



(51) International Patent Classification:
C07D 471/04 (2006.01) *A61P 35/00* (2006.01)
A61K 31/505 (2006.01)

(21) International Application Number:
PCT/EP2014/068404

(22) International Filing Date:
29 August 2014 (29.08.2014)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
13306192.9 2 September 2013 (02.09.2013) EP

(71) Applicants: INSERM (INSTITUT NATIONAL DE LA SANTÉ ET DE LA RECHERCHE MÉDICALE) [FR/FR]; 101, rue de Tolbiac, Paris, 75013 (FR). UNIVERSITE DE BOURGOGNE [FR/FR]; Esplanade Erasme, F-21000 Dijon (FR). CENTRE NATIONAL DE LA RECHERCHE SCIENTIFIQUE - CNRS [FR/FR]; 3, rue Michel Ange, Paris Cedex 16, 75794 (FR). INSTITUT CURIE [FR/FR]; 26 rue d'Ulm, F-75005 Paris (FR).

(72) Inventors: NGUYEN, Chi-Hung; UMR 176 CNRS Institut Curie, Bât 110 Centre Universitaire d'Orsay, F-91405 Orsay (FR). QUERE, Ronan; U866 Faculté de Médecine, 7 boulevard Jeanne d'Arc, F-21000 Dijon (FR).

(74) Agent: CABINET PLASSERAUD; 52 rue de la Victoire, F-75440 Paris Cedex 09 (FR).

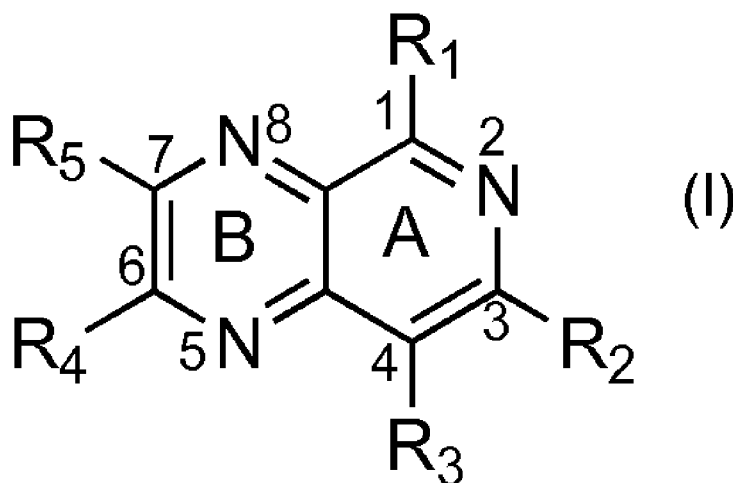
(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, 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, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, 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, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Published:

— with international search report (Art. 21(3))

(54) Title: PYRIDO[4,3-b]PYRAZINE COMPOUNDS AND COMPOSITIONS



(57) Abstract: A pyrido[4,3-b]pyrazine compound of formula (I) the tautomers, the stereoisomers, the mixtures and the pharmaceutical acceptable salts thereof, for use in a method of therapeutic treatment of the human or animal body, pharmaceutical compositions comprising as active ingredient a pyrido[4,3-b]pyrazine compound of formula I, use as chemotherapeutic agent in cancers and new pyrido[4,3-b]pyrazine compounds.

Pyrido[4,3-b]pyrazine compounds and compositions

It is an object of the invention to provide pyrido[4,3-b]pyrazine compounds and compositions, useful for treatment of cancer, particularly of leukaemia. It is another object of the invention to provide novel compounds of formula I and their non-toxic, pharmaceutically acceptable acid addition salts and a process for their preparation.

These and other objects and advantages of the invention will become obvious from the following detailed description.

Possible anti cancer effects of pluricyclic compounds, more particularly of pyrido[4,3-b]pyrazine compounds have been investigated in the past. Anti-Cancer Drug Design (1995), **10**, 277-297, discloses the synthesis and biological activities of a series of pyrido[4,3-b]pyrazine compounds. However said compounds were considered as inactive and the authors conclude that whereas tricyclic compounds comprising a pyrrole nucleus were active, the replacement of the pyrrole nucleus by a pyrazine ring abolishes anti tumour properties. The use of such compounds as drugs has therefore not been evidenced nor disclosed.

WO201113002 generically discloses a broad family of condensed pyridine derivatives useful as potent inhibitors of protein kinase CK2. Two compounds have been prepared and tested for protein kinase CK2 activity.

Hongbo Liu et Al. in International Journal Of Molecular Sciences, vol. 12. No. 10, page 7008, and Fabarice et Al: in J. Med. Chem. vol. 54. 2011 page 642 disclose a same pyrazino quinoline compound wherein one of the carbon atoms of the pyridine ring is substituted by a [3-]chloro phenyl amino substituent directly linked to said carbon atom.

The usual treatment of acute myeloid leukemia (AML) is chemotherapy. It usually results in complete remission. But despite consolidation chemotherapy, a significant number of patients will relapse. It was therefore hypothesized that this treatment does not remove all the cancer cells. Thus, because of its lack of action, chemotherapy eliminates differentiated cells or cells during differentiation, but the population of leukemic cells can persist and cause therapeutic exhausts.

There remains a need in the art to provide molecules specifically targeting this population of leukemic cells.

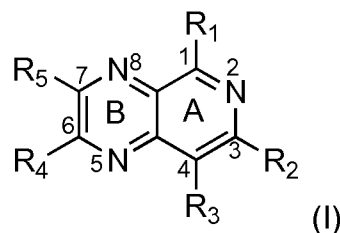
The present invention fulfills this need, and further provides other related advantages. This object has been also solved by the provision of novel compounds.

Surprisingly, it has now been shown that pyrido[4,3-b]pyrazine compounds are indeed endowed with anti-cancer properties. More precisely, the applicant has discovered that derivatives of pyrido[4,3-b]pyrazine which are devoid of protein kinase CK2 activity, allow inhibiting the multiplication of leukaemic cells.

It has been noted in a surprising and unexpected fashion that the derivatives of pyrido[4,3-b]pyrazine of the invention stop the process of replication of malignant, particularly leukaemia cells. In addition, most of them are not toxic for haematopoietic cells. Therefore, they have a high therapeutic index: toxicity for leukaemia cells/ toxicity for haematopoietic cells. Accordingly they allow eliminating leukemia cells but do not affect the normal hematopoietic stem cells.

These properties are illustrated below in the experimental section. They justify the use of the pyrido[4,3-b]pyrazine compounds of the invention, as drugs.

A subject of the present application is therefore a pyrido[4,3-b]pyrazine compound of formula I



wherein

R₁ represents

- hydrogen,

- a (C1-C6)alkoxyl,

- a (C1-C6)alkylthio,

- a —NRaRb group, wherein

Ra represents

- hydrogen or

- an (C1-C6) alkyl radical and

Rb represents

- hydrogen or
- a (C1-C6) alkyl, or
- a (C2-C6)NRcRd group, wherein

Rc represents

- 5
- hydrogen or
 - a (C1-C6) alkyl radical and

Rd represents

- 10
- hydrogen or
 - a (C1-C6) alkyl radical, or
 - a (C2-C6)NReRf group, wherein

Re and Rf each independently represents

- hydrogen or
- a (C1-C6) alkyl radical, or

NReRf represents

- 15
- N(CH₂CH₂)₂O, or
 - N(CH₂CH₂)₂S, or
 - N(CH₂CH₂)₂N(C1-C6)alkyl, or
 - N(CH₂CH₂)₂CH(C1-C6)alkyl, or
 - N(CH₂)_n wherein n = 3, 4, 5 or 6, or

20 NRcRd represents

- N(CH₂CH₂)₂O, or
- N(CH₂CH₂)₂S, or
- N(CH₂CH₂)₂N(C1-C6)alkyl, or
- N(CH₂CH₂)₂CH(C1-C6)alkyl, or
- 25 - N(CH₂)_n wherein n = 3, 4, 5 or 6, or
- N(CH₂CH₂)₂NReRf, wherein Re and Rf each independently have the above definitions

