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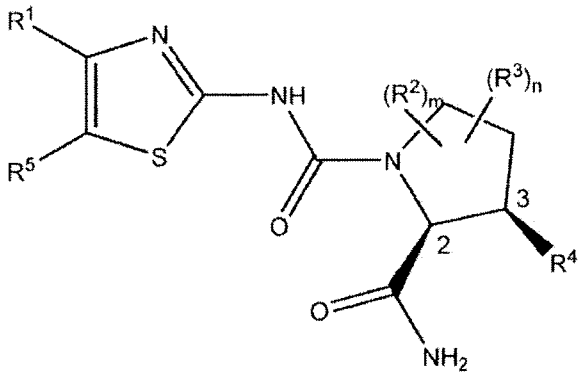
(54) Title: SUBSTITUTED 2-CARBOXAMIDE CYCLOAMINO UREAS



(I)

(57) Abstract: The present invention relates to compounds of formula (I), and salts thereof, wherein the substituents are as defined in the description, to compositions and use of the compounds in the treatment of diseases ameloriated by inhibition of phosphatidylinositol 3-kinase.

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Substituted 2-carboxamide cycloamino ureas

The present invention relates to substituted 2-carboxamide cycloamino ureas, as new phosphatidylinositol (PI) 3-kinase inhibitor compounds, their pharmaceutically acceptable salts, prodrugs thereof and processes for their production. This invention also relates to compositions of these compounds, either alone or in combination with at least one additional therapeutic agent, and optionally in combination with a pharmaceutically acceptable carrier. This invention still further relates to methods of use of these compounds, either alone or in combination with at least one additional therapeutic agent, in the prophylaxis or treatment of a number of diseases, in particular, those mediated by one or more of abnormal activity of growth factors, receptor tyrosine kinases, protein serine/heroine kinases, G protein coupled receptors and phospholipid kinases and phosphatases.

Phosphatidylinositol 3-kinases (PI3Ks) comprise a family of lipid kinases that catalyze the transfer of phosphate to the D-3' position of inositol lipids to produce phosphoinositol-3-phosphate (PIP), phosphoinositol-3,4-diphosphate (PIP_2) and phosphoinositol-3,4,5-triphosphate (PIP_3) that, in turn, act as second messengers in signaling cascades by docking proteins containing pleckstrin-homology, FYVE, Phox and other phospholipid-binding domains into a variety of signaling complexes often at the plasma membrane ((Vanhaesebroeck et al., *Annu. Rev. Biochem* 70:535 (2001); Katso et al., *Annu. Rev. Cell Dev. Biol.* 17:615 (2001)). Of the two Class 1 PI3Ks, Class 1A PI3Ks are heterodimers composed of a catalytic p110 subunit (α, β, δ isoforms) constitutively associated with a regulatory subunit that can be p85α, p55α, p50α, p85β or p55γ. The Class 1B sub-class has one family member, a heterodimer composed of a catalytic p110γ subunit associated with one of two regulatory subunits, p101 or p84 (Fruman et al., *Annu Rev. Biochem.* 67:481 (1998); Suire et al., *Curr. Biol.* 15:566 (2005)). The modular domains of the p85/55/50 subunits include Src Homology (SH2) domains that bind phosphotyrosine residues in a specific sequence context on activated receptor and cytoplasmic tyrosine kinases, resulting in activation and localization of Class 1A PI3Ks. Class 1B PI3K is activated directly by G protein-coupled receptors that bind a diverse repertoire of peptide and non-peptide ligands (Stephens et al., *Cell* 89:105 (1997)); Katso et al., *Annu. Rev. Cell Dev. Biol.* 17:615-675 (2001)). Consequently, the resultant phospholipid products of class I PI3K link upstream receptors with downstream cellular activities including proliferation, survival, chemotaxis, cellular trafficking, motility, metabolism, inflammatory and allergic responses, transcription

and translation (Cantley et al., *Cell* 64:281 (1991); Escobedo and Williams, *Nature* 335:85 (1988); Fantl et al., *Cell* 69:413 (1992)).

In many cases, PIP2 and PIP3 recruit Akt, the product of the human homologue of the viral oncogene *v-Akt*, to the plasma membrane where it acts as a nodal point for many intracellular signaling pathways important for growth and survival (Fantl et al., *Cell* 69:413-423(1992); Bader et al., *Nature Rev. Cancer* 5:921 (2005); Vivanco and Sawyer, *Nature Rev. Cancer* 2:489 (2002)). Aberrant regulation of PI3K, which often increases survival through Akt activation, is one of the most prevalent events in human cancer and has been shown to occur at multiple levels. The tumor suppressor gene *PTEN*, which dephosphorylates phosphoinositides at the 3' position of the inositol ring and in so doing antagonizes PI3K activity, is functionally deleted in a variety of tumors. In other tumors, the genes for the p110α isoform, *PIK3CA*, and for *Akt* are amplified and increased protein expression of their gene products has been demonstrated in several human cancers. Furthermore, mutations and translocation of p85α that serve to up-regulate the p85-p110 complex have been described in human cancers. Finally, somatic missense mutations in *PIK3CA* that activate downstream signaling pathways have been described at significant frequencies in a wide diversity of human cancers (Kang at el., *Proc. Natl. Acad. Sci. USA* 102:802 (2005); Samuels et al., *Science* 304:554 (2004); Samuels et al., *Cancer Cell* 7:561-573 (2005)). These observations show that deregulation of phosphoinositol-3 kinase and the upstream and downstream components of this signaling pathway is one of the most common deregulations associated with human cancers and proliferative diseases (Parsons et al., *Nature* 436:792 (2005); Hennessey at el., *Nature Rev. Drug Disc.* 4:988-1004 (2005)).

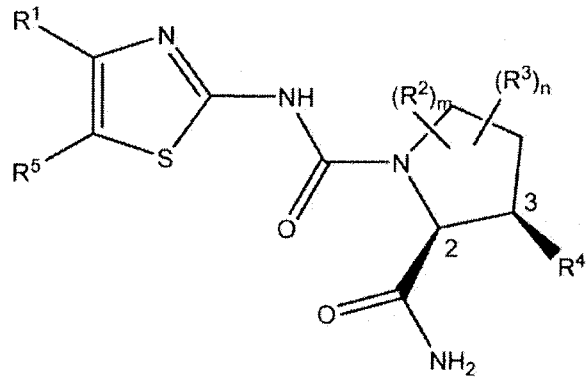
In view of the above, inhibitors of PI3Ks would be of particular value in the treatment of proliferative disease and other disorders. Selectivity towards the PI3K α isoform is desirable, and further desirable properties include improved pharmacokinetic properties and/or chemical stability.

WO2004/096797 discloses certain thiazole derivatives as inhibitors of PI3 kinase and their use as pharmaceutical.

WO 2005/021519 also discloses certain thiazole derivatives as inhibitors of PI3 kinase and their use as pharmaceutical.

It has now been found that the substituted 2-carboxamide cycloamino ureas of the formula I given below have advantageous pharmacological properties and inhibit, for example, the PI3 kinases (phosphatidylinositol 3-kinase). In particular, preferably, these compounds show selectivity for PI3K alpha versus beta and/or delta and/or gamma subtypes in the biochemical and/or in the cellular assay. A further property which is preferably desirable for compounds of formula I includes improved stability, for example, improved chemical stability e.g. in solid form and/or in buffer solution. Hence, the compounds of formula I are suitable, for example, to be used in the treatment of diseases depending on the PI3 kinase (in particular PI3K alpha, such as those showing somatic mutation of PIK3CA or germline mutations or somatic mutation of PTEN), especially proliferative diseases such as tumor diseases and leukaemias.

In a first aspect, the present invention provides compounds of formula I,



(I)

wherein,

m is 0 or 1;

n is 0 or 1;

R^1 represents H, halogen, unsubstituted C_1-C_4-alkyl or substituted C_1-C_4-alkyl;

R^2 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, unsubstituted or substituted amino, halogen or hydroxy;

R^3 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, unsubstituted or substituted amino, halogen or hydroxy;

R^4 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, halogen or hydroxy; or

R^3 and R^4 form, together with the same or different carbon atom to which they are attached, C_3-C_8-cycloalkyl or heterocyclyl;

R^5 is unsubstituted or substituted heteroaryl;

or a salt, solvate, hydrate or prodrug thereof; and

excluding the compound (1*R*,2*S*,5*S*)-3-aza-bicyclo[3.1.0]hexane-2,3-dicarboxylic acid 2-amide 3-{[5-(2-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide}.

The invention may be more fully appreciated by reference to the following description, including the following glossary of terms and the concluding examples. As used herein, the terms "including", "containing" and "comprising" are used herein in their open, non-limiting sense.

Any formula given herein is intended to represent compounds having structures depicted by the structural formula as well as certain variations or forms. In particular, compounds of any formula given herein may have asymmetric centers and therefore exist in different stereoisomeric forms such as different enantiomeric forms. If at least one asymmetrical carbon atom is present in a compound of the formula I, such a compound may exist in optically active form or in the form of a mixture of optical isomers, e. g. in the form of a racemic mixture. Thus an asymmetric carbon atom may be present in the (*R*)-, (*S*)- or (*R,S*)-configuration, preferably in the (*R*)- or (*S*)-configuration. All optical isomers and their mixtures, including the racemic mixtures, are part of the present invention. Thus, any given formula given herein is intended to represent a racemate, one or more enantiomeric forms, one or more diastereomeric forms, one or more atropisomeric forms, and mixtures thereof. Furthermore, certain structures may exist as geometric isomers (e.g. cis and trans isomers), as tautomers, or as atropisomers. For example, substituents at a double bond or a ring may be present in *cis*- (=*Z*-) or *trans* (=*E*-) form. The compounds of the invention may thus be present as mixtures of isomers or preferably as pure isomers, preferably as enantiomer-pure diastereomers or pure enantiomers.

Any formula given herein is intended to represent hydrates, solvates, and polymorphs of such compounds, and mixtures thereof.

Any formula given herein is also intended to represent unlabeled forms as well as isotopically labeled forms of the compounds. Isotopically labeled compounds have structures depicted by the formulas given herein except that one or more atoms are replaced by an atom having a selected atomic mass or mass number. Examples of isotopes that can be incorporated into compounds of the invention include isotopes of hydrogen, carbon, nitrogen, oxygen, phosphorous, fluorine, and chlorine, such as ^{2}H, ^{3}H, ^{11}C, ^{13}C, ^{14}C, ^{15}N, ^{31}P, ^{32}P, ^{18}F ^{35}S, ^{36}Cl, ^{125}I respectively. Various isotopically labeled compounds of the present invention, for example those into which radioactive isotopes such as ^{3}H, ^{13}C, and ^{14}C are incorporated. Such isotopically labeled compounds are useful in metabolic studies (preferably with ^{14}C), reaction kinetic studies (with, for example ^{2}H or ^{3}H), detection or imaging techniques, such as positron emission tomography (PET) or single-photon emission computed tomography (SPECT) including drug or substrate tissue distribution assays, or in radioactive treatment of patients. In particular, an ^{18}F or labeled compound may be particularly preferred for PET or SPECT studies. Further, substitution with heavier isotopes such as deuterium (i.e., ^{2}H) may afford certain therapeutic advantages resulting from greater metabolic stability, for example increased in vivo half-life or reduced dosage requirements. Isotopically labeled compounds of this invention and prodrugs thereof can generally be prepared by carrying out the procedures disclosed in the schemes or in the examples and preparations described below by substituting a readily available isotopically labeled reagent for a non-isotopically labeled reagent.

Further, substitution with heavier isotopes, particularly deuterium (i.e., ^{2}H or D) may afford certain therapeutic advantages resulting from greater metabolic stability, for example increased in vivo half-life or reduced dosage requirements or an improvement in therapeutic index. It is understood that deuterium in this context is regarded as a substituent in the compound of the formula (I). The concentration of such a heavier isotope, specifically deuterium, may be defined by the isotopic enrichment factor. The term "isotopic enrichment factor" as used herein means the ratio between the isotopic abundance and the natural abundance of a specified isotope. If a substituent in a compound of this invention is denoted deuterium, such compound has an isotopic enrichment factor for each designated deuterium atom of at least 3500 (52.5% deuterium incorporation at each designated deuterium atom), at least 4000 (60% deuterium incorporation), at least 4500 (67.5% deuterium incorporation), at least 5000 (75% deuterium incorporation), at least 5500 (82.5% deuterium incorporation), at least 6000 (90% deuterium incorporation), at least 6333.3 (95% deuterium incorporation),

at least 6466.7 (97% deuterium incorporation), at least 6600 (99% deuterium incorporation), or at least 6633.3 (99.5% deuterium incorporation). In the compounds of this invention any atom not specifically designated as a particular isotope is meant to represent any stable isotope of that atom. Unless otherwise stated, when a position is designated specifically as "H" or "hydrogen", the position is understood to have hydrogen at its natural abundance isotopic composition. Accordingly, in the compounds of this invention any atom specifically designated as a deuterium (D) is meant to represent deuterium, for example in the ranges given above.

When referring to any formula given herein, the selection of a particular moiety from a list of possible species for a specified variable is not intended to define the moiety for the variable appearing elsewhere. In other words, where a variable appears more than once, the choice of the species from a specified list is independent of the choice of the species for the same variable elsewhere in the formula (where one or more up to all more general expressions in embodiments characterized as preferred above or below can be replaced with a more specific definition, thus leading to a more preferred embodiment of the invention, respectively).

Where the plural form (e.g. compounds, salts, pharmaceutical preparations, diseases and the like) is used, this includes the singular (e.g. a single compound, a single salt, a single pharmaceutical preparation, a single disease, and the like). "A compound" does not exclude that (e.g. in a pharmaceutical formulation) more than one compound of the formula (I) (or a salt thereof) is present.

Salts are preferably the pharmaceutically acceptable salts of compounds of formula (I) if they are carrying salt-forming groups. Acids/bases required to form the salts are generally known in the field.

The following general definitions shall apply in this specification, unless otherwise specified:

Halogen (or halo) denotes fluorine, bromine, chlorine or iodine, in particular fluorine, chlorine. Halogen-substituted groups and moieties, such as alkyl substituted by halogen (halogenalkyl) can be mono-, poly- or per-halogenated.

Hetero atoms are atoms other than carbon and hydrogen, preferably nitrogen (N), oxygen (O) or sulfur (S), in particular nitrogen.

"**Alkyl**" refers to a straight-chain or branched-chain alkyl group, and includes C_{1-4}alkyl and C_{1-8}alkyl where stated. Such alkyl groups include, for example, methyl, ethyl, n- or iso-propyl, n-, iso-, sec- or tert-butyl, n-pentyl, n-hexyl, n-heptyl, n-octyl, with particular preference given to methyl, ethyl, n-propyl, iso-propyl, n-butyl and iso-butyl. Alkyl may be unsubstituted or substituted. Exemplary substituents include, but are not limited to hydroxy, alkoxy, halogen (especially fluoro), amino, mono- or di-alkyl substituted amino, acetylamino and morpholinyl. An example of a substituted alkyl is trifluoromethyl. Cycloalkyl may also be a substituent to alkyl. An example of such a case is the moiety (alkyl)-cycloalkyl, such as (alkyl)-cyclopropyl or (alkyl)-cyclobutyl, e.g. methyl-cyclopropyl or methyl-cyclobutyl. A more specific example of an (alkyl)-cycloalkyl moiety includes geminal-type of substitution pattern, e.g. 1-alkyl cycloalkyl, such as 1-methyl cyclopropyl. Another example of cycloalkyl as a substituent to alkyl is alkandiyl-cycloalkyl, such as alkandiyl-cyclopropyl, e.g. $-CH_2$-cyclopropyl. C_1-C_8-alkyl is alkyl with from and including 1 up to and including 8 carbon atoms, preferably from and including 1 up to and including 4 carbon atoms (C_1-C_4-alkyl), and is linear or branched; preferably, lower alkyl is butyl, such as n-butyl, sec-butyl, isobutyl, tert-butyl, propyl, such as n-propyl or isopropyl, ethyl or preferably methyl.

Each alkyl part of other groups like "alkoxy", "alkoxyalkyl", "alkoxycarbonyl", "alkoxy-carbonylalkyl", "alkylsulfonyl", "alkylsulfoxyl", "alkylamino", "halogenalkyl" shall have the same meaning as described in the above-mentioned definition of "alkyl".

"**C_{3-8}-Cycloalkyl**" refers to a saturated or partially saturated, monocyclic, fused polycyclic, or spiro polycyclic, carbocycle having from 3 to 8 ring atoms per carbocycle. Illustrative examples of cycloalkyl groups include the following moieties: cyclopropyl, cyclobutyl, cyclpentyl and cylclohexyl. Cycloalkyl may be unsubstituted or substituted; exemplary substituents are provided in the definition for alkyl.

"**Heterocyclyl**" refers to a heterocyclic radical that is saturated or partially saturated and is preferably a monocyclic or in a broader aspect of the invention bicyclic, tricyclic or spirocyclic ring; and has 3 to 24, more preferably 4 to 16, most preferably 5 to 10 and most preferably 5 or 6 ring atoms; wherein one or more, preferably one to four, especially one or two carbon

ring atoms are replaced by a heteroatom, the bonding ring preferably having 4 to 12, especially 5 to 7 ring atoms. The heterocyclic radical (heterocyclyl) may be unsubstituted or substituted by one or more, especially 1 to 3, substituents independently selected from the group consisting of alkyl, or the substituents defined above for substituted alkyl and / or from one or more of the following substituents: oxo (=O), thiocarbonyl (=S), imino(=NH), imino-lower alkyl. Further, heterocyclyl is especially a heterocyclyl radical selected from the group consisting of oxiranyl, aziridinyl, 1,2-oxathiolanyl, tetrahydrofuryl, tetrahydropyranyl, pyrrolinyl, pyrrolidinyl, piperidinyl, piperazinyl, morpholinyl, thiomorpholinyl, (S-oxo or S,S-dioxo)-thiomorpholinyl, azepanyl, diazepanyl, especially 1,4-diazepanyl, tetrahydroquinolyl, tetrahydroisoquinolyl, decahydroquinolyl, octahydroisoquinolyl, isochromanyl, chromanyl and 2,3-dihydro-benzo[1,4]dioxin-6-yl, each of these radicals being unsubstituted or substituted by one or more, preferably up to three, substituents selected from those mentioned above and/or from one or more of the following substituents: oxo (=O), thiocarbonyl (=S), imino(=NH), imino-lower alkyl.

