy-1,2-phenylene)]bis-5-tert-butyl-1H-benzimidazole	
18	2,2' - [1,3-pentanediy]bis(oxy-1,3-phenylene)]bis-5-tert-butyl-1H-benzimidazole	
19	2,2'-[1,3-propanediy]bis(oxy-1,2-phenylene)]bis-5-bromo-1H-benzimidazole	

Compound	Name	Structure
20	2,2'-[1,4-butanediylbis(oxy-1,3-phenylene)]bis-5-bromo-1H-benzimidazole	
21	2,2'-[1,4-butanediylbis(oxy-1,3-phenylene)]bis-5-diethylamino-1H-benzimidazole	
22	2,2'-[1,2-ethanediylbis(oxy-1,4-phenylene)]bis-5-propoxy-1H-benzimidazole	
23	Diethyl 2,2'-[1,2-ethanediylbis(oxy-1,4-phenylene)]bis-1H-benzimidazole-5-carboxylate	
24	2,2'-[1,7-heptanediylbis(oxy-1,3-phenylene)]bis-5-dodecoxy-1H-benzimidazole	
25	Diocetyl 2,2'-[1,5-pentanediybis(oxy-1,2-phenylene)]bis-1H-benzimidazole-5-carboxylate	

Malaria: In vitro drug screening model against *Plasmodium falciparum***Parasite and cell cultures**

Two strains of *P. falciparum* were used: 1/ GHA strain (*P.falGHA*), derived from a Ghanese patient and chloroquine sensitive and 2/ W-2 strain (*P.falW2*) from CDC/Indochina III and resistant to chloroquine, quinine, pyrimethamine, but susceptible to mefloquine. The strains were maintained in RPMI-1640 medium supplemented with 0.37 mM hypoxanthine, 25 mM HEPES, 25 mM NaHCO₃, and 10% O+ human serum together with 2-4% washed human O+ erythrocytes (Adaptation of the procedures described by Trager, W.; Jensen J. B. Science 1976, 193, 673-677). All cultures and assays were conducted at 37°C under an atmosphere of 4% CO₂, 3% O₂ and 93% N₂.

Compound solutions/dilutions

Compound stock solutions were prepared in 100% DMSO at 20 mM or mg/ml (Concentrations are standard expressed in molar concentrations). The compounds were serially pre-diluted (2-fold or 4- fold) in DMSO followed by a further (intermediate) dilution in demineralized water to assure a final in-test DMSO concentration of <1%.

Drug sensitivity assays

Assays were performed in sterile 96-well microtiter plates, each well containing 10 µl of the watery compound dilutions together with 190 µl of the malaria parasite inoculum (1% parasitaemia, 2% HCT). Parasite growth was compared to untreated-infected (100% growth) and uninfected-controls (0% growth). After 72h incubation at 37°C, plates were frozen at -20°C. After thawing, 20 µl of each well was transferred into another plate together with 100 µl Malstat reagent (Lactate dehydrogenase (LDH)-test, according to Makler et al.: Am.J.Trop Med. Hyg, 48, 7339-741, 1993) and 20 µl of a 1/1 mixture of PES (phenazine ethosulfat, 0.1 mg/ml) and NBT (NitroBlue Tetrazolium Grade III, 2 mg/ml). The plates were kept out of light for 2 hours and change in color was measured spectrophotometrically at 655 nm. The results were expressed as % reduction in parasitaemia compared to control wells. For each compound, an IC₅₀ was calculated.

Primary screen

The GHA strain was used. Compounds were tested at 5 concentrations (64 - 16 - 4 - 1 and 0.25 µM or µg/ml). Artesunate (IC₅₀ = 0.005 + 0.004 µM) and chloroquine (IC₅₀ = 0.05 + 0.08 µM) were included as reference drugs. Some of the compounds were evaluated in a secondary screening.

Secondary screen

Additional strains (GHA, W2, D6, NF54) were included and the IC₅₀-values were determined using an extended dose range (2-fold compound dilutions). Artesunate and chloroquine were included as reference drugs.

- 5 The results are shown in Table 2. Compounds for use in the invention were not cytotoxic.

Table 2

	IC ₅₀ (µg / ml) (score)
Chloroquine	0.04
Compound 1	16.58 (1)
Compound 3	0.49 (3)
Compound 6	3.91 (2)
Compound 7	8.31 (2)

The compounds tested were active against *P. falciparum*. Therefore the compounds represent an interesting class of anti-malarial agents.