R₂ and R₃ being the same or different, represent

- 30
- hydrogen,
 - a (C1-C6) alkyl radical,
 - an aryl radical or
 - a substituted or unsubstituted aryl cycle fused with cycle A via carbon atoms 3 and 4, or

- R₂ and R₃ taken together form a bivalent radical of formula (CH₂)_n wherein n = 3, 4, 5 or 6

R₄ and R₅ being the same or different, represent

- hydrogen,

5 - a (C1-C6) alkyl radical, or

- an aryl group fused with cycle B via carbon atoms 6 and 7, said aryl group optionally being substituted by one or more (C1-C6)alkyl, (C1-C6)alkoxy, OH, halogen atom or NR'R'' group, wherein R' and R'' each independently represents H, a (C1-C6) alkyl, a (C1-C6)alkoxy, a (C1-C6) alkyl or R₄ and R₅

10 may be taken together to form a bivalent radical of formula (CH₂)_n in which n = 3, 4, 5 or 6

the tautomers, the stereoisomers, the mixtures and the pharmaceutical acceptable salts thereof, for use in a method of therapeutic treatment of the human or animal body, that is to say as a drug.

15 Hereinafter, general reference to the pyrido[4,3-b]pyrazine compounds of formula I of the present invention also includes the tautomers, the stereoisomers, the mixtures and the pharmaceutical acceptable salts thereof.

Examples of C1-C6 alkyl radicals are methyl, ethyl, n-propyl, isopropyl, n-butyl and isobutyl. Preferred (C1-C6) alkyl radicals are (C1-C4) alkyl
20 radicals, particularly methyl or ethyl radicals. Preferred (C1-C6) alkoxy radicals are (C1-C4) alkoxy radicals, particularly methoxy, or ethoxy radicals.

Examples of suitable acids for the formation of the non-toxic, pharmaceutically acceptable acid addition salts are inorganic acids such as hydrochloric acid, hydrobromic acid, nitric acid, sulphuric acid, phosphoric acid,
25 acetic acid, formic acid, propionic acid, benzoic acid, maleic acid, fumaric acid, succinic acid, tartaric acid, citric acid, oxalic acid, glyoxylic acid, aspartic acid, alkane sulfonic acids such as methane or ethane sulfonic acid, arylsulfonic acids such as benzene or p-toluene sulfonic acid and arylcarboxylic acids.

In the present application "aryl" group means a conventional aryl
30 group, i.e. a group of atoms derived from benzene or from a benzene derivative by removing one hydrogen that is bonded to the benzene ring, such as a phenyl, tolyl or xylyl group or means an heteroaryl group wherein the heteroatom is for example a O atom or preferably a N atom. The aryl group preferably comprises a C4-C10 ring, particularly a C4-C6 ring. Preferred rings are a phenyl, pyridine or

naphthyl, ring and particularly preferred rings are a phenyl or pyridine ring. The aryl group may be substituted by one or more, preferably 1 or 2, (C1-C6) alkyl, (C1-C6) alkoxy, hydroxy or halogeno substituents.

Preferred "aryl" groups of the present application exclude a naphthyl group.

Among the preferred compounds of formula I are those wherein R_1 represents a mono- or di-(C1-C6) alkylamino(C1-C6) alkylamino (C1-C6) alkylamino or a mono- or di-(C1-C6) alkylamino(C1-C6) alkylamino radical, preferably the latter.

Among the preferred compounds of formula I are also those wherein R_2 represents a hydrogen atom or a (C1-C6) alkyl radical and particularly a hydrogen atom or a methyl radical.

Among the preferred compounds of formula I are also those wherein R_3 represents a hydrogen atom or a (C1-C6), preferably (C1-C2), alkyl radical.

Among the preferred compounds of formula I are also those wherein R_4 and R_5 are taken together to form a aryl cycle, particularly wherein a aryl cycle does not represent a naphthyl group, said aryl cycle being preferably a phenyl cycle fused with cycle B via carbon atoms 6 and 7, said aryl cycle being unsubstituted or substituted by one or more hydroxy, (C1-C6)alkyl, (C1-C6)alkoxyl radicals.

Among the preferred compounds, the derivatives wherein R_1 represents a di(C1-C6)alkyl amino (C1-C6)alkyl amino, R_2 represents a hydrogen atom, R_3 represents a (C1-C6) alkyl radical and R_4 and R_5 represent an aryl cycle fused with cycle B via carbon atoms 6 and 7, said aryl cycle optionally being substituted by one or more (C1-C6)alkyl, (C1-C6)alkoxyl, OH, and their acid addition salts are more preferred.

Specific preferred compounds are the compounds cited in the examples and more particularly:

- 1-[(2-Dimethylamino)ethyl]amino-6-hydroxy-4-methylpyrido[4,3-b] quinoxaline
- 1-[(2-Dimethylamino)ethyl]amino-4-methylpyrido[4,3-b] quinoxaline
- 1-[(3-Dimethylamino) propyl]amino-8-methoxy-4-methylpyrido [4,3-b] quinoxaline

- 1-[(2-Dimethylamino)ethyl]amino-8-methoxy-4-methylpyrido[4,3-b] quinoxaline
 - 1-[(2-Dimethylamino)ethyl]amino-9-hydroxy-4-methylpyrido[4,3-b] quinoxaline
- and their non-toxic, pharmaceutically acceptable acid addition salts.

As previously mentioned, a compound of formula I allows
5 eliminating leukemia cells.

These properties also justify the use of the compounds of the invention described above in a pharmaceutical composition.

Therefore, a further subject of the present invention is a pharmaceutical composition comprising as active ingredient a pyrido[4,3-
10 b]pyrazine compound of the invention and an inert pharmaceutical carrier or excipient.

The novel pharmaceutical, particularly anti-cancer, more particularly anti-leukaemic, compositions of the invention are more particularly comprised of an effective amount of at least one compound of formula I and its non-toxic,
15 pharmaceutically acceptable acid addition salts and an inert pharmaceutical carrier or excipient. The compositions may be in the form of tablets, dragees, capsules, granules, suppositories or injectable solutions or suspensions.

Examples of suitable excipients are talc, gum arabic, lactose, starch, magnesium stearate, cocoa butter, aqueous and non-aqueous vehicles, fatty
20 substances of animal or vegetable origin, paraffin derivatives, glycols, various wetting, dispersing or emulsifying agents and preservatives.

The compounds of the present invention may particularly be formulated for parenteral administration (e.g., by injection, for example bolus injection or continuous infusion) and may be presented in unit dose form in
25 ampoules, pre-filled syringes, small volume infusion or in multi-dose containers with or without an added preservative. The compositions may take such forms as suspensions, solutions, or emulsions in oily but preferably aqueous vehicles. Examples of oily or non-aqueous carriers, diluents solvents or vehicles include propylene glycol, polyethylene glycol, vegetable oils (e.g., olive oil, and injectable
30 organic esters (e.g., ethyl oleate), and may contain formulatory agents such as preserving, wetting, emulsifying or suspending, stabilizing and/or dispersing agents. The active ingredient may also be in powder form, obtained by aseptic isolation of sterile solid or by lyophilization from solution for constitution before use

with a suitable vehicle, e.g., sterile, pyrogen-free water. Drugs for parenteral administration are sterile.

The novel compositions of the invention are useful for example in the curative treatment of all hematological malignancies and hematological disorders. They can also be used in the treatment of leukemias and lymphomas, including Acute lymphoblastic leukemia, Acute myelogenous leukemia, Chronic lymphocytic leukemia, Chronic myelogenous leukemia, Chronic myelogenous leukemia, Hodgkin's lymphomas, Non-Hodgkin's lymphomas. They also find their use in treating all solid cancers, tumors and metastasis (for example brain, lung, breast and gastrointestinal tumors).