"**Heteroaryl**" refers to a heterocyclic radical that is unsaturated (in particular maximally unsaturated, eg. carrying the highest possible number of conjugated double bonds in the ring(s)), and is preferably a monocyclic or in a broader aspect of the invention bicyclic or tricyclic ring; and has 3 to 24, more preferably 4 to 16, most preferably 5 to 10 and most preferably 5 or 6 ring atoms; wherein one or more, preferably one to four, especially one or two ring atoms are a heteroatom, the bonding ring (i.e. the ring which is bonded to the rest of the molecule) preferably having 4 to 12, especially 5 to 7 ring atoms. The heteroaryl radical may be unsubstituted or substituted by one or more, especially 1 to 3, substituents independently selected from the group consisting of alkyl or the substituents defined above for substituted alkyl and / or from one or more of the following substituents: oxo (=O), thiocarbonyl (=S), imino(=NH), imino-lower alkyl, and, for nitrogen containing heteroaryls, including N-oxides thereof. Further, heteroaryl is especially a heteroaryl radical selected from the group consisting of azirinyl, thienyl (= thiophenyl), furanyl, pyranyl, thiopyranyl, thianthrenyl, isobenzofuranyl, benzofuranyl, chromenyl, 2H-pyrrolyl, pyrrolyl, imidazolyl, imidazolidinyl, benzimidazolyl, pyrazolyl, pyrazinyl, pyrazolidinyl, thiazolyl, isothiazolyl, dithiazolyl, oxazolyl, isoxazolyl, pyridyl, pyrimidinyl, pyridazinyl, indolizinyl, isoindolyl, 3H-indolyl, indolyl, benzimidazolyl, cumaryl, indazolyl, triazolyl, tetrazolyl, purinyl, 4H-quinolizinyl, isoquinolyl, quinolyl, dibenzofuranyl, benzothiophenyl, dibenzothiophenyl, phthalazinyl, naphthyridinyl, quinoxalyl, quinazolinyl, quinazolinyl, cinnolinyl, pteridinyl, carbazolyl, beta-carbolinyl,

phenanthridinyl, acridinyl, perimidinyl, phenanthrolinyl, furazanyl, phenazinyl, phenothiazinyl, phenoxazinyl, chromenyl and benzo[1,3]dioxol-5-yl, each of these radicals being unsubstituted or substituted by one or more, preferably up to three, substituents selected from those mentioned above aryl and/or from one or more of the following substituents: oxo (=O), thiocarbonyl (=S), imino(=NH), imino-lower alkyl and, for nitrogen containing heteroaryls, including N-oxides thereof.

"**Treatment**" includes prophylactic (preventive) and therapeutic treatment as well as the delay of progression of a disease or disorter.

"**PI3 kinase mediated diseases**" (especially PI3K alpha mediated diseases) are especially such disorders that respond in a beneficial way (e.g. amelioration of one or more symptoms, delay of the onset of a disease, up to temporary or complete cure from a disease) to the inhibition of a PI3 kinase, especially inhibition of PI3Kalpha (where the diseases to be treated may include those showing somatic mutation of PIK3CA or germline mutations or somatic mutation of PTEN). Diseases to be treated include especially proliferative diseases such as tumor diseases, including solid tumors, leukaemias, glioblastoma, breast cancer and prostate cancer may be mentioned).

"**Salts**" (which, what is meant by "or salts thereof" or "or a salt thereof"), can be present alone or in mixture with free compound of the formula I and are preferably pharmaceutically acceptable salts. Salt-forming groups in a compound of formula (I) are groups or radicals having basic or acidic properties. Compounds having at least one basic group or at least one basic radical, e.g., amino; a secondary amino group not forming a peptide bond or a pyridyl radical, may form acid addition salts, e.g., with inorganic acids, such as hydrochloric acid, sulfuric acid or a phosphoric acid; or with suitable organic carboxylic or sulfonic acids, e.g., aliphatic mono- or di-carboxylic acids, such as trifluoroacetic acid, acetic acid, propionic acid, glycolic acid, succinic acid, maleic acid, fumaric acid, hydroxymaleic acid, malic acid, tartaric acid, citric acid or oxalic acid; or amino acids, such as arginine or lysine; aromatic carboxylic acids, such as benzoic acid; 2-phenoxy-benzoic acid; 2-acetoxy-benzoic acid; salicylic acid; 4-aminosalicylic acid; aromatic-aliphatic carboxylic acids, such as mandelic acid or cinnamic acid; heteroaromatic carboxylic acids, such as nicotinic acid or isonicotinic acid; aliphatic sulfonic acids, such as methane-, ethane- or 2-hydroxyethanesulfonic acid; or

aromatic sulfonic acids, e.g., benzene-, *p*-toluene- or naphthalene-2-sulfonic acid. When several basic groups are present mono- or poly-acid addition salts may be formed.
Compounds of formula (I) having acidic groups, a carboxy group or a phenolic hydroxy group, may form metal or ammonium salts, such as alkali metal or alkaline earth metal salts, e.g., sodium, potassium, magnesium or calcium salts; or ammonium salts with ammonia or suitable organic amines, such as tertiary monoamines, e.g., triethylamine or tri(2-hydroxyethyl)-amine, or heterocyclic bases, e.g., *N*-ethyl-piperidine or *N,N'*-dimethylpiperazine. Mixtures of salts are possible.
Compounds of formula (I) having both acidic and basic groups can form internal salts.

For isolation or purification purposes it is also possible to use pharmaceutically unacceptable salts, for example picrates or perchlorates. For therapeutic use, only pharmaceutically acceptable salts or free compounds are employed (where applicable in the form of pharmaceutical preparations), and these are therefore preferred. In view of the close relationship between the novel compounds in free form and those in the form of their salts, including those salts that can be used as intermediates, for example in the purification or identification of the novel compounds, any reference to the free compounds hereinbefore and hereinafter is to be understood as referring also to the corresponding salts, as appropriate and expedient.

Compounds of the present invention may also form **solvates and hydrates**, and as such any reference to a compound of formula (I) is therefore to be understood as referring also to the corresponding solvate and/or hydrate of the compound of formula (I), as appropriate and expedient.

The present invention also relates to **pro-drugs** of a compound of formula (I) that convert *in vivo* to the compound of formula (I) as such. Any reference to a compound of formula (I) is therefore to be understood as referring also to the corresponding pro-drugs of the compound of formula (I), as appropriate and expedient.

Combination refers to either a fixed combination in one dosage unit form, or a kit of parts for the combined administration where a compound of the formula I and a combination partner (e.g. an other drug as explained below, also referred to as "therapeutic agent" or "co-agent") may be administered independently at the same time or separately within time intervals, especially where these time intervals allow that the combination partners show a

cooperative, e.g. synergistic effect. The terms "**co-administration**" or "**combined administration**" or the like as utilized herein are meant to encompass administration of the selected combination partner to a single subject in need thereof (e.g. a patient), and are intended to include treatment regimens in which the agents are not necessarily administered by the same route of administration or at the same time. The term "**pharmaceutical combination**" as used herein means a product that results from the mixing or combining of more than one active ingredient and includes both fixed and non-fixed combinations of the active ingredients. The term "**fixed combination**" means that the active ingredients, e.g. a compound of formula I and a combination partner, are both administered to a patient simultaneously in the form of a single entity or dosage. The term "**non-fixed combination**" means that the active ingredients, e.g. a compound of formula I and a combination partner, are both administered to a patient as separate entities either simultaneously, concurrently or sequentially with no specific time limits, wherein such administration provides therapeutically effective levels of the two compounds in the body of the patient. The latter also applies to cocktail therapy, e.g. the administration of three or more active ingredients.

In preferred embodiments, which are preferred independently, collectively or in any combination or sub-combination, the invention relates to a compound of the formula I, in free base form or in salt form, wherein the substituents are as defined herein.

As shown in formula I, the alpha-amide substituent is at the 2-position on the pyrrolidine ring and the stereochemistry is as drawn and the R^4 substituent is at position 3 of the pyrrolidine ring, and each substituent has the defined stereochemistry which is cis- relative to each other.

R^1 preferably represents C_1-C_4-alkyl, most preferably methyl.

R^2 preferably represents C_1-C_4-alkyl, C_1-C_4-alkoxy, di-C_1-C_4-alkyl-amino, halogen or hydroxy.

R^2 more preferably represents methyl, methoxy, dimethylamino, fluoro or hydroxy.

R^2 even more preferably represents methoxy, dimethylamino, fluoro or hydroxy.

R^3 preferably represents C_1-C_4-alkyl, C_1-C_4-alkoxy, di-C_1-C_4-alkyl-amino, halogen or hydroxy.

R^3 more preferably represents methyl, methoxy, dimethylamino, fluoro or hydroxy.

R^3 even more preferably represents methyl, methoxy, dimethylamino, fluoro or hydroxy.

If present (i.e. m=1 and/or n=1) the R^2 or R^3 group may be attached at the 2-and/or 3- and/or 4- and/or 5- position of the pyrrolidine ring of formula I. Most preferably, the R^2 or R^3 group is attached at the 3-postion of the pyrrolidine ring, i.e. on the same carbon which is simultaneously substituted by the R^4 group.

R^4 preferably represents, hydroxy, C_1-C_4-alkyl or C_1-C_4-alkyl substituted by C_1-C_4-alkoxy, unsubstituted or substituted amino, heterocyclyl or heteroaryl.

R^4 more preferably represents, hydroxy, C_1-C_4-alkyl or C_1-C_4-alkyl substituted by C_1-C_4-alkoxy, di-C_1-C_4-alkyl-amino, acetylamino, morpholinyl or pyridyl.

R^4 more preferably represents, hydroxy, methyl or methyl substituted by C_1-C_4-alkoxy, di-C_1-C_4-alkyl-amino, acetylamino, morpholinyl or pyridyl.

R^4 most preferably represents, hydroxy, methyl, methoxymethyl, dimethylamino-methyl, acetylamino-methyl, morpholin-4-ylmethyl or pyridyl-methyl.

As noted above, compounds according to formula I are provided wherein R^3 and R^4 may form, together with the same or different carbon atom to which they are attached, C_3-C_8-cycloalkyl or heterocyclyl, and wherein the compound (1*R*,2*S*,5*S*)-3-aza-bicyclo[3.1.0]hexane-2,3-dicarboxylic acid 2-amide 3-{[5-(2-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide} is excluded.

Compounds according to formula I include those wherein the substitutents are defined as for a compound of formula (I), and

R^4 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, halogen or hydroxy; or

R^3 and R^4 form, together with the same carbon atom to which they are attached, C_3-C_8-cycloalkyl; or

R^3 and R^4 form, together with the same or different carbon atom to which they are attached, heterocyclyl.

In a further, alternative, compounds according to formula I include those wherein the substitutents are defined as for a compound of formula (I), and

R^4 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, halogen or hydroxy; or

R^3 and R^4 form, together with the same carbon atom to which they are attached, C_3-C_8-cycloalkyl or heterocyclyl (preferably cycloalkyl).

Thus, when R^3 and R^4 form a C_3-C_8-cycloalkyl (preferred) or heterocyclyl, it is preferred that the R^3 group is attached at the 3-position of the pyrrolidine ring, i.e. on the same carbon which is simultaneously substituted by the R^4 group.

R^5 as noted above, represents unsubstituted or preferably substituted heteroaryl, substituted by one or more, preferably one moiety independently selected from the group consisting of halogen, hydroxy, cyano, nitro, C_1-C_7-alkyl, per-deutero C_1-C_7-alkyl, C_3-C_{12}-cycloalkyl, (C_1-C_7-alkyl)-C_3-C_{12}-cycloalkyl, (halo-C_1-C_7-alkyl)-C_3-C_{12}-cycloalkyl, amino-C_1-C_7-alkyl, halo-C_1-C_7-alkyl, N-C_1-C_7-alkanoylamino-C_1-C_7-alkyl, N-C_1-C_7-alkanesulfonyl-amino-C_1-C_7-alkyl, pyrrolidino-C_1-C_7-alkyl, oxo- pyrrolidino-C_1-C_7-alkyl, C_1-C_7-alkanesulfinyl, C_1-C_7-alkanesulfonyl, C_1-C_7-alkoxy, amino, N-mono- or N,N-di-(C_1-C_7-alkyl)-amino, N-mono- or N,N-di-(per-deutero C_1-C_7-alkyl)-amino, N-mono- or N,N-di-(C_1-C_7-cycloalkyl)-amino C_1-C_7-alkanoylamino, pyrrolidino, oxo-pyrrolidino, piperidino, piperazin-1-yl, 4-(C_1-C_7-alkyl, C_1-C_7-alkoxy-C_1-C_7-alkyl, halo-C_1-C_7-alkyl or C_3-C_{10}-cycloalkyl)-piperazin-1-yl, 4-(amino-C_1-C_7-alkyl)-piperazin-1-yl, 4-[N-mono- or N,N-di-(C_1-C_7-alkylamino)-C_1-C_7-alkyl]-piperazin-1-yl, morpholino, thiomorpholino, S-oxo- or S,S-dioxothiomorpholino, C_1-C_7-alkanesulfonylamino, carbamoyl, N-mono- or N,N-di-(C_1-C_7-alkyl, C_1-C_7-alkoxy-C_1-C_7-alkyl, amino-C_1-C_7-alkyl and/or (N'-mono- or N',N'-di-(C_1-C_7-alkyl)-amino-C_1-C_7-alkyl)-carbamoyl, pyrrolidin-1-carbonyl, piperidin-1-carbonyl, piperazin-1-carbonyl, 4-(C_1-C_7-alkyl)piperazin-1-carbonyl, morpholin-1-carbonyl, thiomorpholin-1-carbonyl, S-oxo- or S,S-dioxothiomorpholin-1-carbonyl, sulfo,

C_1-C_7-alkanesulfonyl, C_1-C_7-alkanesulfinyl, sulfamoyl, N-mono- or N,N-di-(C_1-C_7-alkyl)-sulfamoyl, morpholinosulfonyl, thiomorpholinosulfonyl, thiazolyl.

R^5 more preferably represents unsubstituted heteroaryl or heteroaryl substituted by one substituent selected from the group consisting of
C_1-C_4-alkyl (in particular tert.-butyl), per-deutero C_1-C_4-alkyl (in particular d_9-tert-butyl), halo-C_1-C_4-alkyl (in particular 1-fluoro-1-methyl-ethyl, 2,2,2-trifluoro-1,1-dimethyl-ethyl), 1-(C_1-C_4-alkyl)-C_3-C_6-cycloalkyl (in particular 1-methyl-cyclopropyl), (halo-C_1-C_4-alkyl)-C_3-C_6-cycloalkyl (in particular 1-trifluoromethyl-cyclopropyl), di-C_1-C_4-alkylamino (in particular diethylamino), di-(per-deutero C_1-C_4-alkyl)amino (in particular d_{10}-diethylamino).

R^5 more preferably represents heteroaryl selected from the group consisting of pyridyl (especially 4-pyridyl), pyrimidinyl (especially 4-pyrimidinyl), pyrazinyl (especially 2-pyrazinyl) and thiazolyl (especially thiazol-4-yl),
wherein said substituent is selected from the group consisting of C_1-C_4-alkyl (in particular tert.-butyl), per-deutero C_1-C_4-alkyl (in particular d_9-tert-butyl), halo-C_1-C_4-alkyl (in particular 1-fluoro-1-methyl-ethyl, 2,2,2-trifluoro-1,1-dimethyl-ethyl), 1-(C_1-C_4-alkyl)-C_3-C_6-cycloalkyl (in particular 1-methyl-cyclopropyl), (halo-C_1-C_4-alkyl)-C_3-C_6-cycloalkyl (in particular 1-trifluoromethyl-cyclopropyl), di-C_1-C_4-alkylamino (in particular diethylamino), di-(per-deutero C_1-C_4-alkyl)amino (in particular d_{10}-diethylamino).

R^5 very preferably represents a heteroaryl selected from the group consisting of 2-(1-methyl-cyclopropyl)-pyridin-4-yl, 2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl, 2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl, 2-(1-(trifluoromethyl)cyclopropyl)thiazole-4-yl, 2-d_9-tert-butyl-pyrimidin-4-yl, 6-tert-butyl-pyrazin-2-yl.

R^5 very preferably represents in another embodiment a heteroaryl selected from the group consisting of 2-(1-methyl-cyclopropyl)-pyridin-4-yl, 2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl, 2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl, 2-(1-(trifluoromethyl)cyclopropyl) thiazole-4-yl, 2-d_9-tert-butyl-pyrimidin-4-yl, 6-tert-butyl-pyrazin-2-yl, 2-tert-butyl-pyridin-4-yl.

It is preferred that when R^5 is substituted pyridyl, e.g. 4-pyridyl, substituted by at least one substituent (as defined herein above), said substituent is at least at the 2-position of the pyridyl group.

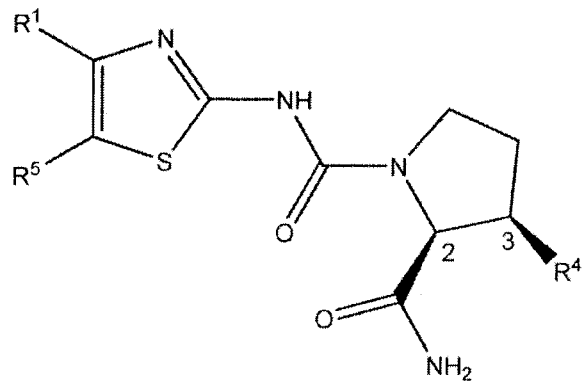
It is preferred that when R^5 is substituted pyrimidinyl, e.g. 4-pyrimidinyl, substituted by at least one substituent (as defined herein above), said substituent is at least at the 2-position of the pyrimidinyl group.

It is preferred that when R^5 is substituted pyrazinyl, e.g. 2-pyrazinyl, substituted by at least one substituent (as defined herein above), said substituent is at least at the 6-position of the pyrazinyl group.

It is preferred that when R^5 is substituted thiazole, e.g. thiazole-4-yl, substituted by at least one substituent (as defined herein above), said substituent is at least at the 2-position of the thiazole group.

An embodiment of the present invention includes compounds of the formula I wherein m and/or n are 0.