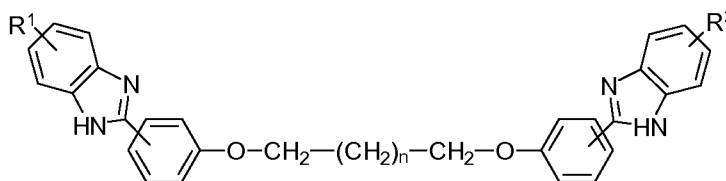
- 10 Compared to chloroquine some of the compounds, such as compound 3, were particularly selective since not active against some other strains of parasites.

All patents, patent applications, and published references cited herein are hereby incorporated by reference in their entirety. While this invention has been particularly shown and described with references to preferred embodiments, it will be understood by

- 15 those skilled in the art that various changes in form and details may be made without departing from the scope of the invention encompassed by the claims.

Claim

1. Use of at least one compound of formula I or a pharmaceutically acceptable salt or solvate thereof for the preparation of a medicament for the prevention and/or treatment of malaria,



I

wherein n is an integer selected from 0 to 10, and

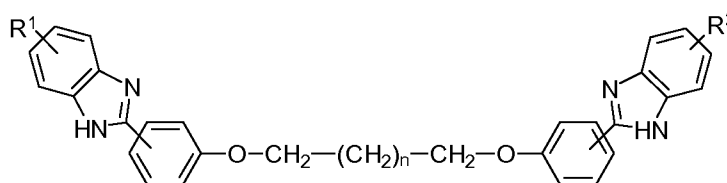
R¹ and R² are each independently selected from hydrogen, halogen, hydroxyl, alkyl, alkoxy, amino, alkylamino, alkylcarbonylamino, alkylcarbonyloxy, formyl, alkylcarbonyl, alkyloxycarbonyl, halocarbonyl, haloalkyl, haloalkoxy, carbamoyl, cyano, nitro, sulfo, or a carboxyl group.

10

2. Use according to claim 1, wherein n is 0, 1, 2, 3, 4, 5, 6 or 7.
3. Use according to claim 1 or 2, wherein R¹ and R² are each independently hydrogen, hydroxyl, halogen, alkyl, alkylamino, alkoxy, haloalkyl, haloalkoxy, alkyloxycarbonyl, alkylcarbonyl, amino, cyano, nitro or a carboxyl group.
- 15
4. Use according to any of claims 1 to 3, wherein the at least one compound of formula I is used in combination with at least one additional drug selected from the group consisting of chloroquine, artemesin, qinghaosu, 8-aminoquinoline, amodiaquine, arteether, artemether, artemisinin, artesunate, artesunic acid, artelinic acid, atovoquone, azithromycine, biguanide, chloroquine phosphate, chlorproguanil, cycloguanil, dapson, desbutyl halofantrine, desipramine, doxycycline, dihydrofolate reductase inhibitors, dipyridamole, halofantrine, haloperidol, hydroxychloroquine sulfate, imipramine, mefloquine, penfluridol, phospholipid inhibitors, primaquine, proguanil, pyrimethamine, pyronaridine, quinine, quinidine, quinacrineartemisinin, sulfonamides, sulfones, sulfadoxine, sulfalene, tafenoquine, tetracycline, tetrandine, triazine, salts or mixture thereof.
- 20
5. Use according to any of claims 1 to 4, for the treatment of drug-resistant malaria.
- 25

6. Use according to any of claims 1 to 5, wherein said malaria is selected from *Plasmodium falciparum* malaria, *P. vivax* malaria, *P. ovale* malaria, or *P. malariae* malaria.

7. A method for the therapeutic and/or prophylactic treatment of malaria in a subject in need of such treatment comprising administering to the subject a therapeutically effective amount of a compound of formula I or a pharmaceutically acceptable salt or solvate thereof



I

10 wherein

n is an integer selected from 0 to 10, and

R¹ and R² are each independently selected from hydrogen, halogen, hydroxyl, alkyl, alkoxy, amino, alkylamino, alkylcarbonylamino, alkylcarbonyloxy, formyl, alkylcarbonyl, alkyloxycarbonyl, halocarbonyl, haloalkyl, haloalkoxy, carbamoyl, cyano, nitro, sulfo, or a carboxyl group.

8. The method according to claim 7, wherein n is 0, 1, 2, 3, 4, 5, 6 or 7.

9. The method according to claim 7 or 8, wherein R¹ and R² are each independently hydrogen, hydroxyl, halogen, alkyl, alkoxy, haloalkyl, haloalkoxy, alkylcarbonyl, amino, alkylamino, alkyloxycarbonyl, cyano, nitro, or a carboxyl group.