The usual dose, which varies depending on the subject and the condition of the patient, may be, for example, from 1 to 100 mg per day, injected intravenously (into a vein), in humans of the compound of Example 1, for 21 to 28 days, the composition administered being a solution (liquid) or a powder to be mixed with liquid to be injected intravenously) by a doctor or nurse, for example in a medical facility. The length of treatment depends on the drug, how well the body responds to it, and the type of cancer.

Among the preferred compositions of the invention are those cited above.

The present invention also relates to a process for preparing a composition described above, characterized in that, according to methods known per se, the active ingredient or ingredients are mixed with acceptable excipients, particularly pharmaceutically acceptable excipients and packaged for use if desired, for example in vials.

The compounds of formula I of the invention possess very useful pharmacological properties; they stop the process of replication of malignant, particularly leukaemia cells. In addition, most of them are not toxic for haematopoietic cells. Therefore, they have a high therapeutic index toxicity for leukaemia cells/ toxicity for haematopoietic cells, in contrast with active ingredients such as doxorubicin which can cause a severe decrease in the number of blood cells in bone marrow.

The invention accordingly also relates to the use of a compound of formula I, for manufacturing a medicament for use as a chemotherapeutic anti-cancer agent. The invention further relates to a compound of formula I, for use as

chemotherapeutic agent in cancers, particularly for the treatment of haematological malignancies, more particularly for the treatment of leukaemia conditions.

The novel method of the invention for treating cancers, particularly haematological malignancies, more particularly leukaemia, in warm-blooded animals comprises administering to warm-blooded animals an anti cancer, particularly anti leukaemia effective amount of at least one compound of formula I. The compound(s) can be administered orally, rectally or preferably parenterally and the usual daily dose is 1 to 200 mg/kg depending on the specific compound, the condition treated, the weight of the patient and the method of administration. For example, the intravenous administration of 1 to 100 mg/kg of the product of Example 2 is useful for the treatment of leukemias or lymphomas.

The compounds of formula (I) are described in the literature, particularly in Anti-Cancer Drug Design (1995), 10, 277-297 or may be manufactured as follows:

4-chloro-5-methyl-3-nitropyridin-2 (1H)-one may be synthesized in four steps from malonyldichloride and propionitrile as described previously (Davis et al., 1962; Nguyen et al., 1992b) and converted by alcoholic ammonia into the corresponding 4-aminopyridone. The 4-aminopyridone may be submitted to catalytic (Pd) reduction under hydrogen for obtaining the corresponding 3,4-diaminopyridinone which may be immediately condensed with 1,2-cyclohexanedione. The resulting 6,7,8,9-tetrahydropyridoquinoxaline may then be subjected to aromatization by refluxing in diphenyl oxide and in the presence of palladium on charcoal to afford the pyridoquinoxaline. Chlorination may be carried out with a mixture of phosphorus oxychloride and benzyl-triethylammonium chloride in acetonitrile according to the method of Robins & Uznanski, 1981. 1-chloro-4-methyl-pyrido[3,4-b]quinoxaline may then be easily substituted by various dialkylaminoalkylamines to provide the corresponding 1-amino-substituted derivatives.

Pyridoquinoxalines bearing methoxy and hydroxy groups on their various benzo-ring positions may for example be prepared as follows. The condensation of p-anisidine with chloropyridone provides a nitrodiarylamine which may be submitted to a reduction-cyclization reaction according to the modified process described by Rewcastle et al. (1987) for the synthesis of phenazine

derivatives. By refluxing a mixture of nitrodiarylamines with NaBH₄, preferably in a 5M sodium methoxide in methanol, methoxypyrido-quinoxalin-1-ones are obtained.

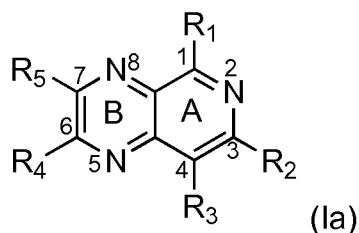
In the same conditions as described above for
 5 pyridoquinoxalinone, methoxypyrido-quinoxalin-1-ones may be transformed into 1-chloropyridoquinoxalines. Demethylation of alkoxy substituents may be achieved using boron tribromide, giving the corresponding 6,7,8 or 9-hydroxypyridoquinoxalines.

Advantageously, in the case where the reduction-cyclization
 10 provides mixtures of the cyclised products, next chlorinations may be performed from pyridoquinoxalinone mixtures. The desired products may be separated by column chromatography.

The compounds of formula (I) have a basic character and the acid addition salts thereof can with advantage be prepared by reacting a mineral or
 15 organic acid in more or less stoichiometric proportions with the compounds of formula (I). The salts can be prepared without isolating the corresponding bases.

The present invention is also directed to novel compounds of formula I.

20 The novel compounds of the invention are selected from the group consisting of pyrido pyrazines of formula IA



wherein

R₁ represents

- 25 - hydrogen,
 - a (C1-C6)alkoxyl,
 - a (C1-C6)alkylthio,
 - a —NRaRb group, wherein

Ra represents

- 30 - hydrogen or

- an (C1-C6) alkyl radical and

Rb represents

- hydrogen or

- a (C1-C6) alkyl, or

5 - a (C2-C6)NRcRd group, wherein

Rc represents

- hydrogen or

- a (C1-C6) alkyl radical and

Rd represents

10 - hydrogen or

- a (C1-C6) alkyl radical, or

- a (C2-C6)NReRf group, wherein

Re and Rf each independently represents

- hydrogen or

15 - a (C1-C6) alkyl radical, or

NReRf represents

- N(CH₂CH₂)₂O, or

- N(CH₂CH₂)₂S, or

- N(CH₂CH₂)₂N(C1-C6)alkyl, or

20 - N(CH₂CH₂)₂CH(C1-C6)alkyl, or

- N(CH₂)_n wherein n = 3, 4, 5 or 6, or

NRcRd represents

- N(CH₂CH₂)₂O, or

- N(CH₂CH₂)₂S, or

25 - N(CH₂CH₂)₂N(C1-C6)alkyl, or

- N(CH₂CH₂)₂CH(C1-C6)alkyl, or

- N(CH₂)_n wherein n = 3, 4, 5 or 6, or

- N(CH₂CH₂)₂NReRf, wherein Re and Rf each independently have the above definitions

30 R₂ and R₃ being the same or different, represent

- hydrogen,

- a (C1-C6) alkyl radical,

- an aryl radical or

- a substituted or unsubstituted aryl cycle fused with cycle A via carbon atoms 3 and 4, or
- R_2 and R_3 taken together form a bivalent radical of formula $(CH_2)_n$ wherein $n = 3, 4, 5$ or 6

5 R_4 and R_5 being the same or different, represent

- hydrogen, or
- a (C1-C6) alkyl radical,

the tautomers, the stereoisomers, the mixtures and the pharmaceutical acceptable salts thereof.

10 Preferred selections of compounds of formula I described above also apply to the other subjects of the invention envisaged above: drugs, compositions, pyrido pyrazines of formula IA etc.

The invention will now be described by means of the following examples.

15

Figure 1 represents the toxicity on hematopoietic stem cells and leukemic cells of chemical compounds, including the compound of example 1, tested at 5 microgrammes/mL during 18 hours of in vitro culture. Toxicity on leukemic cells is measured by disappearance of GFP+ cells by FACS. High toxic
20 chemical compounds show low % of remaining GFP+ cells, while for non-toxic compounds the % of GFP+ cells is high. Toxicity on hematopoietic stem cells is measured according the % of Propidium Iodide positive (IP+) cells detected by FACS. The % of dead cells is correlated to the % of IP+cells.

Figure 2 shows the % of GFP+ leukemic cells (black bars) and
25 healthy hematopoietic stem cells (white bars) reconstitutions in peripheral blood of recipient mice treated with the compound of example 1. Recipient mice were lethally irradiated (900 cGy) and transplanted with 50 000 leukemic cells and 50 000 healthy hematopoietic stem cells, injected in the tail vein. Mice were treated 10 and 13 days post transplantation by intraperitoneal injection of 3 mg/Kg of the
30 chemical compound. Reconstitution was measured by FACS, 3 weeks after the transplantation and compared to control mice injected with DMSO. $n = 5$ mice analyzed per group.