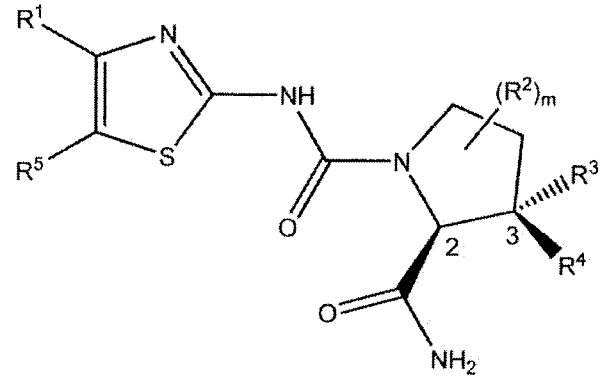
Another embodiment of the present invention includes compounds of the formula I wherein m and n are both 0, that is, wherein the pyrrolidine ring is substituted only by the amide at position 2 and the R^4 group at position 3, i.e. compounds of formula IA:



(IA)

wherein the substitutents are defined as for a compound of formula (I).

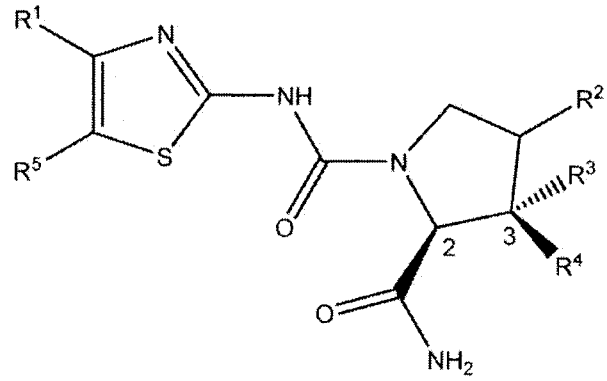
A further embodiment of the present invention includes compounds of the formula I wherein m is 0 or 1 and n is 1. In this embodiment, it is preferred that R^3 is bonded at position 3 of the pyrrolidine ring, i.e. to provide compounds of formula IB:



(IB)

wherein the substituents are defined as for a compound of formula (I).

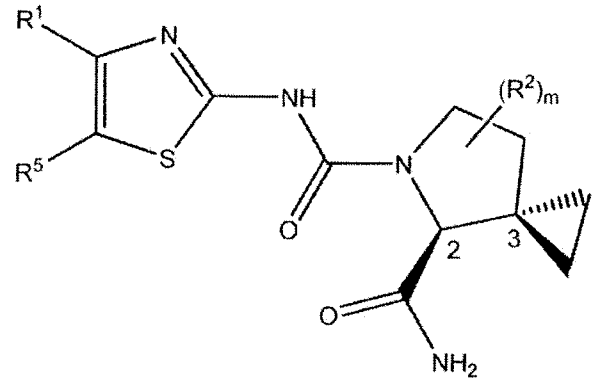
In compounds according to formula IB, when m is 1 (i.e. R^2 is present), the R^2 group is preferably attached at the 4- or 5-position (preferably the 4-position) of the pyrrolidine ring, thus providing compounds of formula IB':



(IB')

In formulae IB or IB', preferably, R^3 is C_1-C_4-alkyl, most preferably methyl.

A further embodiment of the present invention includes compounds of the formula I wherein m is 0 or 1 and n is 1, wherein R^3 is bonded at position 3 of the pyrrolidine ring, and, together with R^4 forms a C_3-C_8-cycloalkyl or heterocyclyl, preferably a C_3-C_8-cycloalkyl, in particular cyclopropyl, i.e. to provide compounds of formula IC:



(IC)

wherein the substitutents are defined as for a compound of formula (I).

In compounds according to formula IC, when m is 1 (i.e. R^2 is present), the R^2 group is preferably attached at the 4- or 5-position (preferably the 4-position) of the pyrrolidine ring.

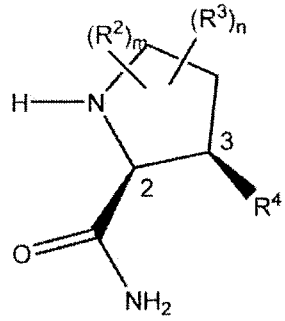
The invention further relates to pharmaceutically acceptable prodrugs of a compound of formula (I), (IA), (IB), (IB') and/or (IC).

The invention further relates to pharmaceutically acceptable metabolites of a compound of formula (I), (IA), (IB), (IB') and/or (IC).

The invention relates especially to the compounds of the formula (I), (IA), (IB), (IB') and/or (IC) given in the Examples, as well as the methods of manufacture described herein.

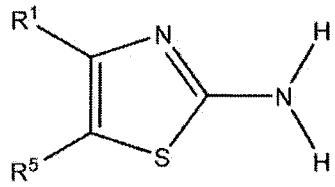
The present invention also relates to processes for the production of a compound of formula (I), (IA), (IB), (IB') and/or (IC). In principle all known processes which convert two different amines into a corresponding urea derivative are suitable and may be applied by using the respective starting material.

Thus, the invention in particular relates to a process which comprises reacting a compound of formula II



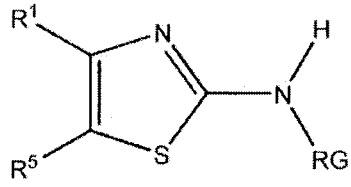
(II)

wherein the substituents are as defined above, either with a compound of formula IIIA



(IIIA)

wherein the substituents are as defined above, in the presence of an activating agent ("method A") or with a compound of formula IIIB



(IIIB)

wherein R^1 is as defined above; RG represents a reactive group (such as imidazolylcarbonyl) ("method B"),

in each case optionally in the presence of a diluent and optionally in the presence of a reaction aid and

recovering the resulting compound of formula I in free form or in form of a salt and, optionally converting a compound of the formula I obtainable according to method A or method B into a different compound of the formula I, and/or converting an obtainable salt of a compound of the formula I into a different salt thereof, and/or converting an obtainable free compound of the formula I into a salt thereof, and/or separating an obtainable isomer of a compound of the formula I from one or more different obtainable isomers of the formula I.

Reaction conditions

The process may be performed according to methods known in the art, or as disclosed below in the Examples. For example a compound of formula II may be reacted with a compound of formula IIIA or IIIB in a solvent, e.g. dimethylformamide, in the presence of a base e.g. an organic amine, e.g. triethylamine.

Where temperatures are given hereinbefore or hereinafter, "about" has to be added, as minor deviations from the numeric values given, e.g. variations of ±10 %, are typically tolerable.

All reactions may take place in the presence of one or more diluents and/or solvents. The starting materials may be used in equimolar amounts; alternatively, a compound may be used in excess, e.g. to function as a solvent or to shift equilibrium or to generally accelerate reaction rates.

Reaction aids, such as acids, bases or catalysts may be added in suitable amounts, as known in the field, required by a reaction and in line with generally known procedures.

Protecting groups

If one or more other functional groups, for example carboxy, hydroxy, amino, sulfhydryl or the like are or need to be protected in a starting material as described herein or any other precursor, because they should not take part in the reaction or disturb the reaction, these are such groups as are usually used in the synthesis of peptide compounds, and also of cephalosporins and penicillins, as well as nucleic acid derivatives and sugars. Protecting groups are such groups that are no longer present in the final compounds once they are removed, while groups that remain as substituents are not protecting groups in the sense used here which are groups that are added at a starting material or intermediate stage and removed to obtain a final compound. Also in the case of conversions of a compound of the formula (I), (IA), (IB), (IB') and/or (IC) into a different compound of the formula (I), (IA), (IB), (IB') and/or (IC), protecting groups may be introduced and removed, if useful or required. The protecting groups may already be present in precursors and should protect the functional groups concerned against unwanted secondary reactions, such as acylations, etherifications, esterifications, oxidations, solvolysis, and similar reactions. It is a characteristic of protecting groups that they lend themselves readily, i.e. without undesired secondary reactions, to removal, typically by acetolysis, protonolysis, solvolysis, reduction, photolysis or also by enzyme activity, for example under conditions analogous to physiological conditions, and

that they are not present in the end-products. The specialist knows, or can easily establish, which protecting groups are suitable with the reactions mentioned above and below.

The protection of such functional groups by such protecting groups, the protecting groups themselves, and their removal reactions are described for example in standard reference works, such as J. F. W. McOmie, "Protective Groups in Organic Chemistry", Plenum Press, London and New York 1973, in T. W. Greene, "Protective Groups in Organic Synthesis", Third edition, Wiley, New York 1999, in "The Peptides"; Volume 3 (editors: E. Gross and J. Meienhofer), Academic Press, London and New York 1981, in "Methoden der organischen Chemie" (*Methods of organic chemistry*), Houben Weyl, 4th edition, Volume 15/I, Georg Thieme Verlag, Stuttgart 1974, in H.-D. Jakubke and H. Jescheit, "Aminosäuren, Peptide, Proteine" (*Amino acids, peptides, proteins*), Verlag Chemie, Weinheim, Deerfield Beach, and Basel 1982, and in Jochen Lehmann, "Chemie der Kohlenhydrate: Monosaccharide und Derivate" (*Chemistry of carbohydrates: monosaccharides and derivatives*), Georg Thieme Verlag, Stuttgart 1974.

Optional Reactions and Conversions

A compound of the formula (I), (IA), (IB), (IB') and/or (IC) may be converted into a different compound of the formula (I), (IA), (IB), (IB') and/or (IC).

In a compound of the formula (I), (IA), (IB), (IB') and/or (IC) wherein a substituent carries an amino or amino-C_1-C_7-alkyl substituent, the amino can be converted into acylamino, e.g. C_1-C_7-alkanoylamino, by reaction with a corresponding C_1-C_7-alkanoylhalogenide, e.g. a corresponding chloride, in the presence of a tertiary nitrogen base, such as triethylamine or pyridine, in the absence or presence of an appropriate solvent, such a methylene chloride, for example at temperatures in the range from –20 to 50 °C, e.g. at about room temperature.

Salts of a compound of formula (I), (IA), (IB), (IB') and/or (IC) with a salt-forming group may be prepared in a manner known *per se*. Acid addition salts of compounds of formula (I), (IA), (IB), (IB') and/or (IC) may thus be obtained by treatment with an acid or with a suitable anion exchange reagent. A salt with two acid molecules (for example a dihalogenide of a compound of formula (I), (IA), (IB), (IB') and/or (IC)) may also be converted into a salt with one acid molecule per compound (for example a monohalogenide); this may be done by

heating to a melt, or for example by heating as a solid under a high vacuum at elevated temperature, for example from 130 to 170°C, one molecule of the acid being expelled per molecule of a compound of formula (I), (IA), (IB), (IB') and/or (IC). Salts can usually be converted to free compounds, e.g. by treating with suitable basic compounds, for example with alkali metal carbonates, alkali metal hydrogencarbonates, or alkali metal hydroxides, typically potassium carbonate or sodium hydroxide.

Stereoisomeric mixtures, e.g. mixtures of diastereomers, can be separated into their corresponding isomers in a manner known *per se* by means of suitable separation methods. Diastereomeric mixtures for example may be separated into their individual diastereomers by means of fractionated crystallization, chromatography, solvent distribution, and similar procedures. This separation may take place either at the level of a starting compound or in a compound of formula (I), (IA), (IB), (IB') and/or (IC) itself. Enantiomers may be separated through the formation of diastereomeric salts, for example by salt formation with an enantiomer-pure chiral acid, or by means of chromatography, for example by HPLC, using chromatographic substrates with chiral ligands.

It should be emphasized that reactions analogous to the conversions mentioned herein may also take place at the level of appropriate intermediates (and are thus useful in the preparation of corresponding starting materials).

Starting materials:

The starting materials of the formulae II and III, as well as other starting materials mentioned herein, e.g. below, can be prepared according to or in analogy to methods that are known in the art, are known in the art and/or are commercially available. Insofar as the production of the starting materials is not particularly described, the compounds are either known or may be prepared analogously to methods known in the art, e.g. in WO 05/021519 or WO04/096797, or as disclosed hereinafter. Novel starting materials, as well as processes for the preparation thereof, are likewise an embodiment of the present invention. In the preferred embodiments, such starting materials are used and the reaction chosen are selected so as to enable the preferred compounds to be obtained.

In the starting materials (including intermediates), which may also be used and/or obtained as salts where appropriate and expedient, the substituents are preferably as defined for a compound of the formula (I), (IA), (IB), (IB') and/or (IC).

Pharmaceutical compositions, uses and methods of treatment

The present invention also relates to use of the compounds of formula (I), (IA), (IB), (IB') and/or (IC) as disclosed herein as pharmaceuticals. The present invention includes in one embodiment compositions comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC), e.g. for human or veterinary use, e.g. where inhibition of PI3K is indicated.

In one embodiment, the invention relates to the treatment of cellular proliferative diseases such as tumor (benign or malignant) and/or cancerous cell growth, e.g. mediated by PI3K. Diseases may include those showing somatic mutation of PIK3CA or germline mutations or somatic mutation of PTEN. In particular, the compounds may be useful in the treatment of human or animal (e.g., murine) cancers, including, for example, sarcoma; lung; bronchus; prostate; breast (including sporadic breast cancers and sufferers of Cowden disease); pancreas; gastrointestinal cancer; colon; rectum; colon carcinoma; colorectal adenoma; thyroid; liver; intrahepatic bile duct; hepatocellular; adrenal gland; stomach; gastric; glioma; glioblastoma; endometrial; melanoma; kidney; renal pelvis; urinary bladder; uterine corpus; uterine cervix; vagina; ovary; multiple myeloma; esophagus; a leukaemia; acute myelogenous leukemia; chronic myelogenous leukemia; lymphocytic leukemia; myeloid leukemia; brain; oral cavity and pharynx; larynx; small intestine; non-Hodgkin lymphoma; melanoma; villous colon adenoma; a neoplasia; a neoplasia of epithelial character; lymphomas; a mammary carcinoma; basal cell carcinoma; squamous cell carcinoma; actinic keratosis; tumor diseases, including solid tumors; a tumor of the neck or head; polycythemia vera; essential thrombocythemia; and myelofibrosis with myeloid metaplasia.

In other embodiments, the condition or disorder (e.g. PI3K-mediated) is selected from the group consisting of: an epidermal hyperproliferation, prostate hyperplasia, a neoplasia, a neoplasia of epithelial character, Cowden syndrome, Lhermitte-Dudos disease or Bannayan-Zonana syndrome, asthma, COPD, ARDS, Loffler's syndrome, eosinophilic pneumonia, parasitic (in particular metazoan) infestation (including tropical eosinophilia), bronchopulmonary aspergillosis, polyarteritis nodosa (including Churg-Strauss syndrome), eosinophilic granuloma, eosinophil-related disorders affecting the airways occasioned by

drug-reaction, psoriasis, contact dermatitis, atopic dermatitis, alopecia areata, erythema multiforme, dermatitis herpetiformis, scleroderma, vitiligo, hypersensitivity angiitis, urticaria, bullous pemphigoid, lupus erythematosus, pemphisus, epidermolysis bullosa acquisita, autoimmune haematogical disorders (e.g. haemolytic anaemia, aplastic anaemia, pure red cell anaemia and idiopathic thrombocytopenia), systemic lupus erythematosus, polychondritis, scleroderma, Wegener granulomatosis, dermatomyositis, chronic active hepatitis, myasthenia gravis, Steven-Johnson syndrome, idiopathic sprue, autoimmune inflammatory bowel disease (e.g. ulcerative colitis and Crohn's disease), endocrine opthalmopathy, Grave's disease, sarcoidosis, alveolitis, chronic hypersensitivity pneumonitis, multiple sclerosis, primary biliary cirrhosis, uveitis (anterior and posterior), interstitial lung fibrosis, psoriatic arthritis, glomerulonephritis, cardiovascular diseases, atherosclerosis, hypertension, deep venous thrombosis, stroke, myocardial infarction, unstable angina, thromboembolism, pulmonary embolism, thrombolytic diseases, acute arterial ischemia, peripheral thrombotic occlusions, and coronary artery disease, reperfusion injuries, retinopathy, such as diabetic retinopathy or hyperbaric oxygen-induced retinopathy, and conditions characterized by elevated intraocular pressure or secretion of ocular aqueous humor, such as glaucoma.

For the above uses the required dosage will of course vary depending on the mode of administration, the particular condition to be treated and the effect desired. In general, satisfactory results are indicated to be obtained systemically at daily dosages of from about 0.03 to 10.0 mg/kg per body weight. An indicated daily dosage in the larger mammal, e.g. humans, is in the range from about 0.5 mg to about 1 g, conveniently administered, for example, in divided doses up to four times a day or in retard form. Suitable unit dosage forms for oral administration comprise from ca. 0.1 to 500 mg active ingredient.

The compounds of formula (I), (IA), (IB), (IB') and/or (IC) may be administered by any conventional route, in particular enterally, e.g. orally, e.g. in the form of tablets or capsules, or parenterally, e.g. in the form of injectable solutions or suspensions, topically, e.g. in the form of lotions, gels, ointments or creams, by inhalation, intranasally, or in a suppository form.

The compounds of formula (I), (IA), (IB), (IB') and/or (IC) may be administered in free form or in pharmaceutically acceptable salt form e.g. as indicated above. Such salts may be

prepared in conventional manner and exhibit the same order of activity as the free compounds.

Consequently, the invention also provides:

- a method for preventing or treating conditions, disorders or diseases mediated by the activation of the PI3, e.g. the PI3 kinase alpha enzyme e.g. such as indicated above, in a subject in need of such treatment, which method comprises administering to said subject an effective amount of a compound of formula (I), (IA), (IB), (IB') and/or (IC) or a pharmaceutically acceptable salt thereof
- use of a compound of formula (I), (IA), (IB), (IB') and/or (IC), in free form or in a pharmaceutically acceptable salt form as a pharmaceutical, e.g. in any of the methods as indicated herein.
- a compound of the formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form for use as pharmaceutical, e.g. in any of the methods as indicated herein, in particular for the use in one or more phosphatidylinositol 3-kinase mediated diseases.
- the use of a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form in any of the methods as indicated herein, in particular for the treatment of one or more phosphatidylinositol 3-kinase mediated diseases.
- the use of a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form in any of the methods as indicated herein, in particular for the manufacture of a medicament for the treatment of one or more phosphatidylinositol 3-kinase mediated diseases.

PI3K serves as a second messenger node that integrates parallel signaling pathways, evidence is emerging that the combination of a PI3K inhibitor with inhibitors of other pathways will be useful in treating cancer and proliferative diseases in humans. Approximately 20-30% of human breast cancers overexpress Her-2/neu-ErbB2, the target for the drug trastuzumab. Although trastuzumab has demonstrated durable responses in some patients expressing Her2/neu-ErbB2, only a subset of these patients respond. Recent work has indicated that this limited response rate can be substantially improved by the combination of trastuzumab with inhibitors of PI3K or the PI3K/AKT pathway (Chan et al.,

Breast Can. Res. Treat. 91:187 (2005), Woods Ignatoski et al., Brit. J. Cancer 82:666 (2000), Nagata et al., Cancer Cell 6:117 (2004)).