10. The method according to any of claims 7 to 9, wherein said compound of formula I is administered in combination with at least one additional drug selected from the group consisting of chloroquine, artemesin, qinghaosu, 8-aminoquinoline, amodiaquine, arteether, artemether, artemisinin, artesunate, artesunic acid, artelinic acid, atovoquone, azithromycine, biguanide, chloroquine phosphate, chlorproguanil, cycloguanil, dapson, desbutyl halofantrine, desipramine, doxycycline, dihydrofolate reductase inhibitors, dipyrindamole, halofantrine, haloperidol, hydroxychloroquine sulfate, imipramine, mefloquine, penfluridol, phospholipid inhibitors, primaquine, proguanil, pyrimethamine, pyronaridine, quinine, quinidine, quinacrineartemisinin, sulfonamides, sulfones, sulfadoxine, sulfalene, tafenoquine, tetracycline, tetrandine, triazine, salts or mixture thereof.

11. The method according to any of claims 7 to 10, wherein the subject has been infected with *Plasmodium falciparum*.
12. The method according to any of claims 7 to 10, wherein the subject has been infected with *P. vivax*.
- 5 13. The method according to any of claims 7 to 10, wherein the subject has been infected with *P. ovale*.
14. The method according to any of claims 7 to 10, wherein the subject has been infected with *P. malariae*.
- 10 15. The method according to any of claims 7 to 14, wherein the compound of Formula I is administered after the subject has been exposed to the malaria parasite.
16. The method according to any of claims 7 to 15, wherein the malaria parasite is a drug-resistant malarial strain.
17. The method according to any of claims 7 to 16, wherein the compound of Formula I is administered before the subject travels to a country where malaria is endemic.

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2008/063224

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K31/4184 A61P33/06 A61K45/06

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>BELL C A ET AL: "STRUCTURE-ACTIVITY RELATIONSHIPS OF ANALOGS OF PENTAMIDINE AGAINST PLASMODIUM FALCIPARUM AND LEISHMANIA MEXICANA AMAZONENSIS" ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, AMERICAN SOCIETY FOR MICROBIOLOGY, WASHINGTON, DC, US, vol. 34, no. 7, 1 July 1990 (1990-07-01), pages 1381-1386, XP008013577 ISSN: 0066-4804 abstract page 1381, column 1, paragraph 1 page 1382; tables 1,2 page 1384; table 6; compounds 31-33 page 1383; table 3 page 1385, column 1, paragraph 2</p> <p align="center">----- -/--</p>	1-17

Further documents are listed in the continuation of Box C.

See patent family annex.

* Special categories of cited documents :

- *A* document defining the general state of the art which is not considered to be of particular relevance
- *E* earlier document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

- *T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- *X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- *Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- *&* document member of the same patent family

Date of the actual completion of the international search

11 February 2009

Date of mailing of the international search report

23/02/2009

Name and mailing address of the ISA/

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040,
Fax: (+31-70) 340-3016

Authorized officer

Haider, Ursula

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2008/063224

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	DE 198 53 944 A1 (BOURDICHON ALAIN JACQUES [DE]) 25 May 2000 (2000-05-25) claim 1 -----	1-17
A	DONKOR I O ET AL: "Trypanocidal activity of dicationic compounds related to pentamidine" EUROPEAN JOURNAL OF MEDICINAL CHEMISTRY, EDITIONS SCIENTIFIQUE ELSEVIER, PARIS, FR, vol. 36, no. 6, 1 June 2001 (2001-06-01), pages 531-538, XP004372864 ISSN: 0223-5234 page 533, column 1 - column 2 -----	1-17
A	WO 95/08540 A (WELLCOME FOUND [GB]; CLEARY DARRYL GENE [US]; CORY MICHAEL [US]; SHERM) 30 March 1995 (1995-03-30) page 57, paragraph 4 - page 58; claim 1 -----	1-17

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/EP2008/063224

Patent document cited in search report	Publication date	Publication date	Patent family member(s)	Publication date
DE 19853944	A1	25-05-2000	AT 275949 T	15-10-2004
			AU 1653100 A	13-06-2000
			WO 0030631 A1	02-06-2000
			EP 1220668 A1	10-07-2002
WO 9508540	A	30-03-1995	AU 7661594 A	10-04-1995
			EP 0720603 A1	10-07-1996
			HU 71345 A2	28-11-1995
			JP 9506335 T	24-06-1997