Figure 3 shows the toxicity on leukemic cells (upper graph) and hematopoietic stem cells (bottom graph) of 17 different analogues of the compound 1 (compounds 1 to 17).

Figure 4 shows GFP+ leukemic cells (black bars) and healthy hematopoietic stem cells (white bars) reconstitutions in peripheral blood of recipient mice treated with compounds 1, 2, 3, 4 or 5. Recipient mice were lethally irradiated (900 cGy) and transplanted with 50 000 leukemic cells and 50 000 healthy hematopoietic stem cells, injected in the tail vein. Mice were treated 10 and 13 days post transplantation by intraperitoneal injection of 3 mg/Kg of the chemical compounds 1, 2, 3, 4 or 5. Reconstitution was measured by FACS, 3 weeks after the transplantation and compared to control mice injected with DMSO. n = 9 mice analyzed for each group.

Figure 5 shows Kaplan–Meier plot of survival mice over time when mice were treated with the compounds of examples 1, 2, 3, 4 and 5. Mice were treated with DMSO (n=19 mice), compound 1 (n = 17 mice), 2 (n = 10 mice), 3 (n = 11 mice), 4 (n = 10 mice), 5 (n = 9 mice) and Doxorubicine (n = 20 mice). Mice were treated 10 and 13 days post transplantation by intraperitoneal injection of 3 mg/Kg of the chemical compounds 1, 2, 3, 4, 5 or Doxorubicine.

EXAMPLE 1: 1-[(2-Dimethylamino)ethyl]amino-6-hydroxy-4-methylpyrido[4,3-b] quinoxaline

Anti-Cancer Drug Design (1995), **10**, 277-297, compound 21a.

EXAMPLE 2: 1-[(2-Dimethylamino)ethyl]amino-4-methylpyrido[4,3-b] quinoxaline

Anti-Cancer Drug Design (1995), **10**, 277-297, compound 14a.

EXAMPLE 3: 1-[(3-Dimethylamino) propyl]amino-8-methoxy-4-methylpyrido [4,3-b] quinoxaline

Anti-Cancer Drug Design (1995), **10**, 277-297, compound 18b.

EXAMPLE 4: 1-[(2-Dimethylamino)ethyl]amino-8-methoxy-4-methyl pyrido[4,3-b] quinoxaline

Anti-Cancer Drug Design (1995), **10**, 277-297, compound 18a.

EXAMPLE 5: 1-[(2-Dimethylamino)ethyl]amino-9-hydroxy-4-methylpyrido[4,3-b] quinoxaline

Anti-Cancer Drug Design (1995), 10, 277-297, compound 21b.

5 EXAMPLE 6: 5-[(3-Dimethylamino)propyl]amino-2,3,8-trimethylpyrido[4,3-b]pyrazine

The title compound was synthesized in 3 steps from 3,4-diamino-5-methylpyridin-2(1H)-one.

Step 1 : The mixture of 3,4-diamino-5-methylpyridin-2(1H)-one
10 (prepared as described in *Anti-Cancer Drug design* (1995), **10** , 277-2971; 4 g, 10 mmol), diacetyl (1.3 g, 15 mmol) and absolute ethanol (65 mL) was heated under reflux for 1.5 h. Evaporation into dryness under reduced pressure of the resulting mixture afforded the crude intermediate 1, 2,3,8-trimethylpyrido[4,3-*b*]pyrazin-5(6H)-one which was submitted for the next step without any further
15 purification.

Step 2 : Under nitrogen atmosphere, the mixture of 2,3,8-trimethylpyrido[4,3-*b*]pyrazin-5(6H)-one obtained above (intermediate 1, 10 mmol) benzyltriethylammonium chloride (9.0g, 40 mmol), phosphoryl trichloride (55 mL, large excess) and acetonitrile (100 mL) was heated under reflux for 4 h.
20 The volatile material was evaporated in vacuum, cold water (200 ml) was added and the medium was basified by addition of 28% ammonia. The organic layer obtained after extraction using dichloromethane as solvent was dried over MgSO₄ and evaporated in vacuum into dryness to give a residue which was purified by flash chromatography (silica, gradient of ethyl acetate (0 to 10%) in
25 dichloromethane to give the intermediate 2, 5-chloro-2,3,8-trimethylpyrido[4,3-*b*]pyrazine, (930 mg, 44% yield). ¹H NMR (CDCl₃) δ 8.33 (s, 1H), 2.82 (s, 3H), 2.80 (s, 3H), 2.65 (s, 3H). Microanalyses, calculated for C₁₀H₁₀ClN₃: C, 57.84; H, 4.85; N, 20.24 ; found: C, 57.74; H, 4.87; N, 20.11.

30 Step 3 : Under nitrogen atmosphere, the mixture of 5-chloro-2,3,8-trimethylpyrido[4,3-*b*]pyrazine obtained above (intermediate 2, 200 mg, 1 mmol), K₂CO₃ (370 mg, 26 mmol), *N*¹,*N*¹-dimethylpropane-1,3-diamine (1.3 g, 13 mmol) and diglyme (3 mL) was heated under reflux for 2.5 h. The volatile material was evaporated in vacuum, water (50 ml) was added and the medium
35 was basified by addition of 28% ammonia. The organic layer obtained after

extraction using dichloromethane as solvent was dried over MgSO_4 and evaporated in vacuum into dryness to give a residue which was purified by flash chromatography (neutral alumina, gradient of ethanol (0 to 1.5%) in dichloromethane to give of free base of the titled 5-[(3-
5 dimethylamino)propyl]amino-2,3,8-trimethylpyrido[4,3-*b*]pyrazine (220 mg, 83% yield). ^1H NMR (CDCl_3) δ 7.94 (s, 1H), 7.06 (m, 1H), 3.66-3.60 (m, 1H), 2.70 (s, 3H), 2.66 (s, 3H), 2.44 (m, 2H), 2.27 (s, 6H), 1.94-1.84 (m, 2H). Maleate salt formation: A solution of the free base (220 mg) in boiling acetone (15 mL) was poured into a solution of maleic acid (200 mg) in hot acetone (5 ml). The
10 precipitate obtained was collected by filtration at room temperature, washed with minimum amount of acetone and dried in a desiccator affording the maleate salt (340 mg, 83%). Microanalyses, calculated for $\text{C}_{15}\text{H}_{23}\text{N}_5 \cdot 2 \text{C}_4\text{H}_4\text{O}_4$: C, 54.65; H, 6.58; N, 13.85; found: C, 54.71; H, 6.25; N, 13.87.

15 EXAMPLE 7: 5-[(3-Dimethylamino)propyl]amino-2,3-
dimethylpyrazino[2,3-*c*]quinoline

The title compound was synthesized in 2 steps from 2,3-dimethylpyrazino[2,3-*c*]quinolin-5(6H)-one prepared as disclosed by Gewald K. et al. (Chemische Berichte (1991), 124(5), 1237-1241).

20 Step 1: Under nitrogen atmosphere, a mixture of 2,3-dimethylpyrazino[2,3-*c*]quinolin-5(6H)-one (1.42 g, 6.3 mmol), benzyltriethylammonium chloride (5.8 g, 25 mmol), phosphoryl trichloride (40 mL, 440 mmol) and acetonitrile (70 mL) was heated under reflux for 4 h. The
25 volatile material was evaporated in vacuum, cold water (150 ml) was added and the medium was basified by addition of 28% ammonia. The organic layer obtained after extraction using dichloromethane as solvent was dried over MgSO_4 and evaporated in vacuum into dryness to give a residue which was purified by flash chromatography (silica, gradient of ethyl acetate (0 to 8%) in
30 dichloromethane to give 5-chloro-2,3-dimethylpyrazino[2,3-*c*]quinoline (1.47 g, 95 % yield). ^1H NMR (CDCl_3) δ 9.01 (d, 1H), 8.13 (d, 1H), 7.85 (t, 1H), 7.76 (t, 1H), 2.88 (s, 3H), 2.87 (s, 3H). Microanalyses, calculated for $\text{C}_{13}\text{H}_{10}\text{ClN}_3$: C, 64.07; H, 4.14; N, 17.24; found: C, 63.77; H, 4.04; N, 16.86.