A variety of human malignancies express activitating mutations or increased levels of Her1/EGFR and a number of antibody and small molecule inhibitors have been developed against this receptor tyrosine kinase including tarceva, gefitinib and erbitux. However, while EGFR inhibitors demonstrate anti-tumor activity in certain human tumors (e.g., NSCLC), they fail to increase overall patient survival in all patients with EGFR-expressing tumors. This may be rationalized by the fact that many downstream targets of Her1/EGFR are mutated or deregulated at high frequencies in a variety of malignancies, including the PI3K/Akt pathway. For example, gefitinib inhibits the growth of an adenocarcinoma cell line in in vitro assays. Nonetheless, sub-clones of these cell lines can be selected that are resistant to gefitinib that demonstrate increased activation of the PI3/Akt pathway. Down-regulation or inhibition of this pathway renders the resistant sub-clones sensitive to gefitinib (Kokubo et al., Brit. J. Cancer 92:1711 (2005)). Furthermore, in an in vitro model of breast cancer with a cell line that harbors a PTEN mutation and over-expresses EGFR inhibition of both the PI3K/Akt pathway and EGFR produced a synergistic effect (She et al., Cancer Cell 8:287-297(2005)). These results indicate that the combination of gefitinib and PI3K/Akt pathway inhibitors would be an attractive therapeutic strategy in cancer.

The combination of AEE778 (an inhibitor of Her-2/neu/ErbB2, VEGFR and EGFR) and RAD001 (an inhibitor of mTOR, a downstream target of Akt) produced greater combined efficacy that either agent alone in a glioblastoma xenograft model (Goudar et al., Mol. Cancer. Ther. 4:101-112 (2005)).

Anti-estrogens, such as tamoxifen, inhibit breast cancer growth through induction of cell cycle arrest that requires the action of the cell cycle inhibitor p27Kip. Recently, it has been shown that activation of the Ras-Raf-MAP Kinase pathway alters the phosphorylation status of p27Kip such that its inhibitory activity in arresting the cell cycle is attenuated, thereby contributing to anti-estrogen resistance (Donovan, et al, J. Biol. Chem. 276:40888, (2001)). As reported by Donovan et al., inhibition of MAPK signaling through treatment with MEK inhibitor reversed the aberrant phosphorylation status of p27 in hormone refractory breast cancer cell lines and in so doing restored hormone sensitivity. Similarly, phosphorylation of p27Kip by Akt also abrogates its role to arrest the cell cycle (Viglietto et al., Nat Med. 8:1145 (2002)).

Accordingly, the present invention provides, in a further aspect, compounds of formulae (I), (IA), (IB), (IB') and/or (IC) for use in the treatment of hormone dependent cancers, such as breast and prostate cancers. By this use, it is aimed to reverse hormone resistance commonly seen in these cancers with conventional anticancer agents.

In hematological cancers, such as chronic myelogenous leukemia (CML), chromosomal translocation is responsible for the constitutively activated BCR-Abl tyrosine kinase. The afflicted patients are responsive to imatinib, a small molecule tyrosine kinase inhibitor, as a result of inhibition of Abl kinase activity. However, many patients with advanced stage disease respond to imatinib initially, but then relapse later due to resistance-conferring mutations in the Abl kinase domain. In vitro studies have demonstrated that BCR-Ab1 employs the Ras-Raf kinase pathway to elicit its effects. In addition, inhibiting more than one kinase in the same pathway provides additional protection against resistance-conferring mutations.

Accordingly, in another aspect, the present invention provides the compounds of formulae (I), (IA), (IB), (IB') and/or (IC) for use in combination with at least one additional agent selected from the group of kinase inhibitors, such as Gleevec®, in the treatment of hematological cancers, such as chronic myelogenous leukemia (CML). By this use, it is aimed to reverse or prevent resistance to said at least one additional agent.

Because activation of the PI3K/Akt pathway drives cell survival, inhibition of the pathway in combination with therapies that drive apoptosis in cancer cells, including radiotherapy and chemotherapy, will result in improved responses (Ghobrial et al., CA Cancer J. Clin 55:178-194 (2005)). As an example, combination of PI3 kinase inhibitor with carboplatin demonstrated synergistic effects in both in vitro proliferation and apoptosis assays as well as in in vivo tumor efficacy in a xenograft model of ovarian cancer (Westfall and Skinner, Mol. Cancer Ther. 4:1764-1771 (2005)).

In addition to cancer and proliferative diseases, there is accumulating evidence that inhibitors of Class 1A and 1B PI3 kinases would be therapeutically useful in others disease areas. The inhibition of p110β, the PI3K isoform product of the PIK3CB gene, has been shown to be involved in shear-induced platelet activation (Jackson et al., Nature Medicine 11:507-514 (2005)). Thus, a PI3K inhibitor that inhibits p110β would be useful as a single

agent or in combination in anti-thrombotic therapy. The isoform p110δ, the product of the PIK3CD gene, is important in B cell function and differentiation (Clayton et al., J. Exp. Med. 196:753-763 (2002)), T-cell dependent and independent antigen responses (Jou et al., Mol. Cell. Biol. 22:8580-8590 (2002)) and mast cell differentiation (Ali et al., Nature 431:1007-1011 (2004)). Thus, it is expected that p110δ-inhibitors would be useful in the treatment of B-cell driven autoimmune diseases and asthma. Finally, the inhibition of p110γ, the isoform product of the PI3KCG gene, results in reduced T, but not B cell, response (Reif et al., J. Immunol. 173:2236-2240 (2004)) and its inhibition demonstrates efficacy in animal models of autoimmune diseases (Camps et al., Nature Medicine 11:936-943 (2005), Barber et al., Nature Medicine 11:933-935 (2005)).

The invention further provides pharmaceutical compositions comprising at least one compound of formula (I), (IA), (IB), (IB') and/or (IC), together with a pharmaceutically acceptable excepient suitable for administration to a human or animal subject, either alone or together with another therapeutic agent, for example another anticancer agent.

The invention further provides methods of treating human or animal subjects suffering from a cellular proliferative disease, such as cancer. The invention thus provides methods of treating a human or animal subject in need of such treatment, comprising administering to the subject a therapeutically effective amount of a compound of formula (I), (IA), (IB), (IB') and/or (IC) either alone or in combination with one or more other therapeutic agents, e.g. other anticancer agents. In particular, compositions will either be formulated together as a combination therapeutic or administered separately. Suitable anticancer agents for use with a compound of formula I include, but are not limited to, one or more compounds selected from the group consisting of kinase inhibitors, anti-estrogens, anti androgens, other inhibitors, cancer chemotherapeutic drugs, alkylating agents, chelating agents, biological response modifiers, cancer vaccines, agents for antisense therapy as set forth below:

A. Kinase Inhibitors: Kinase inhibitors for use as anticancer agents in conjunction with a compound of the formula (I), (IA), (IB), (IB') and/or (IC) include inhibitors of Epidermal Growth Factor Receptor (EGFR) kinases such as small molecule quinazolines, for example gefitinib (US 5457105, US 5616582, and US 5770599), ZD-6474 (WO 01/32651), erlotinib (Tarceva®, US 5,747,498 and WO 96/30347), and lapatinib (US 6,727,256 and WO 02/02552); Vascular Endothelial Growth Factor Receptor (VEGFR) kinase inhibitors, including SU-11248 (WO 01/60814), SU 5416 (US 5,883,113 and WO 99/61422), SU 6668

(US 5,883,113 and WO 99/61422), CHIR-258 (US 6,605,617 and US 6,774,237), vatalanib or PTK-787 (US 6,258,812), VEGF-Trap (WO 02/57423), B43-Genistein (WO-09606116), fenretinide (retinoic acid p-hydroxyphenylamine) (US 4,323,581), IM-862 (WO 02/62826), bevacizumab or Avastin® (WO 94/10202), KRN-951, 3-[5-(methylsulfonylpiperadine methyl)-indolyl]-quinolone, AG-13736 and AG-13925, pyrrolo[2,1-f][1,2,4]triazines, ZK-304709, Veglin®, VMDA-3601, EG-004, CEP-701 (US 5,621,100), Cand5 (WO 04/09769); Erb2 tyrosine kinase inhibitors such as pertuzumab (WO 01/00245), trastuzumab, and rituximab; Akt protein kinase inhibitors, such as RX-0201; Protein Kinase C (PKC) inhibitors, such as LY-317615 (WO 95/17182), and perifosine (US 2003171303); Raf/Map/MEK/Ras kinase inhibitors including sorafenib (BAY 43-9006), ARQ-350RP, LErafAON, BMS-354825 AMG-548, and others disclosed in WO 03/82272; Fibroblast Growth Factor Receptor (FGFR) kinase inhibitors; Cell Dependent Kinase (CDK) inhibitors, including CYC-202 or roscovitine (WO 97/20842 and WO 99/02162); Platelet-Derived Growth Factor Receptor (PDGFR) kinase inhibitors such as CHIR-258, 3G3 mAb, AG-13736, SU-11248 and SU6668; and Bcr-Abl kinase inhibitors and fusion proteins such as STI-571 or Gleevec® (imatinib).

B. Anti-Estrogens: Estrogen-targeting agents for use in anticancer therapy in conjunction with a compoound of formula (I), (IA), (IB), (IB') and/or (IC) include Selective Estrogen Receptor Modulators (SERMs) including tamoxifen, toremifene, raloxifene; aromatase inhibitors including Arimidex® or anastrozole; Estrogen Receptor Downregulators (ERDs) including Faslodex® or fulvestrant.

C. Anti-Androgens: Androgen-targeting agents for use in anticancer therapy in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) include flutamide, bicalutamide, finasteride, aminoglutethamide, ketoconazole, and corticosteroids.

D. Other Inhibitors: Other inhibitors for use as anticancer agents in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) include protein farnesyl transferase inhibitors including tipifarnib or R-115777 (US 2003134846 and WO 97/21701), BMS-214662, AZD-3409, and FTI-277; topoisomerase inhibitors including merbarone and diflomotecan (BN-80915); mitotic kinesin spindle protein (KSP) inhibitors including SB-743921 and MKI-833; proteasome modulators such as bortezomib or Velcade® (US 5,780,454), XL-784; and cyclooxygenase 2 (COX-2) inhibitors including non-steroidal antiinflammatory drugs I (NSAIDs).

E. Cancer Chemotherapeutic Drugs: Particular cancer chemotherapeutic agents for use as anticancer agents in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or

(IC) include anastrozole (Arimidex®), bicalutamide (Casodex®), bleomycin sulfate (Blenoxane®), busulfan (Myleran®), busulfan injection (Busulfex®), capecitabine (Xeloda®), N4-pentoxycarbonyl-5-deoxy-5-fluorocytidine, carboplatin (Paraplatin®), carmustine (BiCNU®), chlorambucil (Leukeran®), cisplatin (Platinol®), cladribine (Leustatin®), cyclophosphamide (Cytoxan® or Neosar®), cytarabine, cytosine arabinoside (Cytosar-U®), cytarabine liposome injection (DepoCyt®), dacarbazine (DTIC-Dome®), dactinomycin (Actinomycin D, Cosmegan), daunorubicin hydrochloride (Cerubidine®), daunorubicin citrate liposome injection (DaunoXome®), dexamethasone, docetaxel (Taxotere®, US 2004073044), doxorubicin hydrochloride (Adriamycin®, Rubex®), etoposide (Vepesid®), fludarabine phosphate (Fludara®), 5-fluorouracil (Adrucil®, Efudex®), flutamide (Eulexin®), tezacitibine, Gemcitabine (difluorodeoxycitidine), hydroxyurea (Hydrea®), Idarubicin (Idamycin®), ifosfamide (IFEX®), irinotecan (Camptosar®), L-asparaginase (ELSPAR®), leucovorin calcium, melphalan (Alkeran®), 6-mercaptopurine (Purinethol®), methotrexate (Folex®), mitoxantrone (Novantrone®), mylotarg, paclitaxel (Taxol®), phoenix (Yttrium90/MX-DTPA), pentostatin, polifeprosan 20 with carmustine implant (Gliadel®), tamoxifen citrate (Nolvadex®), teniposide (Vumon®), 6-thioguanine, thiotepa, tirapazamine (Tirazone®), topotecan hydrochloride for injection (Hycamptin®), vinblastine (Velban®), vincristine (Oncovin®), and vinorelbine (Navelbine®).

F. Alkylating Agents: Alkylating agents for use in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) include VNP-40101M or cloretizine, oxaliplatin (US 4,169,846, WO 03/24978 and WO 03/04505), glufosfamide, mafosfamide, etopophos (US 5,041,424), prednimustine; treosulfan; busulfan; irofluven (acylfulvene); penclomedine; pyrazoloacridine (PD-115934); O6-benzylguanine; decitabine (5-aza-2-deoxycytidine); brostallicin; mitomycin C (MitoExtra); TLK-286 (Telcyta®); temozolomide; trabectedin (US 5,478,932); AP-5280 (Platinate formulation of Cisplatin); porfiromycin; and clearazide (meclorethamine).

G. Chelating Agents: Chelating agents for use in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) include tetrathiomolybdate (WO 01/60814); RP-697; Chimeric T84.66 (cT84.66); gadofosveset (Vasovist®); deferoxamine; and bleomycin optionally in combination with electorporation (EPT).

H. Biological Response Modifiers: Biological response modifiers, such as immune modulators, for use in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) include staurosprine and macrocyclic analogs thereof, including UCN-01, CEP-701 and

midostaurin (see WO 02/30941, WO 97/07081, WO 89/07105, US 5,621,100, WO 93/07153, WO 01/04125, WO 02/30941, WO 93/08809, WO 94/06799, WO 00/27422, WO 96/13506 and WO 88/07045); squalamine (WO 01/79255); DA-9601 (WO 98/04541 and US 6,025,387); alemtuzumab; interferons (e.g. IFN-a, IFN-b etc.); interleukins, specifically IL-2 or aldesleukin as well as IL-1, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-12, and active biological variants thereof having amino acid sequences greater than 70% of the native human sequence; altretamine (Hexalen®); SU 101 or leflunomide (WO 04/06834 and US 6,331,555); imidazoquinolines such as resiquimod and imiquimod (US 4,689,338, 5,389,640, 5,268,376, 4,929,624, 5,266,575, 5,352,784, 5,494,916, 5,482,936, 5,346,905, 5,395,937, 5,238,944, and 5,525,612); and SMIPs, including benzazoles, anthraquinones, thiosemicarbazones, and tryptanthrins (WO 04/87153, WO 04/64759, and WO 04/60308).

I. Cancer Vaccines: Anticancer vaccines for use in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) include Avicine® (Tetrahedron Lett. 26:2269-70 (1974)); oregovomab (OvaRex®); Theratope® (STn-KLH); Melanoma Vaccines; GI-4000 series (GI-4014, GI-4015, and GI-4016), which are directed to five mutations in the Ras protein; GlioVax-1; MelaVax; Advexin® or INGN-201 (WO 95/12660); Sig/E7/LAMP-1, encoding HPV-16 E7; MAGE-3 Vaccine or M3TK (WO 94/05304); HER-2VAX; ACTIVE, which stimulates T-cells specific for tumors; GM-CSF cancer vaccine; and Listeria monocytogenes-based vaccines.

J. Antisense Therapy: Anticancer agents for use in conjunction with a compound of formula (I), (IA), (IB), (IB') and/or (IC) also include antisense compositions, such as AEG-35156 (GEM-640); AP-12009 and AP-11014 (TGF-beta2-specific antisense oligonucleotides); AVI-4126; AVI-4557; AVI-4472; oblimersen (Genasense®); JFS2; aprinocarsen (WO 97/29780); GTI-2040 (R2 ribonucleotide reductase mRNA antisense oligo) (WO 98/05769); GTI-2501 (WO 98/05769); liposome-encapsulated c-Raf antisense oligodeoxynucleotides (LErafAON) (WO 98/43095); and Sirna-027 (RNAi-based therapeutic targeting VEGFR-1 mRNA).

A compound of formula (I), (IA), (IB), (IB') and/or (IC) can also be combined in a pharmaceutical composition with bronchiodilatory or antihistamine drugs substances. Such bronchiodilatory drugs include anticholinergic or antimuscarinic agents, in particular glycopyrrolate, ipratropium bromide, oxitropium bromide, and tiotropium bromide, OrM3, aclidinium, CHF5407, GSK233705 and β-2- adrenoreceptor agonists such as salbutamol, terbutaline, salmeterol, carmoterol, milveterol and, especially, indacaterol and formoterol. Co-therapeutic antihistamine drug substances include cetirizine hydrochloride, clemastine

fumarate, promethazine, loratadine, desloratadine diphenhydramine and fexofenadine hydrochloride.

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and one or more compounds that are useful for the treatment of a thrombolytic disease, heart disease, stroke, etc. Such compounds include aspirin, a streptokinase, a tissue plasminogen activator, a urokinase, a anticoagulant, antiplatelet drugs (e.g, PLAVIX; clopidogrel bisulfate), a statin (e.g., LIPITOR or Atorvastatin calcium), ZOCOR (Simvastatin), CRESTOR (Rosuvastatin), etc.), a Beta blocker (e.g., Atenolol), NORVASC (amlodipine besylate), and an ACE inhibitor (e.g., lisinopril).

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and one or more compounds that are useful for the treatment of antihypertension. Such compounds include ACE inhibitors, lipid lowering agents such as statins, LIPITOR (Atorvastatin calcium), calcium channel blockers such as NORVASC (amlodipine besylate).

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and one or more compounds selected from the group consisting of fibrates, beta-blockers, NEPI inhibitors, Angiotensin-2 receptor antagonists and platelet aggregation inhibitors.

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and a compound suitable for the treatment of inflammatory diseases, including rheumatoid arthritis. Such compound may be selected from the group consisting of TNF-α inhibitors such as anti-TNF-α monoclonal antibodies (such as REMICADE, CDP-870) and D2E7 (HUMIRA) and TNF receptor immunoglobulin fusion molecules (such as ENBREL), IL-1 inhibitors, receptor antagonists or soluble IL-1Rα(e.g. KINERET or ICE inhibitors), nonsterodial anti-inflammatory agents (NSAIDS), piroxicam, diclofenac, naproxen, flurbiprofen, fenoprofen, ketoprofen ibuprofen, fenamates, mefenamic acid, indomethacin, sulindac, apazone, pyrazolones, phenylbutazone, aspirin, COX-2 inhibitors (such as CELEBREX (celecoxib), PREXIGE (lumiracoxib)), metalloprotease inhibitors (preferably MMP-13 selective inhibitors), p2x7 inhibitors, α2αinhibitors,

NEUROTIN, pregabalin, low dose methotrexate, leflunomide, hydroxyxchloroquine, d-penicillamine, auranofin or parenteral or oral gold.

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and a compound suitable for the treatment of osteoarthritis. Such compound may be selected from the group consisting of standard non-steroidal anti-inflammatory agents (hereinafter NSAID's) such as piroxicam, diclofenac, propionic acids such as naproxen, flurbiprofen, fenoprofen, ketoprofen and ibuprofen, fenamates such as mefenamic acid, indomethacin, sulindac, apazone, pyrazolones such as phenylbutazone, salicylates such as aspirin, COX-2 inhibitors such as celecoxib, valdecoxib, lumiracoxib and etoricoxib, analgesics and intraarticular therapies such as corticosteroids and hyaluronic acids such as hyalgan and synvisc.

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and an antiviral agent and/or an antisepsis compound. Such antiviral agent may be selected from the group consisting of Viracept, AZT, acyclovir and famciclovir. Such antisepsis compound may be selected from the group consisting of Valant.

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and one or more agents selected from the group consisting of CNS agents such as antidepressants (sertraline), anti-Parkinsonian drugs (such as deprenyl, L-dopa, Requip, Mirapex; MAOB inhibitors (such as selegine and rasagiline); comP inhibitors (such as Tasmar); A-2 inhibitors; dopamine reuptake inhibitors; NMDA antagonists; Nicotine agonists; Dopamine agonists; and inhibitors of neuronal nitric oxide synthase).

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and one or more anti-Alzheimer's drugs. Such anti-Alzheimer Drug may be selected from the group consisting of donepezil, tacrine, $\alpha 2\delta$inhibitors, NEUROTIN, pregabalin, COX-2 inhibitors, propentofylline or metryfonate.

The invention provides in a further aspect a combination comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) and anosteoporosis agents and/or an immunosuppressant agent. Such osteoporosis agents ma be selected from the group consisting of EVISTA

(raloxifene hydrochloride), droloxifene, lasofoxifene or fosomax. Such immunosuppressant agents may be selected from the group consisting of FK-506 and rapamycin.

In another aspect of the preferred embodiments, kits that include one or more compound of formula (I), (IA), (IB), (IB') and/or (IC) and a combination partner as disclosed herein are provided. Representative kits include a PI3K inhibitor compound (e.g., a compound of formula (I), (IA), (IB), (IB') and/or (IC)) and a package insert or other labeling including directions for treating a cellular proliferative disease by administering a PI3K inhibitory amount of the compound(s).

In general, the compounds of formula (I), (IA), (IB), (IB') and/or (IC) will be administered in a therapeutically effective amount by any of the accepted modes of administration for agents that serve similar utilities. The actual amount of the compound of formula (I), (IA), (IB), (IB') and/or (IC), i.e., the active ingredient, will depend upon numerous factors such as the severity of the disease to be treated, the age and relative health of the subject, the potency of the compound used, the route and form of administration, and other factors. The drug can be administered more than once a day, preferably once or twice a day. All of these factors are within the skill of the attending clinician. Therapeutically effective amounts of compounds of formulas I may range from about 0.05 to about 50 mg per kilogram body weight of the recipient per day; preferably about 0.1-25 mg/kg/day, more preferably from about 0.5 to 10 mg/kg/day. Thus, for administration to a 70 kg person, the dosage range would most preferably be about 35-70 mg per day.

In general, compounds of formula (I), (IA), (IB), (IB') and/or (IC) will be administered as pharmaceutical compositions by any one of the following routes: oral, systemic (e.g., transdermal, intranasal or by suppository), or parenteral (e.g., intramuscular, intravenous or subcutaneous) administration. The preferred manner of administration is oral using a convenient daily dosage regimen that can be adjusted according to the degree of affliction. Compositions can take the form of tablets, pills, capsules, semisolids, powders, sustained release formulations, solutions, suspensions, elixirs, aerosols, or any other appropriate compositions. Another preferred manner for administering compounds of the formula I is inhalation. This is an effective method for delivering a therapeutic agent directly to the respiratory tract.

The choice of formulation depends on various factors such as the mode of drug administration and bioavailability of the drug substance. For delivery via inhalation the compound can be formulated as liquid solution, suspensions, aerosol propellants or dry powder and loaded into a suitable dispenser for administration. There are several types of pharmaceutical inhalation devices-nebulizer inhalers, metered dose inhalers (MDI) and dry powder inhalers (DPI). Nebulizer devices produce a stream of high velocity air that causes the therapeutic agents (which are formulated in a liquid form) to spray as a mist that is carried into the patient's respiratory tract. MDI's typically are formulation packaged with a compressed gas. Upon actuation, the device discharges a measured amount of therapeutic agent by compressed gas, thus affording a reliable method of administering a set amount of agent. DPI dispenses therapeutic agents in the form of a free flowing powder that can be dispersed in the patient's inspiratory air-stream during breathing by the device. In order to achieve a free flowing powder, the therapeutic agent is formulated with an excipient such as lactose. A measured amount of the therapeutic agent is stored in a capsule form and is dispensed with each actuation.

The inventions also relates to formulations wherein the particle size of a compound of formula I between 10 – 1000 nm, preferably 10 – 400 nm. Such pharmaceutical formulations have been developed especially for drugs that show poor bioavailability based upon the principle that bioavailability can be increased by increasing the surface area i.e., decreasing particle size. For example, U.S. 4,107,288 describes a pharmaceutical formulation having particles in the size range from 10 to 1,000 nm in which the active material is supported on a crosslinked matrix of macromolecules. U.S. 5,145,684 describes the production of a pharmaceutical formulation in which the drug substance is pulverized to nanoparticles (average particle size of 400 nm) in the presence of a surface modifier and then dispersed in a liquid medium to give a pharmaceutical formulation that exhibits remarkably high bioavailability. Both documents are included by reference.

In a further aspect, the invention provides pharmaceutical compositions comprising a (therapeutically effective amount) of a compound of formula (I), (IA), (IB), (IB') and/or (IC), and at least one pharmaceutically acceptable excipient. Acceptable excipients are non-toxic, aid administration, and do not adversely affect the therapeutic benefit of the compound of formula (I), (IA), (IB), (IB') and/or (IC). Such excipient may be any solid, liquid, semi-solid or,

in the case of an aerosol composition, gaseous excipient that is generally available to one of skill in the art.

Solid pharmaceutical excipients include starch, cellulose, talc, glucose, lactose, sucrose, gelatin, malt, rice, flour, chalk, silica gel, magnesium stearate, sodium stearate, glycerol monostearate, sodium chloride, dried skim milk and the like.

Liquid and semisolid excipients may be selected from glycerol, propylene glycol, water, ethanol and various oils, including those of petroleum, animal, vegetable or synthetic origin, e.g., peanut oil, soybean oil, mineral oil, sesame oil, etc. Preferred liquid carriers, particularly for injectable solutions, include water, saline, aqueous dextrose, and glycols.

Compressed gases may be used to disperse a compound of the formula I in aerosol form. Inert gases suitable for this purpose are nitrogen, carbon dioxide, etc. Other suitable pharmaceutical excipients and their formulations are described in Remington's Pharmaceutical Sciences, edited by E. W. Martin (Mack Publishing Company, I8th ed., I990).

The amount of the compound in a formulation can vary within the full range employed by those skilled in the art. Typically, the formulation will contain, on a weight percent (wt%) basis, from about 0.01-99.99 wt% of a compound of formula I based on the total formulation, with the balance being one or more suitable pharmaceutical excipients. Preferably, the compound is present at a level of about 1-80 wt%.

The invention further relates to pharmaceutical compositions comprising (i.e. containing or consisting of) at least one compound of formula (I), (IA), (IB), (IB') and/or (IC) and at least one pharmaceutically acceptable excipient.

Pharmaceutical compositions comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form in association with at least one pharmaceutical acceptable excipient (such as a carrier and/or diluent) may be manufactured in conventional manner by mixing the components.

Combined pharmaceutical compositions comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form and further comprising a combination partner (either in one dosage unit form or as a kit of parts) in association with at least one pharmaceutical acceptable carrier and/or diluent may be manufactured in conventional manner by mixing with a pharmaceutically acceptable carrier and/or diluent with said active ingredients.

Consequently, the invention provides in further aspects

- a combined pharmaceutical composition, e.g. for use in any of the methods described herein, comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or pharmaceutically acceptable salt form in association with a pharmaceutically acceptable diluent and/or carrier.
- a combined pharmaceutical composition comprising a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form as active ingredient; one or more pharmaceutically acceptable carrier material(s) and / or diluents and optionally one or more further drug substances. Such combined pharmaceutical composition may be in the form of one dosage unit form or as a kit of parts.
- a combined pharmaceutical composition comprising a therapeutically effective amount of a compound of formula (I), (IA), (IB), (IB') and/or (IC) in free form or in pharmaceutically acceptable salt form and a second drug substance, for simultaneous or sequential administration.
- a method as defined above comprising co-administration, e.g. concomitantly or in sequence, of a therapeutically effective non-toxic amount of a compound of formula (I), (IA), (IB), (IB') and/or (IC) or a pharmaceutically acceptable salt thereof, and at least a second drug substance, e.g. as indicated above.
- a pharmaceutical combination, e.g. a kit, comprising a) a first agent which is a compound of formula (I), (IA), (IB), (IB') and/or (IC) as disclosed herein, in free form or in pharmaceutically acceptable salt form, and b) at least one co-agent, e.g. as indicated above; whereby such kit may comprise instructions for its administration.

The following examples of compounds formula (I), (IA), (IB), (IB') and/or (IC) illustrate the invention without limiting the scope thereof. Methods for preparing such compounds are described.

Temperatures are measured in degrees Celsius. Unless otherwise indicated, the reactions take place under an argon atmosphere at rt and the MS are obtained with ESI. The following HPLC and LC-MS methods are used in the preparation and analysis of the Intermediates and Examples:

HPLC (Method A):

System: Agilent 1100 Series

Column: HP Hypersil BDS C18, 4 x 125 mm, 5 micron

Temperature: 25°C

Eluent A: H_2O, containing 0.1% v/v TFA

Eluent B: acetonitrile, containing 0.1% v/v TFA

Gradient: 10% → 100% B in 5 min, 2.5 min with 100% B, then → 10% B in 1 min

Flow Rate: 1.5 mL/min

Detection: UV 215 nm

HPLC (Method B):

System: Agilent 1100 Series

Column: Macherey-Nagel Nucleosil 100-3 C18HD, 4 x 125 mm, 3 micron

Temperature: 30°C

Eluent A: H_2O, containing 0.1% v/v TFA

Eluent B: acetonitrile, containing 0.1% v/v TFA

Gradient: 2% → 100% B in 7 min, 2 min with 100% B, then → 2% B in 1 min

Flow Rate: 1.0 mL/min

Detection: UV 215 nm

LC-MS (Method A):

System: Waters Acquity UPLC with Waters Micromass ZQ 2000 ESI+/-

Column: Acquity HSS T3 C18, 2.1 x 50 mm, 1.8 micron

Temperature: 50 °C

Eluent A: H_2O, containing 0.05% v/v HCOOH and 3.75 mM ammonium acetate

Eluent B: acetonitrile, containing 0.04% HCOOH

Gradient: 2% → 98% B in 4.3 min, 0.7 min with 98% B, then → 2% B in 0.1 min and 0.9 min with 2% B

Flow Rate: 1.0 mL/min

LC-MS (Method B):

System: Agilent 1100 Series; MS: G1946D

Column: Symmetry C8, 2.1 x 50mm, 3.5 micron

Eluent A: H2O, containing 0.1% v/v HCOOH

Eluent B: acetonitrile, containing 0.1% v/v HCOOH
Gradient: 0 – 3.3 min: 5% to 95% of B
Flow rate: 1.0 mL/min

LC-MS (Method C):
System: Waters Acquity UPLC
Column: Acquity HSS T3 C18, 2.1 x 50 mm, 1.8 micron
Eluent A: H_2O, containing 0.05% v/v HCOOH and 0.05% ammonium acetate
Eluent B: acetonitrile, containing 0.04% HCOOH
Gradient: 2% → 98% B in 1.7 min, 0.45 min with 98% B, then → 2% B in 0.04 min
Flow Rate: 1.2 ml/min

LC-MS (Method D):
System: Waters Aquity UPLC; MS: Waters AQ Detector
Column: Aquity HSS, 1.8 µm 2.1 x 50mm, 3/pk
Eluent A: H_2O, containing 0.1% v/v HCOOH
Eluent B: acetonitrile, containing 0.1% v/v HCOOH
Gradient: 0 – 1.5 min: 10% to 95% of B, then 1 min: 95% B
Flow Rate: 1.2 mL/min

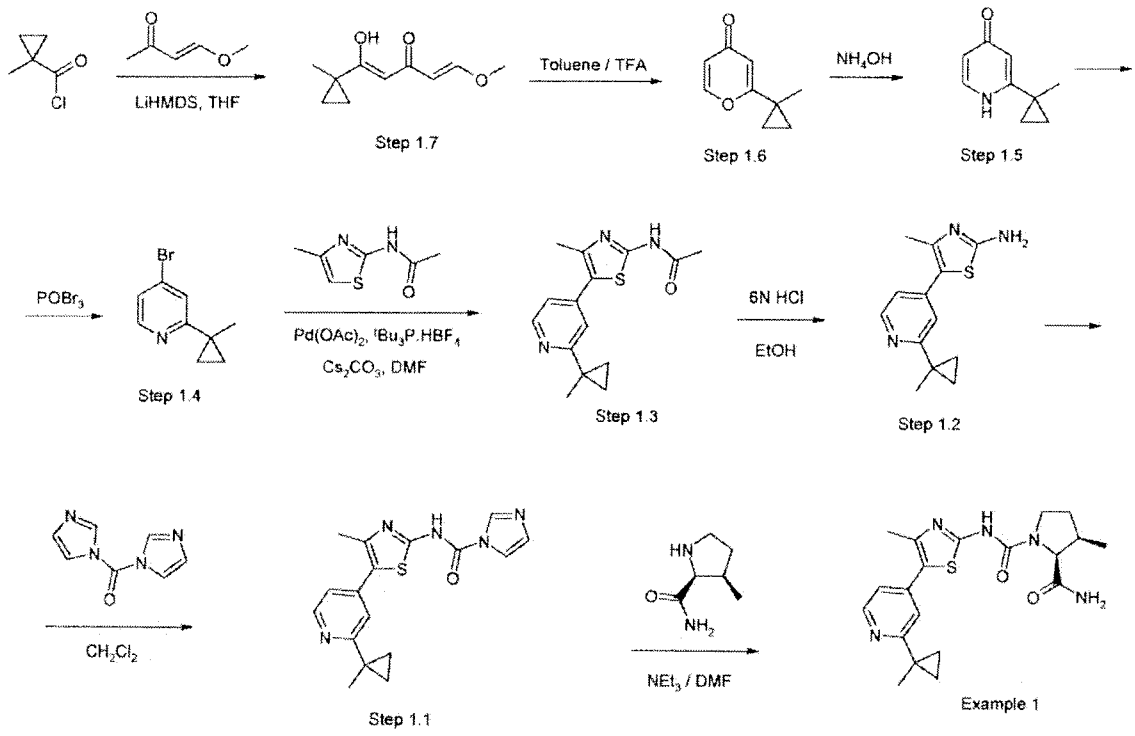
ESI-MS:
Instrument: Micromass Platform II
Eluent: 15% v/v methanol in water containing 0.2% v/v of a 25% ammonium hydroxide solution
Flow rate: 0.05 mL/min

In the following examples, the abbreviations given below are used:

atm.	atmosphere
CDI	1,1'-carbonyldiimidazole
CH_2Cl_2	dichloromethane
DCE	1,2-dichloroethane
DMF	N,N-dimethylformamide
DMSO	dimethyl sulfoxide
Et_2O	diethyl ether

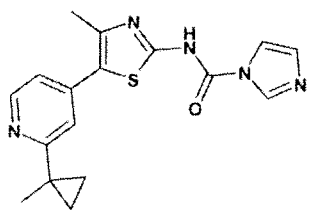
EtOAc	ethyl acetate
EtOH	ethanol
Et_3N	triethylamine
eq	equivalent(s)
h	hour(s)
Hex	hexanes
HPLC	High Performance Liquid Chromatography
HV	high vacuum
LC-MS	liquid chromatography coupled with mass spectrometry
LiHMDS	lithium bis(trimethylsilyl)amide
MeOH	methanol
mL	milliliter(s)
min	minute(s)
MS-ESI	electrospray ionisation mass spectrometry
MTBE	methyl tert-butyl ether
MW	microwave
R_f	ratio of fronts in TLC
rt	room temperature
TFA	trifluoroacetic acid
THF	tetrahydrofuran
TLC	thin layer chromatography
t_R	retention time
UV	ultraviolet

Example 1: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



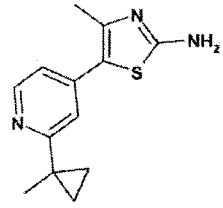
Et_3N (0.53 mmol) was added to a mixture of imidazole-1-carboxylic acid {4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide (Step 1.1) (0.177 mmol) and (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide (Step 1.8) (0.194 mmol) in DMF (1 mL) at rt. After stirring for 20 h, the reaction mixture was concentrated. The residue was purified using a RediSep® silica gel column followed by trituration with EtOAc to afford the title compound as a white solid. HPLC: t_R = 4.27 min (method B); LC-MS: t_R= 1.34 min, $[M+H]^+$ 400 (method A); TLC: R_f = 0.12 (95:5 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.92 (br s, 1H), 8.41 (d, 1H), 7.38 (br s, 1H), 7.28 (s, 1H), 7.16 (d, 1H), 6.98 (br s, 1H), 4.15 (m, 1H), 3.69 (m, 1H), 3.37 (m, 1H), 2.40 (s, 3H), 2.36 (m, 1H), 1.95 (m, 1H), 1.69 (m, 1H), 1.48 (s, 3H), 1.18 (m, 2H), 0.98 (d, 3H), 0.80 (m, 2H).