Step 2 : Under nitrogen atmosphere, the mixture of 5-chloro-2,3-dimethylpyrazino[2,3-c]quinoline obtained above (intermediate 4, 250 mg, 1 mmol) and *N*¹,*N*¹-dimethylpropane-1,3-diamine (3 mL, large excess) was heated under reflux for 4 h. After evaporation of the excess of amine in vacuum, water (30 ml) was added and the medium was basified by addition of 28% ammonia. The organic layer obtained after extraction using dichloromethane as solvent was dried over MgSO₄ and evaporated in vacuum into dryness to give a residue which was purified by flash chromatography (neutral alumina, gradient of ethanol (0 to 1.5%) in dichloromethane to give of free base of the expected 5-[(3-dimethylamino)propyl]amino-2,3-dimethylpyrazino[2,3-c]quinoline (320 mg, quantitative yield). ¹H NMR (CDCl₃) δ 8.62 (d, 1H), 7.64 (d, 1H), 7.53-7.48 (m, 1H), 7.38 (br t, 1H), 7.26-7.18 (m, 1H), 3.71 (q, 2H), 2.68 (s, 3H), 2.61 (s, 3H), 2.41 (t, 2H), 2.23 (s, 6H), 1.86 (qi, 2H).

Maleate salt formation: A solution of the free base (300 mg) in boiling acetone (20 mL) was poured into a solution of maleic acid (360 mg) in hot acetone (5 ml). The precipitate obtained was collected by filtration at room temperature, washed with minimum amount of acetone and dried in a desiccator affording the maleate salt (470 mg, 74%). Microanalyses, calculated for C₁₈H₂₃N₅ · 2 C₄H₄O₄: C, 57.66; H, 5.77; N, 12.93; found: C, 57.85; H, 5.58; N, 13.11.

EXAMPLE 8: Tablets

Tablets were prepared containing respectively 25 and 100 mg of 1-[(2-dimethylamino)ethyl]amino-6-hydroxy-4-methylpyrido[4,3-b] quinoxaline bi-maleate and sufficient excipient of lactose, starch, talc and magnesium stearate to obtain a final weight of 100 mg.

EXAMPLE 9: Vials for injections

Vials containing an injectable sterile solution consisting of 200 mg of 1-[(3-dimethylamino) propyl]amino-8-methoxy-4-methylpyrido [4,3-b] quinoxaline bi-maleate in an aqueous vehicle for injections were prepared.

PHARMACOLOGICAL STUDY

Toxicity for leukaemia cells

Isolation of bone marrow cells

Femurs and tibias were harvested from 7- to 12-week-old C57BL/6
5 × B6SJL mice. Bones were crushed, the bone marrow (BM) cell suspension
was filtered through a 100-µm-pore size Cell Strainer filter (Fisher) and the
lineage-negative (Lin⁻) population was depleted from BM cells, with a cocktail
of antibodies: rat anti-mouse-B220, -Gr1, -Mac1, -CD4, -CD5, -CD8 and -
Ter119 (Miltenyi Biotec).

10 Anti-rat immunoglobulin G-conjugated immunomagnetic beads
(Miltenyi Biotec) were used for enrichment of the Lin⁻ population. For
prestimulation before the retroviral transduction, cells were cultured in SFEM
medium (StemCell) for 24 h, supplemented with 100 U/ml PenStrep (Sigma),
50 ng/ml murine stem cell factor, 10 ng/ml murine interleukin-3 and 50 ng/ml
15 murine interleukin-6 (Miltenyi Biotec).

Retroviral transduction and BM transplantation to generate Acute Myeloid Leukemia in mice

The MEIS1 and HOXA9 retroviral vectors expressed reciprocally
20 the MEIS1 cofactor and HOXA9 transcription factor, together with green
fluorescent protein (GFP). The vectors were transfected into Phoenix Amphi
cells (Nolan Lab), and supernatants were harvested for transduction of Lin⁻ BM
cells.

A total of 2×10^5 transduced Lin⁻ cells were mixed with 2×10^5
25 support BM cells and transplanted into the tail vein of lethally irradiated
(900 cGy) C57BL/6 × B6SJL recipient mice.

Mice were monitored for occurrence of Acute Myeloid Leukemia
(AML). By using fluorescence-activated cell sorting (FACS), the development of
Acute Myeloid Leukemia (AML) in mice was monitored by measuring the
30 proportion of GFP⁺ cells in peripheral blood. Recipient mice developed
leukemia, from around 8-12 weeks following the transplantation.

The phenotype of the leukemia was analyzed in peripheral blood
by staining with Mac1 antibody (Becton Dickinson). Stained cells were run
throughout a FACS LSRII (Becton Dickinson Biosciences) and the subsequent

data were analyzed with the FlowJo software (Tree Star). When development of AML was observed, femurs and tibias were harvested from mice, bones were crushed and the BM cell suspension was filtered through a 100- μ m-pore size Cell Strainer filter (Fisher).

5

Screening of chemical compounds on leukemic cells versus healthy hematopoietic cells

The tested compounds were collected on 96-well plates. A screening was performed *in vitro* on GFP⁺ BM cells harvested from mice suffering of AML. Chemical compounds were tested at 5 micrograms/mL together with cells (2×10^5 cells per well) cultured in SFEM medium, supplemented with 100 U/ml PenStrep, 50 ng/ml murine stem cell factor, 10 ng/ml murine interleukin-3 and 50 ng/ml murine interleukin-6.

After 18 hours of culture, FACS (GUAVA, Millipore) was performed to measure the effect of each chemical compound (percentage of remaining GFP⁺ cells).

High level of GFP is associated with no effect while low level of GFP means that the chemical molecule has induced death of AML cells.

Another screening was performed to quantify the toxicity of each chemical compounds on hematopoietic cells (Lin⁻ bone marrow cells).

After 18 hours of *in vitro* culture, toxicity was measured on leukemic cells by disappearance of GFP⁺ cells by FACS. Toxicity on hematopoietic stem cells was measured according the % of Propidium Iodide positive (IP⁺) cells detected by FACS.

Results: the results are shown on figure 3. Compounds of examples 1 to 5 have the capacity to induce death of leukemic cells and leave healthy hematopoietic stem cells intact. Low level of IP⁺ cells and effect on leukemic cells : disappearance of GFP⁺ cells.

Effect of chemical compounds on the development of AML

In order to test the effect of the compounds on the development of AML *in vivo*, 50 000 GFP⁺ cells were transplanted together with 50 000 Lin⁻

hematopoietic stem cells and 200 000 support cells (Lin⁺ cells depleted from Lin⁻ cells) into the tail vein of lethally irradiated (900 cGy) C57BL/6 × B6SJL recipient mice. The tested compound was reconstituted at 10mg/mL in DMSO and diluted 10 fold in physiological water before *in vivo* injection. The
5 Reconstituted compound was injected into the peritoneum of mice (3 mg/Kg), 10 and 13 days after the transplantation.

As a negative control, DMSO 10% was injected in animals.

As a positive control, Doxorubicin (Sigma Aldrich) was administered by intraperitoneal injection (3 mg/Kg). To control the development
10 of leukemia, GFP+ cells reconstitution was analyzed in peripheral blood by flow cytometry three weeks after the transplantation. Lifetime was monitored for survival analyses and peripheral blood from mice alive 100 days after the transplantation was tested by flow cytometry to quantify the percentage of remaining GFP+ cells.

15

RESULTS

Toxicity for leukaemic cells and haematopoietic cells

The results are reported on Figure 1.