Step 1.1: Imidazole-1-carboxylic acid {4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide



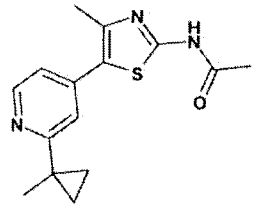
A mixture of 4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-ylamine (Step 1.2) (28.5 mmol) and CDI (42.8 mmol) in CH_2Cl_2 (330 mL) was refluxed for 11 h. After cooling to rt, the reaction mixture was filtered to afford the title compound as a light green solid.

Step 1.2: 4-Methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-ylamine



A mixture of N-{4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-acetamide (Step 1.3) (34.8 mmol), a 6N aqueous solution of HCl (53 mL) and EtOH (265 mL) was stirred for 5 h at 85°C, cooled to rt and then concentrated. The residue was slowly diluted with a saturated solution of $NaHCO_3$ and then extracted with EtOAc (3X). The combined organic phases were successively washed with a saturated solution of $NaHCO_3$ and brine, dried (Na_2SO_4), filtered and concentrated. The residue was triturated with CH_2Cl_2 and then filtered to afford the title compound as a yellow-green solid. HPLC: t_R = 3.55 min (method B); LC-MS: t_R= 1.03 min, $[M+H]^+$ 246; TLC: R_f = 0.26 (1:2 Hex/EtOAc).

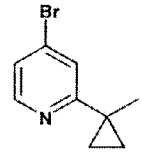
Step 1.3: N-{4-Methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-acetamide



A mixture of 2-acetamido-4-methylthiazole [7336-51-8] (60.4 mmol), cesium carbonate (110 mmol), tri-*tert*-butylphosphinium tetrafluoroborate (10.99 mmol), palladium (II) acetate (5.49 mmol) and 4-bromo-2-(1-methyl-cyclopropyl)-pyridine (Step 1.4) (54.9 mmol) in DMF (230 mL) was stirred for 3.5 h at 100°C. After cooling to rt, the reaction mixture was filtered and then concentrated. The residue was diluted with a saturated solution of $NaHCO_3$ and extracted with EtOAc (3X). The combined organic phases were successively washed with a saturated solution of $NaHCO_3$ and brine, dried (Na_2SO_4), filtered and concentrated. The residue was purified using a RediSep® silica gel column to afford the title compound as a

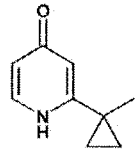
pale yellow solid. HPLC: t_R = 4.37 min (method B); LC-MS: t_R= 1.47 min, $[M+H]^+$ 288; TLC: R_f = 0.26 (1:2 Hex/EtOAc).

Step 1.4: 4-Bromo-2-(1-methyl-cyclopropyl)-pyridine



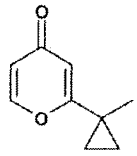
A mixture of 2-(1-methyl-cyclopropyl)-1H-pyridin-4-one (Step 1.5) (13.4 mmol) and $POBr_3$ (14.74 mmol) was stirred for 15 min at 85°C and then 15 min at 120°C. After cooling slightly, the reaction mixture was poured into a saturated solution of $NaHCO_3$ and extracted with CH_2Cl_2 (2X). The combined organic phases were washed with a saturated solution of $NaHCO_3$, dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel column chromatography to afford the title compound as a brown oil. HPLC: t_R = 4.11 min (method B); LC-MS: t_R= 2.39 min, $[M+H]^+$ 212/214; TLC: R_f = 0.31 (CH_2Cl_2).

Step 1.5: 2-(1-Methyl-cyclopropyl)-1H-pyridin-4-one



A mixture of 2-(1-methyl-cyclopropyl)-pyran-4-one (Step 1.6) (66.6 mmol) and a 28-30% aqueous solution of ammonium hydroxide (182 mL) was stirred for 1 h at 65°C. After cooling to rt, the reaction mixture was decanted to remove a dark brown solid and then concentrated. The residue was diluted with MeOH and re-concentrated (3X) to afford the title compound as a brown-orange solid. HPLC: t_R = 3.56 min (method B); LC-MS: t_R= 0.87 min, $[M+H]^+$ 151; TLC: R_f = 0.18 (9:1 CH_2Cl_2/MeOH).

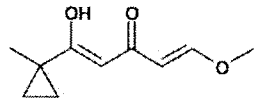
Step 1.6: 2-(1-Methyl-cyclopropyl)-pyran-4-one



A mixture of (1Z,4E)-1-hydroxy-5-methoxy-1-(1-methyl-cyclopropyl)-penta-1,4-dien-3-one one (Step 1.7) (111 mmol) and TFA (221 mmol) in toluene (175 mL) was stirred for 15.5 h at

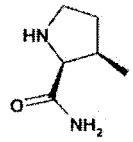
rt and then concentrated. The residue was purified using a RediSep® silica gel column to afford the title compound as a white solid. LC-MS: t_R = 1.48 min, $[M+H]^+$ 151; TLC: R_f = 0.26 (EtOAc).

Step 1.7: (1Z,4E)-1-Hydroxy-5-methoxy-1-(1-methyl-cyclopropyl)-penta-1,4-dien-3-one



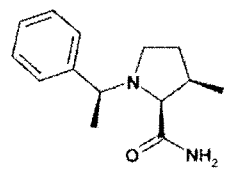
LiHMDS (1M in THF, 845 mmol) was added dropwise to a solution of trans-4-methoxy-3-buten-2-one [51731-17-0] (845 mmol) in THF (2 L) at -78°C. After stirring for 15 min, a solution of 1-methyl-cyclopropanecarbonyl chloride [16480-05-0] (407 mmol) in THF (100 mL) was added. The resulting mixture was allowed to warm to rt over 2.5 h and then quenched by addition of a saturated solution of NH_4Cl. The mixture was extracted with Et_2O (2X). The combined organic phases were successively washed with a saturated solution of $NaHCO_3$ and brine, dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel column chromatography to afford the title compound as a yellow solid. ESI-MS: $[M+H]^+$ 183; TLC: R_f = 0.29 (9:1 Hex/EtOAc).

Step 1.8: (2S,3R)-3-Methyl-pyrrolidine-2-carboxylic acid amide



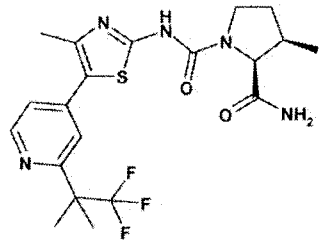
A mixture of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 1.9) (4.76 mmol) and 10% Pd on charcoal (0.947 mmol) in MeOH (20 mL) was hydrogenated for 46 h at rt. The reaction mixture was then filtered through a Fluoropore Membrane Filter (0.2 µm FG) and evaporated. The residue was dissolved in CH_2Cl_2 and evaporated to dryness to afford the title compound as a white solid. ESI-MS: $[M+H]^+$ 129; TLC: R_f = 0.10 (1:3 Hex/EtOAc).

Step 1.9: (2S,3R)-3-Methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide



Trimethylaluminum in toluene (2 M, 6.46 mmol) was added dropwise to a mixture of NH_4Cl (6.47 mmol) in toluene (3.2 mL) at 0 °C with the formation of methane gas. The reaction mixture was allowed to warm to rt, stirred for a further 15 min and then slowly treated with (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester (prepared as described in *Tet. Lett.* **1997**, *38*, 85-88) (6.47 mmol). After stirring for 56 h, the mixture was cooled to 0°C, quenched with 1M HCl and then washed with CH_2Cl_2 (3X). The aqueous phase was basified with a saturated solution of $NaHCO_3$ and extracted with CH_2Cl_2 (3X). The combined organic layers were dried (Na_2SO_4), filtered and concentrated. The residue was purified using a RediSep® silica gel column to afford the title compound as a colorless oil. ESI-MS: $[M+H]^+$ 233; HPLC: t_R 2.35 min (method A).

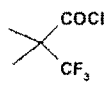
Example 2: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 1 but in Step 1.7, 3,3,3-trifluoro-2,2-dimethyl-propionyl chloride (Step 2.1) was used instead of 1-methyl-cyclopropanecarbonyl chloride.

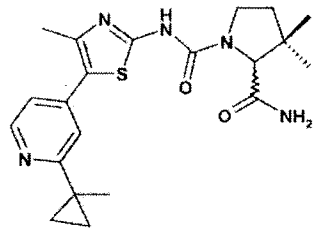
The title compound was obtained as a white solid. HPLC: t_R = 5.16 min (method B); LC-MS: t_R= 2.29 min, $[M+H]^+$ 456 (method A); TLC: R_f = 0.17 (95:5 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.98 (br s, 1H), 8.59 (d, 1H), 7.54 (s, 1H), 7.40 (d, 1H), 7.39 (br s, 1H), 6.98 (br s, 1H), 4.15 (m, 1H), 3.69 (m, 1H), 3.37 (m, 1H), 2.40 (s, 3H), 2.36 (m, 1H), 1.95 (m, 1H), 1.69 (m, 1H), 1.61 (s, 6H), 0.97 (d, 3H).

Step 2.1: 3,3,3-Trifluoro-2,2-dimethyl-propionyl chloride



DMF (3 drops) was added to a solution of 3,3,3-trifluoro-2,2-dimethyl-propionic acid [889940-13-0] (179 mmol) in CH_2Cl_2 (160 mL) at rt. Slowly added a solution of oxalyl chloride (197 mmol) in CH_2Cl_2 (20 mL). After stirring for 14 h, the reaction mixture was carefully evaporated (500 mbar, 33°C) to afford the title compound (volatile!) as a yellow solution.

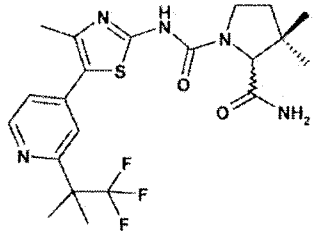
Example 3: (rac)-3,3-Dimethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 1 but (rac)-3,3-dimethyl-pyrrolidine-2-carboxylic acid amide (prepared as described in *J. Org. Chem.*, **2008**, *73*, 3946-3949) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

The title compound was obtained as a yellow solid. HPLC: t_R = 2.90 min (method A); LC-MS: t_R= 1.32 min, $[M+H]^+$ 414 (method A); TLC: R_f = 0.17 (9:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.97 (br s, 1H), 8.43 (d, 1H), 7.39 (m, 2H), 7.31 (br s, 1H), 6.97 (m, 1H), 3.82 (m, 1H), 3.65 (m, 1H), 3.46 (m, 1H), 2.43 (s, 3H), 1.86 (m, 1H), 1.62 (m, 1H), 1.48 (s, 3H), 1.19 (m, 2H), 1.04 (s, 3H), 0.99 (s, 3H), 0.85 (m, 2H).

Example 4: (rac)-3,3-Dimethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide)

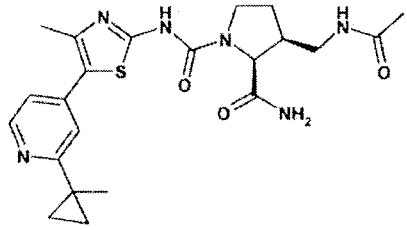


The title compound was prepared in analogy to the procedure described in Example 2 but (rac)-3,3-dimethyl-pyrrolidine-2-carboxylic acid amide (prepared as described in *J. Org. Chem.*, **2008**, *73*, 3946-3949) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

The title compound was obtained as a yellow solid. HPLC: t_R = 5.38 min (method B); LC-MS: t_R= 2.12 min, $[M+H]^+$ 470 (method A); TLC: R_f = 0.12 (9:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.93 (br s, 1H), 8.58 (d, 1H), 7.54 (s, 1H), 7.39 (d, 1H), 7.39 (br s, 1H),

6.97 (br s, 1H), 3.82 (m, 1H), 3.65 (m, 1H), 3.46 (m, 1H), 2.39 (s, 3H), 1.86 (m, 1H), 1.61 (m, 1H), 1.59 (s, 6H), 1.04 (s, 3H), 1.00 (s, 3H).

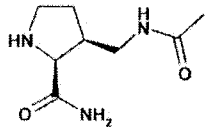
Example 5: (2S,3S)-3-(Acetylamino-methyl)-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 1 but (2S,3S)-3-(acetylamino-methyl)-pyrrolidine-2-carboxylic acid amide (Step 5.1) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

The title compound was obtained as a white solid. HPLC: t_R = 4.00 min (method B); LC-MS: t_R= 1.04 min, $[M+H]^+$ 457 (method A); TLC: R_f = 0.17 (9:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.96 (br s, 1H), 8.42 (d, 1H), 7.80 (br s, 1H), 7.49 (br s, 1H), 7.29 (s, 1H), 7.17 (d, 1H), 7.10 (br s, 1H), 4.25 (m, 1H), 3.72 (m, 1H), 3.37 (m, 1H), 3.23 (m, 1H), 2.92 (m, 1H), 2.41 (s, 3H), 2.39 (m, 1H), 2.00 (m, 1H), 1.82 (s, 3H), 1.73 (m, 1H), 1.49 (s, 3H), 1.19 (m, 2H), 0.81 (m, 2H).

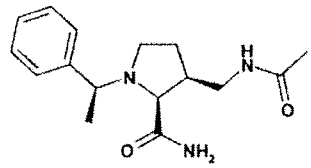
Step 5.1: (2S,3S)-3-(Acetylamino-methyl)-pyrrolidine-2-carboxylic acid amide



The title compound was prepared in analogy to the procedure described in Step 1.8 but (2S,3S)-3-(acetylamino-methyl)-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 5.2) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide. Moreover, 10% Pd on charcoal, wet with 50% H2O (Aldrich 330108) was used instead of the dry catalyst.

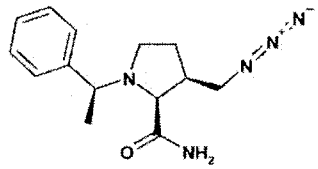
The title compound was obtained as an off-white solid. ESI-MS: $[M+H]^+$ 186; TLC: R_f = 0.08 (200:20:1 CH_2Cl_2/MeOH/conc. NH_4OH).

Step 5.2: (2S,3S)-3-(Acetylamino-methyl)-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide



Thioacetic acid (2.312 mmol) was added to (2S,3S)-3-azidomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 5.3) (0.578 mmol) at rt with the formation of nitrogen gas. After stirring for 16 h, the reaction mixture was diluted with Et_2O, the solids were removed by filtration and the filtrate was concentrated. The residue was purified by silica gel column chromatography to afford the title compound as a light yellow oil (thiol odor). HPLC: t_R = 3.71 min (method B); LC-MS: t_R= 0.64 min, $[M+H]^+$ 290; TLC: R_f = 0.38 (200:20:1 CH_2Cl_2/MeOH/conc. NH_4OH).

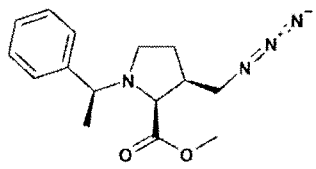
Step 5.3: (2S,3S)-3-Azidomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide



The title compound was prepared in analogy to the procedure described in Step 1.9 but (2S,3S)-3-azidomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester (Step 5.4) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester. Moreover, a 1:1 saturated solution of $NaHCO_3$/saturated solution of Rochelle's salt was used for the basification and the basified aqueous layer was extracted exhaustively with THF.

The title compound was obtained as a yellow oil. HPLC: t_R = 2.50 min (method A); ESI-MS: $[M+H]^+$ 274; TLC: R_f = 0.26 (3:1 Hex/EtOAc).

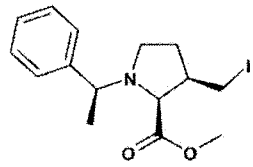
Step 5.4: (2S,3S)-3-Azidomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester



Sodium azide (5.34 mmol) was added to a solution of (2S,3R)-3-iodomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester (Step 5.5) (3.56 mmol) in DMF (30 mL) at rt. After 18 h, the reaction mixture was poured onto water and extracted with MTBE (2X). The

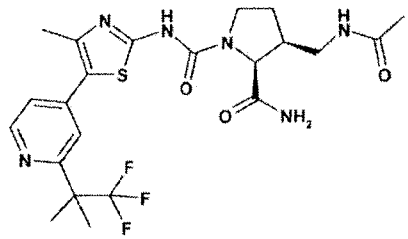
combined organic phases were washed with brine, dried (Na_2SO_4), filtered and concentrated. The residue was purified using a RediSep® silica gel column to to afford the title compound as a brown oil. HPLC: t_R = 3.26 min (method A); ESI-MS: $[M+H]^+$ 289.

Step 5.5: (2S,3R)-3-Iodomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester



A solution of [but-3-enyl-((S)-1-phenyl-ethyl)-amino]-acetic acid methyl ester [432555-77-6] (20.22 mmol) in THF (10 mL) was slowly added to a solution of lithium diisopropylamide (24.26 mmol) in 1:2 hexanes/THF (30 mL) at -78°C. The reaction mixture was warmed to 0°C, stirred for 1 h and then re-cooled to -78°C. A solution of zinc bromide (50.5 mmol) in Et_2O (40 mL) was added and the reaction mixture was then warmed to rt. After stirring for 1 h, the mixture was cooled to 0°C and iodine (22.24 mmol) was added in portions. The reaction mixture was stirred at 0°C for 2 h and at rt for another 2 h, diluted with Et_2O and then successively washed with a saturated solution of $Na_2S_2O_3$ and a saturated solution of NH_4Cl. The aqueous layers were each back-extracted with Et_2O. The combined organic phases were dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel column chromatography to afford the title compound as a red oil. HPLC: t_R = 3.46 min (method A); ESI-MS: $[M+H]^+$ 374.

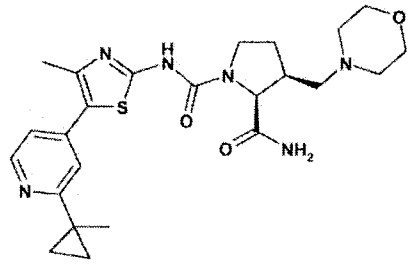
Example 6: (2S,3S)-3-(Acetylamino-methyl)-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 2 but (2S,3S)-3-(acetylamino-methyl)-pyrrolidine-2-carboxylic acid amide (Step 5.1) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

The title compound was obtained as a white solid. HPLC: t_R = 4.73/4.78 min (method B); LC-MS: t_R= 1.81 min, $[M+H]^+$ 513 (method A); TLC: R_f = 0.17 (9:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 11.05 (br s, 1H), 8.59 (d, 1H), 7.80 (br s, 1H), 7.55 (s, 1H), 7.49 (br s, 1H), 7.40 (m, 1H), 7.10 (br s, 1H), 4.25 (m, 1H), 3.72 (m, 1H), 3.39 (m, 1H), 3.23 (m, 1H), 2.92 (m, 1H), 2.41 (s, 3H), 2.39 (m, 1H), 2.00 (m, 1H), 1.82 (s, 3H), 1.72 (m, 1H), 1.61 (s, 6H).

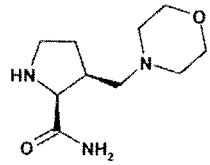
Example 7: (2S,3S)-3-Morpholin-4-ylmethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 1 but (2S,3S)-3-morpholin-4-ylmethyl-pyrrolidine-2-carboxylic acid amide (Step 7.1) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

The title compound was obtained as a pale yellow solid. HPLC: t_R = 2.39 min (method A); LC-MS: t_R= 0.90 min, $[M+H]^+$ 485 (method A); TLC: R_f = 0.09 (19:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.90 (br s, 1H), 8.41 (d, 1H), 7.35 (br s, 1H), 7.29 (s, 1H), 7.17 (d, 1H), 7.02 (br s, 1H), 4.26 (br s, 1H), 3.69 (m, 1H), 3.58 (m, 4H), 3.40 (m, 1H), 2.58 (m, 1H), 2.41 (s, 3H), 2.38 (m, 4H), 2.35 (m, 1H), 2.17 (m, 1H), 2.00 (m, 1H), 1.75 (m, 1H), 1.49 (s, 3H), 1.19 (m, 2H), 0.81 (m, 2H).

Step 7.1: (2S,3S)-3-Morpholin-4-ylmethyl-pyrrolidine-2-carboxylic acid amide

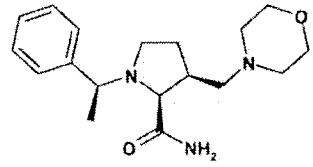


The title compound was prepared in analogy to the procedure described in Step 1.8 but (2S,3S)-3-morpholin-4-ylmethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide

(Step 7.2) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide. Moreover, 10% Pd on charcoal, wet with 50% H2O (Aldrich 330108) was used instead of the dry catalyst.

The title compound was obtained as a colorless oil. ESI-MS: $[M+H]^+$ 214; TLC: R_f = 0.14 (200:20:1 CH_2Cl_2/MeOH/conc. NH_4OH).

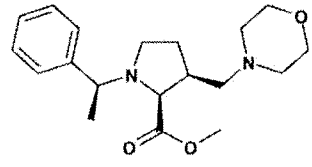
Step 7.2: (2S,3S)-3-Morpholin-4-ylmethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide



The title compound was prepared in analogy to the procedure described in Step 1.9 but (2S,3S)-3-morpholin-4-ylmethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester (Step 7.3) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester. Moreover, the basified aqueous layer was extracted exhaustively with THF instead of CH_2Cl_2.

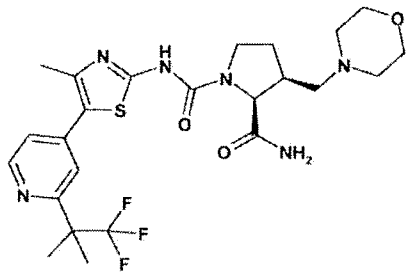
The title compound was obtained as a yellow oil. HPLC: t_R = 2.18 min (method A); ESI-MS: $[M+H]^+$ 318.

Step 7.3: (2S,3S)-3-Morpholin-4-ylmethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester



A mixture of(2S,3R)-3-iodomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester (Step 5.5) (7.07 mmol), K_2CO_3 (21.22 mmol) and morpholine (10.61 mmol) in acetonitrile (24 mL) was stirred at 50°C for 62 h. The reaction mixture was poured onto ice water and extracted with EtOAc (3X). The combined organic layers were successively washed with water and brine, dried (Na_2SO_4), filtered and concentrated. The residue was purified using a RediSep® silica gel column to afford the title compound as a yellow oil. HPLC: t_R = 2.56 min (method A); ESI-MS: $[M+H]^+$ 333.

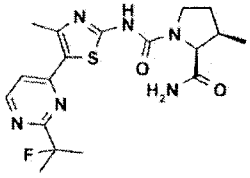
Example 8: (2S,3S)-3-Morpholin-4-ylmethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 2 but (2S,3S)-3-morpholin-4-ylmethyl-pyrrolidine-2-carboxylic acid amide (Step 7.1) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

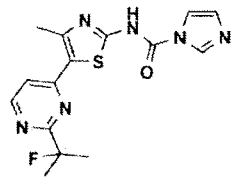
The title compound was obtained as a yellow foam. HPLC: t_R = 4.81 min (method B); LC-MS: t_R= 1.53 min, $[M+H]^+$ 541 (method A); TLC: R_f = 0.22 (9:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 11.02 (br s, 1H), 8.60 (d, 1H), 7.55 (s, 1H), 7.41 (d, 1H), 7.36 (br s, 1H), 7.03 (br s, 1H), 4.26 (m, 1H), 3.69 (m, 1H), 3.58 (m, 4H), 3.41 (m, 1H), 2.58 (m, 1H), 2.41 (s, 3H), 2.38 (m, 4H), 2.35 (m, 1H), 2.17 (m, 1H), 2.01 (m, 1H), 1.75 (m, 1H), 1.61 (s, 6H).

Example 9: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({5-[2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl]-4-methyl-thiazol-2-yl}-amide)



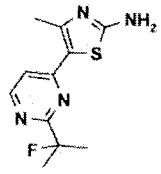
The title compound was prepared in analogy to the procedure described in Example 1 but imidazole-1-carboxylic acid {5-[2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl]-4-methyl-thiazol-2-yl}-amide (Step 9.1) was used instead of imidazole-1-carboxylic acid {4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide. LC-MS: t_R = 1.53 min, M+H = 407.0 (method B); ^{1}H-NMR (d_6-DMSO, 600.13 MHz) 11 (s, br, 1H) 8.7 (s, 1H), 7.5 (s, 1H), 7.35 (s, 1H), 6.95 (s, 1H), 4.15 (s, 1H), 3.65 (m, 1H), 3.4 (m, 1H); 2.6 (s, 3H), 2.3 (m, 1H), 1.95 (m, 1H), 1.75 (s, 3H), 1.7 (s, 3H), 1.7 (s, 3H), 1.7 (m, 1H), 0.95 (d, 3H).

Step 9.1: Imidazole-1-carboxylic acid {5-[2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl]-4-methyl-thiazol-2-yl}-amide



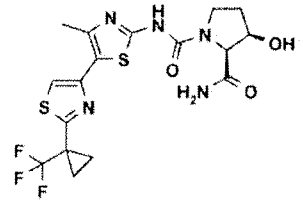
CDI (0.10 g) was added to a stirred solution of 5-[2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl]-4-methyl-thiazol-2-ylamine (Step 9.2) (0.15 g) in CH_2Cl_2 (4 mL) at rt. The reaction mixture was then stood for 56 h at 25°C and the title compound isolated by filtration.

Step 9.2: 5-[2-(1-Fluoro-1-methyl-ethyl)-pyrimidin-4-yl]-4-methyl-thiazol-2-ylamine



A mixture of *N'*-[5-((E)-3-dimethylamino-acryloyl)-4-methyl-thiazol-2-yl]-*N,N*-dimethyl-formamidine [507487-90-3] (2.1 g), 2-fluoro-2-methylpropionamidine (1.7 g prepared by an analogous procedure to that described for Example 1 in EP 0227415) and 2-methoxyethanol (3.8 mL) was stirred at rt for 30 min. NaOH (0.3 g) was added and the mixture stirred at 125°C for 2 h. After cooling to rt, water was added and the mixture evaporated to dryness and purified by normal phase chromatography on silica gel eluting with CH_2Cl_2:MeOH:conc. NH_4OH 97.5:2:0.5 to give the title compound.

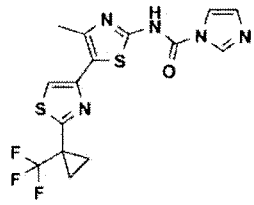
Example 10: (2S,3R)-3-Hydroxy-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide}



The title compound was prepared in analogy to the procedure described in Example 1 but imidazole-1-carboxylic acid [4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide (Step 10.1) was used instead of imidazole-1-carboxylic acid {4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide, and (2S,3R)-3-hydroxy-pyrrolidine-2-carboxylic acid amide (described in H. Fukushima et al., Biorg. Med Chem. **2004**, *12*, 6053; H. Ji et al., J. Med Chem. **2006**, *49*, 6254) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic

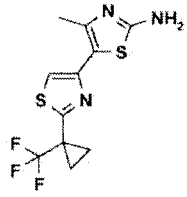
acid amide. LC-MS: t_R =1.78 min, M+H = 462.0, M-H = 460.0 (method B); ^{1}H-NMR (d_6-DMSO, 600.13 MHz) 10.8 (s, br, 1H) 7.6 (s, 1H), 7.1 (s, br, 1H), 6.9 (s, br, 1H), 5.15 (s, 1H), 4.4 (s, 1H), 4.2 (s, br, 1H), 3.6 (m, 1H); 3.40 (m, 1H), 2.4 (s, 3H), 1.9 (m, 1H), 1.7 (m, 1H), 1.6 (s, 1H), 1.52 (s, 2H).

Step 10.1: Imidazole-1-carboxylic acid [4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide



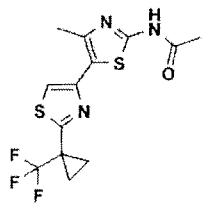
CDI (0.13 g) was added to a stirred solution of 4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-ylamine (Step 10.2) (0.16 g) in CH_2Cl_2 (5 mL) at rt. The reaction mixture was then stood for 3 h at 25°C and the title compound isolated by filtration.

Step 10.2: 4'-Methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-ylamine



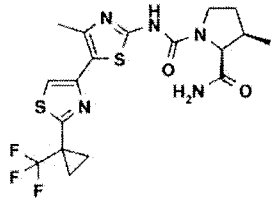
HCl (1.17 g 32% aqueous solution) was added to a solution of N-[4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-acetamide (Step 10.3) (0.18 g) in EtOH (10 mL) and the reaction mixture heated at reflux for 7 h. The cooled reaction mixture was partitioned between EtOAc and aqueous $NaHCO_3$ solution, the organic layers dried over Na_2SO_4 and evaporated to give the title compound. MS (ESI): positive 306.1 (M+H), negative 304.1 (M-H).

Step 10.3: N-[4'-Methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-acetamide



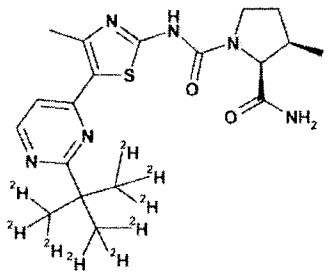
To a solution of *N*-[5-(2-bromo-acetyl)-4-methyl-thiazol-2-yl]-acetamide (0.29 g, prepared as described in WO 2005/068444) in MeOH (15 mL) at rt was added 1-trifluoromethyl-cyclopropanecarbothioic acid amide [871913-36-9] (0.20 g) and ammoniumphosphomolybdate (0.15 g). After stirring 18 h at rt, the reaction mixture was partitioned between EtOAc and water, the organic layers dried over Na_2SO_4 and evaporated to give the crude product. Purification by flash chromatography with an eluent of 1% MeOH in CH_2Cl_2 gave the title compound. MS (ESI): positive 348.1 (M+H), negative 346.1 (M-H).

Example 11: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide}



Imidazole-1-carboxylic acid [4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide (Step 10.1) (20 mg) is added to a stirred solution of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide (Step 1.8) (9 mg) and Et_3N (21 µl) in DMF (0.5 mL) at rt. The reaction mixture was stirred at rt for 56 h, evaporated and crystallized from aqueous MeOH to give the title compound. LC-MS: t_R = 1.90 min, M+H = 460.0, M-H = 458.0 (method B); ^{1}H-NMR (d_6-DMSO, 600.13 MHz) 10.75 (s, br, 1H) 7.62 (s, 1H), 7.35 (s, br, 1H), 6.95 (s, br, 1H), 4.15 (s, br, 1H), 3.65 (m, 1H); 3.35 (m, 1H), 2.4 (s, 3H), 2.35 (m, 1H), 1.9 (m, 1H), 1.7 (m, 1H), 1.52 (s, 1H), 1.50 (s, 2H), 0.95 (d, 3H).

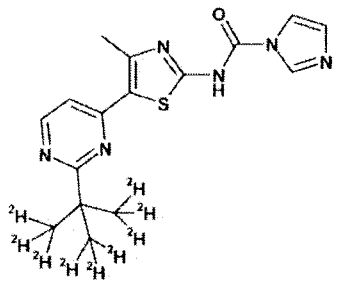
Example 12: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[5-(2-d_9-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide}



Imidazole-1-carboxylic acid [5-(2-d_9-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide (Step 12.1) (222 mg) is added to a stirred solution of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide (Step 1.8) (95 mg) and Et_3N (264 µl) in DMF (3 mL) at rt. The reaction

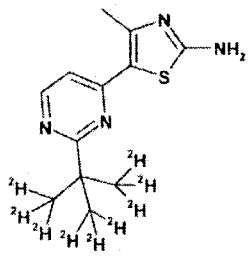
mixture was stirred at rt for 56 h, evaporated and crystallized from aqueous MeOH to give the title compound. MS (ESI): positive 412.1 (M+H), negative 410.2 (M-H).

Step 12.1: Imidazole-1-carboxylic acid [5-(2-d_9-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide



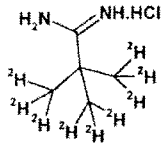
CDI(0.77 g) was added to a stirred solution of 5-(2-d_9-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-ylamine (Step 12.2) (1.11 g) in DMF (4.3 mL) at rt. The reaction mixture was then stood for 18 h at 25°C and the title compound isolated by filtration.

Step 12.2: 5-(2-d_9-tert-Butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-ylamine



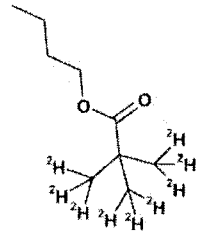
Powdered NaOH (3.71 g) was added to a solution of N'-[5-(3-dimethylamino-acryloyl)-4-methyl-thiazol-2-yl]-N,N-dimethyl-formamidine [507487-90-3] (5.51 g) and d_9-2,2-dimethyl-propionamidine hydrochloride (Step 12.3) (4.50 g) in 2-methoxyethanol (41 mL) and the mixture heated at 125°C for 1 h with stirring. The reaction mixture was cooled, water added, and the crude product isolated by filtration. The crude product was purified by preparative HPLC and the fractions containing the title compound partitioned between CH_2Cl_2 and aqueous $NaHCO_3$. The title compound was obtained as a yellow solid after evaporation of the dried CH_2Cl_2 layers. LC-MS: t_R = 1.12 min, M+H 258.4.

Step 12.3: d_9-2,2-Dimethyl-propionamidine hydrochloride



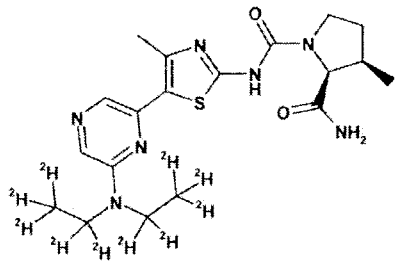
A 2M solution of trimethylaluminium in toluene (61 mL) was added dropwise to a suspension of ammonium chloride (6.53 g) in toluene (46 mL) with ice bath cooling. The reaction mixture was stirred for 4 h at rt and d_9-2,2-dimethyl-propionic acid butyl ester (Step 12.4) (6.3 g) added. After heating at 80°C for 4 days, the reaction mixture was cooled to 0°C and MeOH (200 mL) was carefully added dropwise. After stirring and sonication for 1 h at rt, the reaction mixture was filtered through Hyflo, washing with MeOH, and the filtrate was evaporated to give the title compound as an off-white solid.

Step 12.4: d_9-2,2-Dimethyl-propionic acid butyl ester



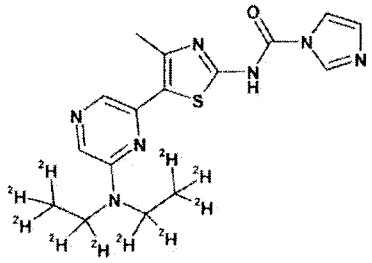
d_9-tert-Butylchloride (5.0 g) was added portionwise to a suspension of magnesium (1.50 g) in THF (20 mL), activated with a catalytic amount of iodine, over 1 h with heating as required to maintain a steady reflux. The reaction mixture was then heated for a further 1 h to ensure complete Grignard formation. The above Grignard solution was then added dropwise to a solution of imidazole-1-carboxylic acid butyl ester (7.5 g, prepared as described by T. Werner and A.G.M. Barrett, J. Org. Chem. 2006, 71, 4302-4304.) in THF (40 mL) cooled with an ice bath. The reaction mixture was stirred for 18 h at rt, (200 mL) was added, the mixture filtered through Hyflo, the filtrate extracted with Et_2O and the Et_2O layers dried over Na_2SO_4 and evaporated to give the tile compound.

Example 13: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[5-(6- d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-yl]-amide}



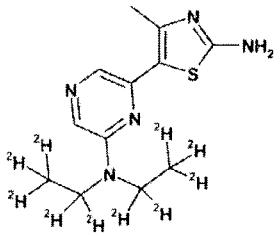
Imidazole-1-carboxylic acid [5-(6-d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-yl]-amide (Step 13.1) (19 mg) was added to a stirred solution of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide (Step 1.8) (9 mg) and Et3N (21 µl) in DMF (0.5 mL) at rt. The reaction mixture was stirred at rt for 56 h, water (1 mL) was added and the title compound collected by filtration. MS (ESI): positive 428.1 (M+H), negative 426.2 (M-H).

Step 13.1: Imidazole-1-carboxylic acid [5-(6-d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-yl]-amide



CDI (78 mg) was added to a solution of 5-(6-d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-ylamine (Step 13.2) (121 mg) in DMF (2 mL) at rt and stood for 3.5 h at rt. The reaction mixture was filtered, washing with CH_2Cl_2, to give the title compound.