20 Observations: The results show that the compound of example 1 tested in vitro on both hematopoietic stem cells and leukemic cells, at the concentration of 5 microgrammes/mL shows high toxicity on leukemic cells and reduced toxicity on hematopoietic stem cells.

Conclusion: The chemical compound 1 has the capacity to induce death of
25 leukemic cells, leaving healthy hematopoietic stem cells intact.

Effect on the development of AML in vivo

The results are reported on figure 2. the compound of example 1 shows reduced percentage of GFP+ leukemic cells in peripheral blood and
30 relevant reconstitution by healthy hematopoietic cells.

In vitro effect on leukemic cells versus healthy hematopoietic cells

The effect of 17 compounds including examples 1 to 5 was analyzed in vitro on GFP⁺ leukemic cells and their toxicity on healthy

hematopoietic stem cells. The results were reported on Figure 3. Compounds of examples 1 to 5 have the capacity to induce death of leukemic cells as assessed by an important reduction of leukemic cells expressing GFP after 18 hours of in vitro treatment (remaining GFP⁺ leukemic cells is below 10%). The compounds of examples 1 to 5 also leave healthy hematopoietic stem cells intact while there are less than 10% dead cells after 18 hours of treatment.

Effect of pyrido[4,3-b]pyrazine on the development of AML in vivo

Compared to control mice injected with DMSO, mice treated with compounds of examples 1 to 5 show an important reduction of the percentage of GFP⁺ leukemic cells in peripheral blood and a good reconstitution by healthy hematopoietic cells (Figure 4). Mice were also monitored for AML disease daily and euthanized when moribund.

By analyzing the percentage of survival mice over time (Figure 5), it was observed that mice treated with the compound of example 1 show a significant increase in their median overall survival (50 days versus 25 days for mice injected with DMSO), but the survival rate over 100 days was lower compared to the Doxorubicin control (23,5% versus 50%).

Mice treated with the compound of example 4 show significant increase in their median overall survival (50 days versus 25 days for mice injected with DMSO), and the survival rate over 100 days was closed to mice treated with Doxorubicin (55% versus 50%).

Mice treated with compounds of examples 2, 3 and 5 show the best survival rate over 100 days, even better than for mice treated with Doxorubicin (>75% versus 50%).

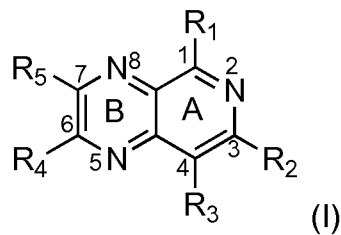
TOXICITY

Acute toxicity

The compounds of the examples of the present invention (3 mg/kg) were injected in mice. No adverse effects were observed.

REVENICATIONS

1. A pyrido[4,3-b]pyrazine compound of formula I



5 wherein

R_1 represents

- hydrogen,
- a (C1-C6)alkoxyl,
- a (C1-C6)alkylthio,

10 - a —NRaRb group, wherein

R_a represents

- hydrogen or
- an (C1-C6) alkyl radical and

R_b represents

15 - hydrogen or
 - a (C1-C6) alkyl, or
 - a (C2-C6) NRcRd group, wherein

R_c represents

20 - hydrogen or
 - a (C1-C6) alkyl radical and

R_d represents

- hydrogen or
- a (C1-C6) alkyl radical, or
- a (C2-C6) NReRf group, wherein

25 R_e and R_f each indedendently represents

- hydrogen or
- a (C1-C6) alkyl radical, or

NReRf represents

30 - $\text{N}(\text{CH}_2\text{CH}_2)_2\text{O}$, or
 - $\text{N}(\text{CH}_2\text{CH}_2)_2\text{S}$, or

- $N(CH_2CH_2)_2N(C1-C6)alkyl$, or
- $N(CH_2CH_2)_2CH(C1-C6)alkyl$, or
- $N(CH_2)_n$ wherein $n = 3, 4, 5$ or 6 , or

NRcRd represents

- 5 - $N(CH_2CH_2)_2O$, or
- $N(CH_2CH_2)_2S$, or
- $N(CH_2CH_2)_2N(C1-C6)alkyl$, or
- $N(CH_2CH_2)_2CH(C1-C6)alkyl$, or
- $N(CH_2)_n$ wherein $n = 3, 4, 5$ or 6 , or
- 10 - $N(CH_2CH_2)_2NReRf$, wherein Re and Rf each independently have the above definitions

R_2 and R_3 being the same or different, represent

- hydrogen,
- a (C1-C6) alkyl radical,
- 15 - an aryl radical or
- a substituted or unsubstituted aryl cycle fused with cycle A via carbon atoms 3 and 4, or
- R_2 and R_3 taken together form a bivalent radical of formula $(CH_2)_n$ wherein $n = 3, 4, 5$ or 6
- 20 R_4 and R_5 being the same or different, represent
- hydrogen,
- a (C1-C6) alkyl radical, or
- an aryl group fused with cycle B via carbon atoms 6 and 7, said aryl cycle optionally being substituted by one or more (C1-C6)alkyl, (C1-C6)alkoxy, OH,
- 25 halogen atom or $NR'R''$ group, wherein R' and R'' each independently represents H, a (C1-C6) alkyl, a (C1-C6)alkoxy, a (C1-C6) alkyl or R_4 and R_5 may be taken together to form a bivalent radical of formula $(CH_2)_n$ in which $n = 3, 4, 5$ or 6

the tautomers, the stereoisomers, the mixtures and the pharmaceutical
30 acceptable salts thereof, for use in a method of therapeutic treatment of the human or animal body.

2. A pyrido[4,3-b]pyrazine compound according to claim 1, wherein R_1 represents a mono- or di-(C1-C6) alkylamino(C1-C6) alkylamino or a mono- or di-(C1-C6) alkylamino(C1-C6) alkylamino (C1-C6) alkylamino radical.

3. A pyrido[4,3-b]pyrazine compound according to claim 1 or 2, wherein R_2 represents a hydrogen atom or a methyl radical.

4. A pyrido[4,3-b]pyrazine compound according to any of claims 1 to 3, wherein R_3 represents a hydrogen atom or a (C1-C2) alkyl radical.

5. A pyrido[4,3-b]pyrazine compound according to any of claims 1 to 4, wherein R_4 and R_5 taken together form a phenyl (or aryl, plus générale ?) cycle fused with cycle B via carbon atoms 6 and 7, said phenyl (or aryl ?) cycle optionally being substituted by one or more hydroxy, (C1-C6)alkyl, (C1-C6)alkoxyl radical.

10 6. Any one of the following compounds:

- 1-[(2-Dimethylamino)ethyl]amino-6-hydroxy-4-methylpyrido[4,3-b] quinoxaline

- 1-[(2-Dimethylamino)ethyl]amino-4-methylpyrido[4,3-b] quinoxaline

- 1-[(3-Dimethylamino) propyl]amino-8-methoxy-4-methylpyrido [4,3-b] quinoxaline

15 - 1-[(2-Dimethylamino)ethyl]amino-8-methoxy-4-methylpyrido[4,3-b] quinoxaline

- 1-[(2-Dimethylamino)ethyl]amino-9-hydroxy-4-methylpyrido[4,3-b] quinoxaline or pharmaceutically acceptable salt thereof for use in a method of therapeutic treatment of the human or animal body.

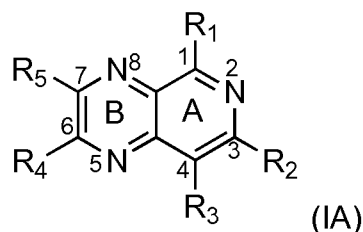
20 7. A pharmaceutical composition comprising as active ingredient a pyrido[4,3-b]pyrazine compound of formula I as defined in any of claims 1 to 6 and an inert pharmaceutical carrier or excipient.

8. A pyrido[4,3-b]pyrazine compound of formula I as defined in any of claims 1 to 6, for use as chemotherapeutic agent in cancers.

25 9. A pyrido[4,3-b]pyrazine compound of formula I according to claim 8 wherein the cancer is a haematological malignancy.

10. A pyrido[4,3-b]pyrazine compound of formula I according to claim 9 wherein the haematological malignancy is leukaemia.

11. A pyrido[4,3-b]pyrazine compound of formula IA



30 wherein

R₁ represents

- hydrogen,
- a (C1-C6)alkoxyl,
- a (C1-C6)alkylthio,

5 - a —NRaRb group, wherein

Ra represents

- hydrogen or
- an (C1-C6) alkyl radical and

Rb represents

- 10
- hydrogen or
 - a (C1-C6) alkyl, or
 - a (C2-C6)NRcRd group, wherein

Rc represents

- 15
- hydrogen or
 - a (C1-C6) alkyl radical and

Rd represents

- 20
- hydrogen or
 - a (C1-C6) alkyl radical, or
 - a (C2-C6)NReRf group, wherein
- Re and Rf each independently represents

- hydrogen or
- a (C1-C6) alkyl radical, or

NReRf represents

- 25
- N(CH₂CH₂)₂O, or
 - N(CH₂CH₂)₂S, or
 - N(CH₂CH₂)₂N(C1-C6)alkyl, or
 - N(CH₂CH₂)₂CH(C1-C6)alkyl, or
 - N(CH₂)_n wherein n = 3, 4, 5 or 6, or

NRcRd represents

- 30
- N(CH₂CH₂)₂O, or
 - N(CH₂CH₂)₂S, or
 - N(CH₂CH₂)₂N(C1-C6)alkyl, or
 - N(CH₂CH₂)₂CH(C1-C6)alkyl, or
 - N(CH₂)_n wherein n = 3, 4, 5 or 6, or

- $N(CH_2CH_2)_2NReRf$, wherein Re and Rf each independently have the above definitions

R_2 and R_3 being the same or different, represent

- hydrogen,

5 - a (C1-C6) alkyl radical,

- an aryl radical or

- a substituted or unsubstituted aryl cycle fused with cycle A via carbon atoms 3 and 4, or

- R_2 and R_3 taken together form a bivalent radical of formula $(CH_2)_n$ wherein $n =$

10 3, 4, 5 or 6

R_4 and R_5 being the same or different, represent

- hydrogen, or

- a (C1-C6) alkyl radical,

the tautomers, the stereoisomers, the mixtures and the pharmaceutical

15 acceptable salts thereof.

12. A pyrido[4,3-b]pyrazine compound of claim 11 wherein R_1 represents a mono- or di-(C1-C6) alkylamino(C1-C6) alkylamino or a mono- or di-(C1-C6) alkylamino(C1-C6) alkylamino (C1-C6) alkylamino radical, R_2 represents a hydrogen atom or a methyl radical, R_3 represents a hydrogen atom or a (C1-C2) alkyl radical.

20

13. A pharmaceutical composition comprising as active ingredient a pyrido[4,3-b]pyrazine compound of formula IA as defined in any of claims 11 to 13 and an inert pharmaceutical carrier or excipient.

14. A pyrido[4,3-b]pyrazine compound of formula IA as defined in any
25 of claims 11 to 13, for use as chemotherapeutic agent in cancers.

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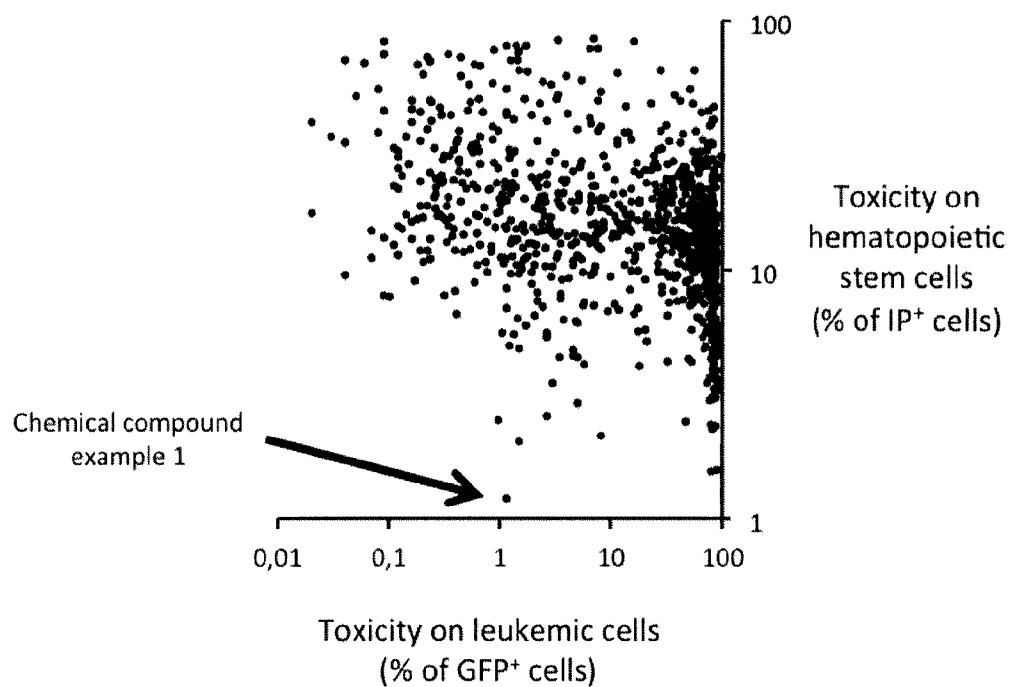


Fig. 1

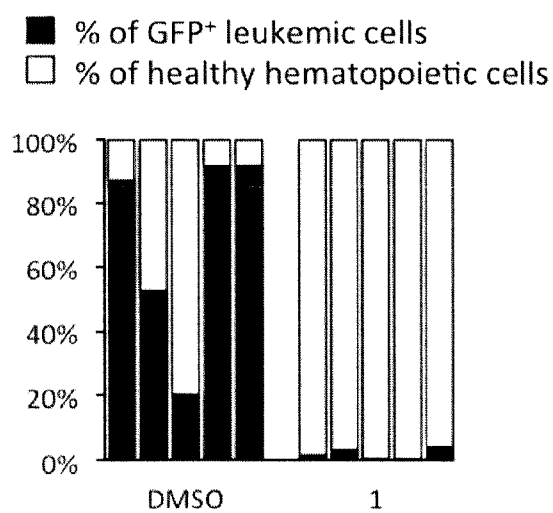


Fig. 2

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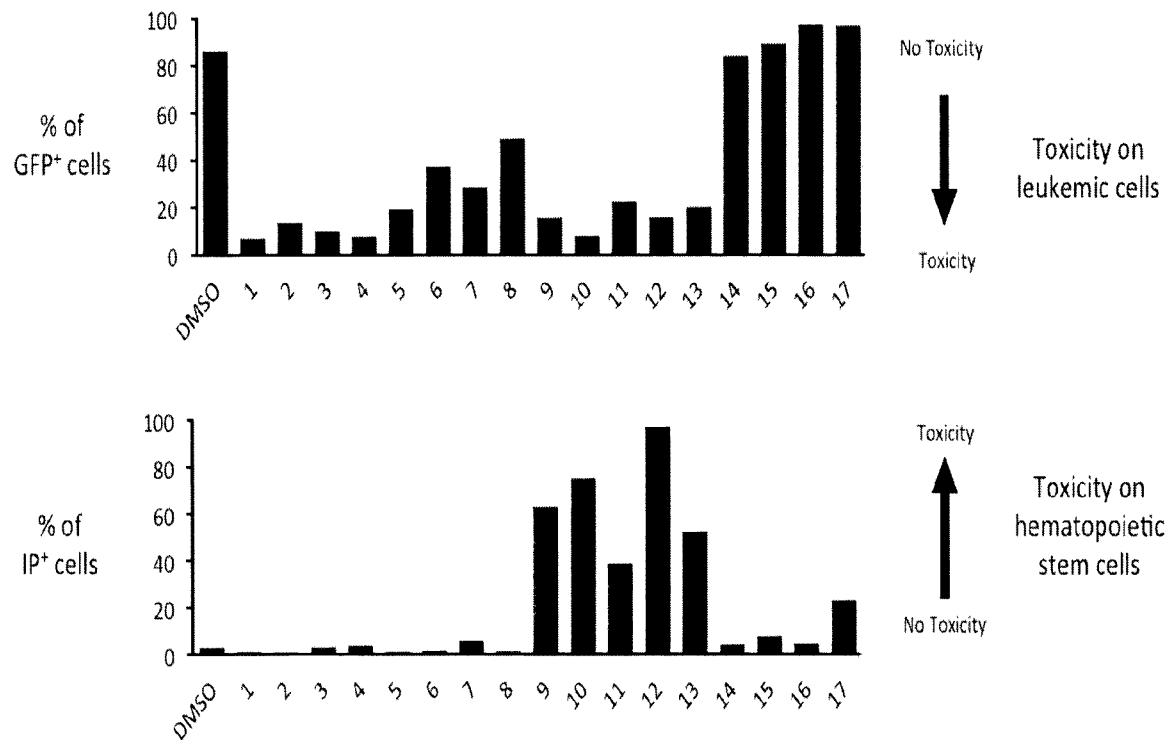


Fig. 3

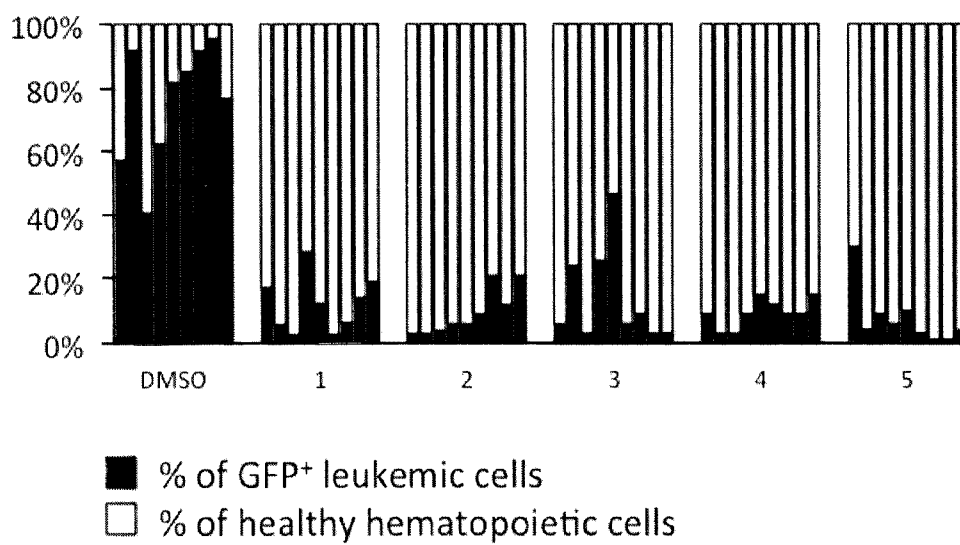


Fig. 4

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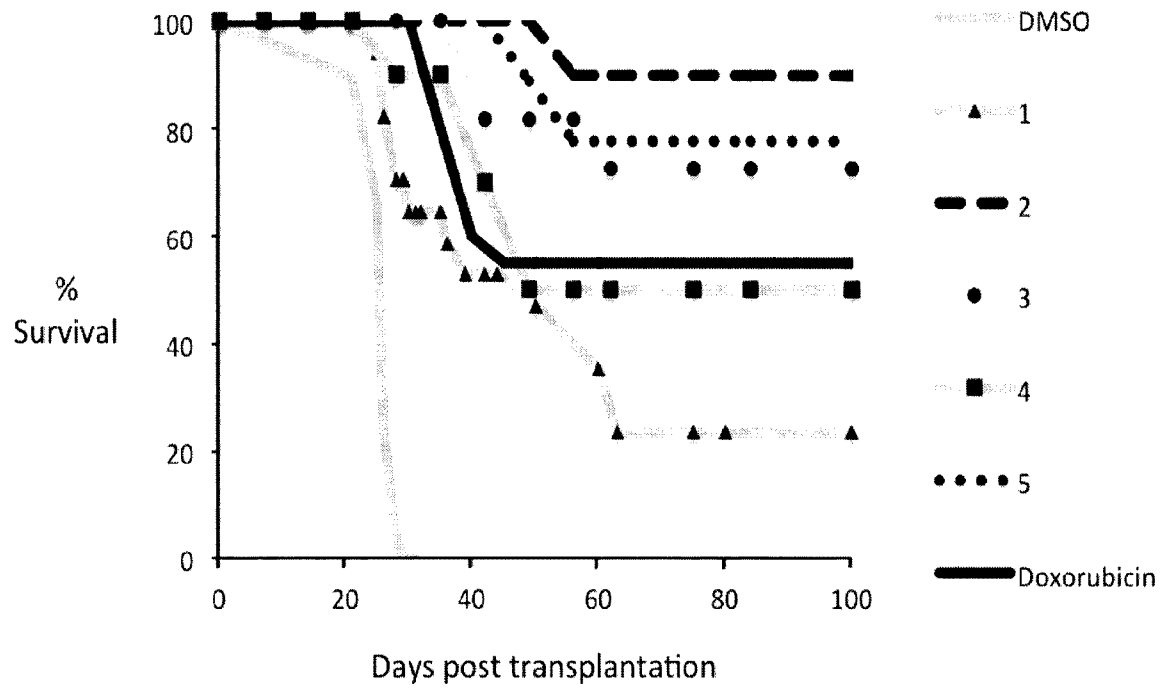


Fig. 5

INTERNATIONAL SEARCH REPORT

International application No

PCT/EP2014/068404

A. CLASSIFICATION OF SUBJECT MATTER

INV. C07D471/04 A61K31/505 A61P35/00
ADD.

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)

C07D A61K A61P

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 practicable, search terms used)

EPO-Internal, CHEM ABS Data, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	CHI HUNG NGUYEN ET AL: "Synthesis and biological evaluation of amino-substituted benzo(f)pyrido(4,3-b) and pyrido (3,4-b)quinoxalines: a new class of antineoplastic agents", ANTI-CANCER DRUG DESIGN, OXFORD UNIVERSITY PRESS, BASINGSTOKE, vol. 10, 1 January 1995 (1995-01-01), pages 277-297, XP009173893, ISSN: 0266-9536 page 278, paragraph in the middle; page 278, compounds 6-8; page 279, compound 14; page 280, compounds 12, 16, 18-21; page 281, compounds 27-32, 35-38; pages 2909-295, "results" ----- -/-	1-14



Further documents are listed in the continuation of Box C.



See patent family annex.

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"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent 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

20 October 2014

Date of mailing of the international search report

31/10/2014

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

Schmid, Arnold

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2014/068404

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	HONGBO LIU ET AL: "Structural Determinants of CX-4945 Derivatives as Protein Kinase CK2 Inhibitors: A Computational Study", INTERNATIONAL JOURNAL OF MOLECULAR SCIENCES, MOLECULAR DIVERSITY PRESERVATION INTERNATIONAL, BASEL, CH, vol. 12, no. 10, 20 October 2011 (2011-10-20), pages 7004-7021, XP009173897, ISSN: 1422-0067, DOI: 10.3390/IJMS12107004 page 7005-7006, "introduction"; table compound 49	1-14
X	----- P. FABARICE ET AL: "Discovery and SAR of 5-(3-Chlorophenylamino)benzo[c][2,6]naphth yridine-8-carboxylic Acid (CX-4945), the First Clinical Stage Inhibitor of Protein Kinase CK2 for the treatment of Cancer", J. MED. CHEM., vol. 54, 2011, pages 635-654, XP002715731, abstract; compound 28f -----	1-14