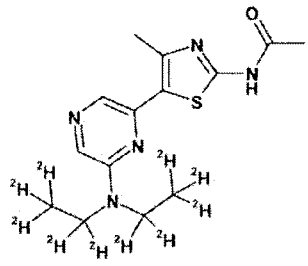
Step 13.2: 5-(6-d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-ylamine



Concentrated HCl (0.4 mL) was added to N-[5-(6-d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-yl]-acetamide (Step 13.3) (140 mg) in EtOH (9 ml) at rt and the mixture heated at reflux for 40 h. The cooled reaction mixture was evaporated, neutralized with aqueous $NaHCO_3$ and extracted with 10% MeOH in CH_2Cl_2. The combined organic extracts were

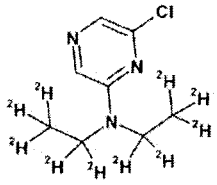
dried over Na_2SO_4 and evaporated to give the title compound. LC-MS: t_R = 1.17 min, M+H 274.4 (method A).

Step 13.3: N-[5-(6-d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-yl]-acetamide



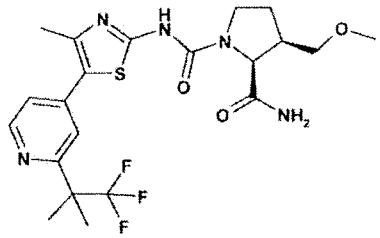
Argon was bubbled through a mixture of 2-d_{10}-diethylamino-6-chloropyrazine (Step 13.4) (293 mg), 2-acetamido-4-methylthiazole (300 mg), palladium acetate (24 mg), tri-tert-butylphosphonium tetrafuoroborate (61 mg) and cesium carbonate (1.02 g) in DMF (3 mL) at rt for 5 min. The reaction mixture was heated in a sealed vial under an argon atmosphere for 45 min at 150°C in a Biotage Initiator™ microwave apparatus, filtered and purified by preparative HPLC. Fractions containing the title compound were combined and evaporated to remove acetonitrile and the title compound obtained as a beige solid by filtration. LC-MS: t_R = 1.68 min, M+H 316.3 (method A).

Step 13.4: 2-d_{10}-diethylamino-6-chloropyrazine



d_{10}-Diethylamine (0.5 g) was added to a stirred mixture of 2,6-dichloropyrazine [4774-14-5] (0.93 g) and potassium carbonate (1.41 g) in acetonitrile (4 mL) at rt. The reaction mixture was then heated at 55°C for 60 h, cooled, water added and extracted with CH_2Cl_2. The combined organic extracts were dried over Na_2SO_4, evaporated and purified by normal phase chromatography, eluent CH_2Cl_2, to give the title compound. LC-MS: t_R = 2.10 min, M+H 196.4 and 198.4 (method A).

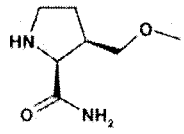
Example 14: (2S,3R)-3-Methoxymethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 2 but (2S,3R)-3-methoxymethyl-pyrrolidine-2-carboxylic acid amide (Step 14.1) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

The title compound was obtained as a white solid. HPLC: t_R = 3.18 min (method A); LC-MS: t_R= 1.88 min, $[M+H]^+$ 486 (method A); TLC: R_f = 0.14 (19:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.97 (br s, 1H), 8.60 (d, 1H), 7.55 (br s, 1H), 7.41 (d, 1H), 7.39 (br s, 1H), 7.04 (br s, 1H), 4.26 (br s, 1H), 3.73 (m, 1H), 3.45 (m, 1H), 3.40 (m, 1H), 3.24 (s, 3H), 3.16 (m, 1H), 2.50 (m, 1H), 2.42 (s, 3H), 2.02 (m, 1H), 1.77 (m, 1H), 1.61 (s, 6H).

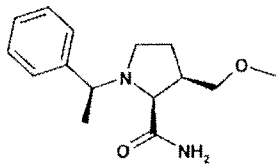
Step 14.1: (2S,3R)-3-Methoxymethyl-pyrrolidine-2-carboxylic acid amide



The title compound was prepared in analogy to the procedure described in Step 1.8 but (2S,3R)-3-methoxymethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 14.2) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide. Moreover, 10% Pd on charcoal, wet with 50% H2O (Aldrich 330108) was used instead of the dry catalyst.

The title compound was obtained as a white solid. ESI-MS: $[M+H]^+$ 159.

Step 14.2: (2S,3R)-3-Methoxymethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide

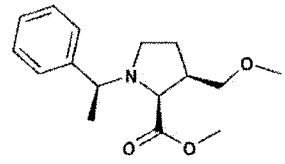


The title compound was prepared in analogy to the procedure described in Step 1.9 but (2S,3R)-3-methoxymethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester (Step 14.3) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-

carboxylic acid methyl ester. Moreover, the basified aqueous layer was extracted exhaustively with THF instead of CH_2Cl_2.

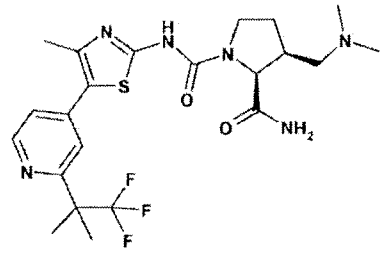
The title compound was obtained as a yellow oil. HPLC: t_R = 2.35 min (method A); LC-MS: t_R= 0.47 min, $[M+H]^+$ 263 (method C); TLC: R_f = 0.05 (1:1 Heptanes/EtOAc).

Step 14.3: (2S,3R)-3-Methoxymethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid methyl ester



A mixture of (3aR,6aS)-1-((S)-1-phenyl-ethyl)-hexahydro-furo[3,4-b]pyrrol-6-one [805246-48-4] (17.05 mmol), KOH (71.60 mmol) and iodomethane (68.20 mmol) in toluene (79 mL) was stirred at 80°C for 1.5 h. The reaction mixture was cooled to rt and partitioned between water and MTBE. The aqueous layer was extracted with MTBE (3X). The combined organic layers were dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel column chromatography to afford the title compound as a yellow oil. HPLC: t_R = 2.98 min (method A); LC-MS: t_R= 0.69 min, $[M+H]^+$ 278 (method C); TLC: R_f = 0.25 (1:3 Heptanes/EtOAc).

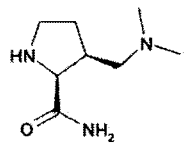
Example 15: (2S,3S)-3-Dimethylaminomethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide)



The title compound was prepared in analogy to the procedure described in Example 2 but (2S,3S)-3-dimethylaminomethyl-pyrrolidine-2-carboxylic acid amide (Step 15.1) was used instead of (2S,3R)-3-methyl-pyrrolidine-2-carboxylic acid amide.

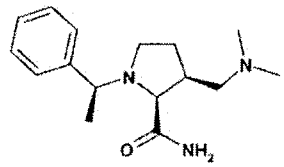
The title compound was obtained as a yellow solid. HPLC: t_R = 4.71 min (method B); LC-MS: t_R= 1.58 min, $[M+H]^+$ 499 (method A); TLC: R_f = 0.08 (4:1 CH_2Cl_2/MeOH); ^{1}H-NMR (d_6-DMSO, 600 MHz): 10.99 (br s, 1H), 8.60 (d, 1H), 7.55 (br s, 1H), 7.41 (d, 1H), 7.40 (br s, 1H), 7.04 (br s, 1H), 4.25 (br s, 1H), 3.69 (m, 1H), 3.41 (m, 1H), 2.51 (m, 1H), 2.41 (s, 3H), 2.32 (m, 1H), 2.17 (m, 1H), 2.17 (s, 6H), 2.01 (m, 1H), 1.72 (m, 1H), 1.61 (s, 6H).

Step 15.1: (2S,3S)-3-Dimethylaminomethyl-pyrrolidine-2-carboxylic acid amide



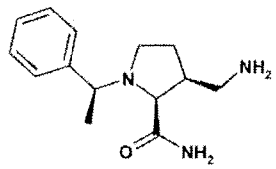
The title compound was prepared in analogy to the procedure described in Step 1.8 but (2S,3S)-3-dimethylaminomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 15.2) was used instead of (2S,3R)-3-methyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide. Moreover, the hydrogenation was performed under 4 bar pressure. The title compound was obtained as a yellow oil. ESI-MS: $[M+H]^+$ 172.

Step 15.2: (2S,3S)-3-Dimethylaminomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide



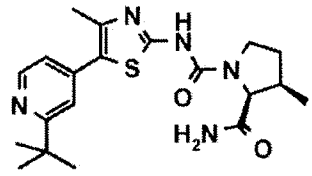
A mixture of (2S,3S)-3-aminomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 15.3) (0.418 mmol), sodium cyanoborohydride (2.86 mmol) and 37% aqueous formaldehyde (2.14 mmol) in MeOH (3.3 mL) was stirred at 55°C for 16 h. The reaction mixture was cooled to rt and concentrated. The residue was purified using a RediSep® silica gel column to afford the title compound as a white foam. HPLC: t_R 3.59 min (method B); LC-MS: t_R= 0.86 min, $[M+H]^+$ 276 (method A); TLC: R_f = 0.13 (9:1 CH_2Cl_2/MeOH).

Step 15.3: (2S,3S)-3-Aminomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide



A mixture of (2S,3S)-3-azidomethyl-1-((S)-1-phenyl-ethyl)-pyrrolidine-2-carboxylic acid amide (Step 5.3) (0.723 mmol) and triphenylphosphine (0.867 mmol) in THF (3 mL) was stirred at rt for 25 h. The reaction mixture was concentrated to afford the crude title compound as a light brown solid. HPLC: t_R 3.53 min (method B); ESI-MS: $[M+H]^+$ 248.

Example 16: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[5-(2-tert-butyl-pyridin-4-yl)-4-methyl-thiazol-2-yl]-amide}



The title compound was prepared from commercially available (Combi-Phos) 4-bromo-2-tert-butyl-pyridine (instead of 4-bromo-2-(1-methyl-cyclopropyl)-pyridine) using synthetic methodology as described for the preparation of **Example 1**.

LC-MS: t_R = 0.45 min, M+H = 402.3, M-H = 400.2 (method D). ^{1}H-NMR (d_6-DMSO, 400 MHz): 8.495 (d, 1H), 7.34 (s, 2H), 7.205 (d, 1H), 6.95 (bs, 1H), 4.15 (m, 1H), 3.68 (dd, 1H), 3.37 (m, 1H), 2.39 (s, 3H), 2.37 (m, 1H), 1.95 (m, 1H), 1.68 (m, 1H), 1.32 (s, 9H), 0.965 (d, 3H).

Efficiency as PI3 kinase inhibitors

PI3K KinaseGlo assay: 50 nL of compound dilutions were dispensed onto black 384-well low volume Non Binding Styrene (NBS) plates (Costar Cat. No. NBS#3676). L-a-phosphatidylinositol (PI), provided as 10 mg/ml solution in methanol, was transferred into a glass tube and dried under nitrogen beam. It was then resuspended in 3% OctylGlucoside (OG) by vortexing and stored at 4°C. The KinaseGlo Luminescent Kinase Assay (Promega, Madison/WI, USA) is a homogeneous HTS method of measuring kinase activity by quantifying the amount of ATP remaining in solution following a kinase reaction.

5 µL of a mix of PI/OG with the PI3K subtype were added (Table 1). Kinase reactions were started by addition of 5 µl of ATP-mix containing in a final volume 10 µL 10 mM TRIS-HCl pH 7.5, 3mM $MgCl_2$, 50 mM NaCl, 0.05% CHAPS, 1mM DTT and 1 µM ATP, and occurred at room temperature. Reactions were stopped with 10 µl of KinaseGlo and plates were read 10 mins later in a Synergy2 reader using an integration time of 0.1 seconds per well. 2.5 µM of a pan-class 1 PI3 kinase inhibitor (standard) was added to the assay plates to generate the 100% inhibition of the kinase reaction, and the 0% inhibition was given by the solvent vehicle (90% DMSO in water). The standard was used as a reference compound and included in all assay plates in the form of 16 dilution points in duplicate.

Table 1 PI3Ks by KinaseGlo: assay conditions and reagent protocol

Vol (10 µL)	Enzyme (nM)	ATP (µM)	PI/OG (µM/ µg/ml)	NaCl (mM)	Mg^{2+} (mM)	CHAPS (%)	DTT (mM)	time (mins)
PI3Kα	10	1	11/10	50	3	0.05	1	30
PI3Kβ	25	1	11/10	50	3	0.05	1	30
PI3Kγ	150	1	22/20	50	3	0.05	1	90
PI3Kd	10	1	11/10	50	3	0.05	1	30

Cloning of PI3Ks

The PI3Kα, PI3Kβ and PI3Kδ constructs are fusion of p85α iSH2 domain and the respective p110 isoforms. The p85α fragment and p110 isoform genes were generated by PCR from first strand cDNA generated by RT-PCR from commercial RNA from placenta, testis and brain as described below. The PI3Kγ construct was obtained from Roger Williams lab, MRC Laboratory of Molecular Biology, Cambridge, UK (November, 2003) and is described (Pacold, Michael E.; Suire, Sabine; Perisic, Olga; Lara-Gonzalez, Samuel; Davis, Colin T.; Walker, Edward H.; Hawkins, Phillip T.; Stephens, Len; Eccleston, John F.; Williams, Roger L. Crystal structure and functional analysis of Ras binding to its effector phosphoinositide 3-kinase gamma. Cell (2000), 103(6), 931-943).

PI3Kα constructs and proteins

PI3Kα wt	BV1075	p85iSH2(461-568)-GGGGGGGGGGGG-p110α(21-1068)-His

BV1075: The construct for Baculovirus BV-1075 was generated by a three-part ligation comprised of a p85 fragment and a p110α fragment cloned into vector pBlueBac4.5. The p85 fragment was derived from plasmid p1661-2 digested with Nhe/Spe. The p110α fragment derived from is clone was verified by sequencing and used in a LR410 as a SpeI/HindIII fragment. For the generation of the baculovirus expression vector LR410 the gateway LR reaction to transfer the insert into the Gateway adapted pBlueBac4.5 (Invitrogen) vector was used,. The cloning vector pBlueBac4.5 (Invitrogen) was digested with Nhe/HindIII. This resulted in the

construct PED 153.8. The p85 component (iSH2) was generated by PCR using ORF 318 (described above) as a template and one forward primer KAC1028 (5'-GCTAGCATGCGAGAATATGATAGAT-TATATGAAG-AATATACC) (SEQ ID No. 1) and two reverse primers, KAC1029 (5'-GCCTCCACCAC-CTCCGCCTG-GTTTAATGCTGTTCATACGTTTGTC) (SEQ ID No. 2) and KAC1039 (5'-TACTAGTC-CGCCTCCAC-CACCTCCGCCTCCACCACCTCCGCC) (SEQ ID No. 3). The two reverse primers overlap and incorporate the 12x Gly linker and the N-terminal sequence of the p110α gene to the SpeI site. The 12x Gly linker replaces the single Gly linker in the BV1052 construct. The PCR fragment was cloned into pCR2.1 TOPO (Invitrogen). Of the resulting clones, p1661-2 was determined to be correct by sequencing. This plasmid was digested with Nhe and SpeI and the resulting fragment was gel-isolated and purified for sub-cloning.

The p110α cloning fragment was generated by enzymatic digest of clone LR410 (see above) with Spe I and HindIII. The SpeI site is in the coding region of the p110α gene. The resulting fragment was gel-isolated and purified for sub-cloning. The cloning vector, pBlueBac4.5 (Invitrogen) was prepared by enzymatic digestion with Nhe and HindIII. The cut vector was purified with Qiagen column and then dephosphorylated with Calf Intestine alkaline phosphatase (CIP) (BioLabs). After completion of the CIP reaction the cut vector was again column purified to generate the final vector. A three-part ligation was performed using Roche Rapid ligase and the vendor specifications. The final plasmid was verified by sequencing.

Kinase domain.

Protein sequence of BV 1075:

```
   1 MREYDRLYEE YTRTSQEIQM KRTAIEAFNE TIKIFEEQCQ TQERYSKEYI EKFKREGNEK
  61 EIQRIMHNYD KLKSRISEII DSRRRLEEDL KKQAAEYREI DKRMNSIKPG GGGGGGGGGG
 121 GLVECLLPNG MIVTLECLRE ATLITIKHEL FKEARKYPLH QLLQDESSYI FVSVTQEAER
 181 EEFFDETRRL CDLRLFQPFL KVIEPVGNRE EKILNREIGF AIGMPVCEFD MVKDPEVQDF
 241 RRNILNVCKE AVDLRDLNSP HSRAMYVYPP NVESSPELPK HIYNKLDKGQ IIVVIWVIVS
 301 PNNDKQKYTL KINHDCVPEQ VIAEAIRKKT RSMLLSSEQL KLCVLEYQGK YILKVCGCDE
 361 YFLEKYPLSQ YKYIRSCIML GRMPNLMLMA KESLYSQLPM DCFTMPSYSR RISTATPYMN
 421 GETSTKSLWV INSALRIKIL CATYVNVNIR DIDKIYVRTG IYHGGEPLCD NVNTQRVPCS
 481 NPRWNEWLNY DIYIPDLPRA ARLCLSICSV KGRKGAKEEH CPLAWGNINL FDYTDTLVSG
 541 KMALNLWPVP HGLEDLLNPI GVTGSNPNKE TPCLELEFDW FSSVVKFPDM SVIEEHANWS
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601 VSREAGFSYS HAGLSNRLAR DNELRENDKE QLKAISTRDP LSEITEQEKD FLWSHRHYCV
661 TIPEILPKLL LSVKWNSRDE VAQMYCLVKD WPPIKPEQAM ELLDCNYPDP MVRGFAVRCL
721 EKYLTDDKLS QYLIQLVQVL KYEQYLDNLL VRFLLKKALT NQRIGHFFFW HLKSEMHNKT
781 VSQRFGLLLE SYCRACGMYL KHLNRQVEAM EKLINLTDIL KQEKKDETQK VQMKFLVEQM
841 RRPDFMDALQ GFLSPLNPAH QLGNLRLEEC RIMSSAKRPL WLNWENPDIM SELLFQNNEI
901 IFKNGDDLRQ DMLTLQIIRI MENIWQNQGL DLRMLPYGCL SIGDCVGLIE VVRNSHTIMQ
961 IQCKGGLKGA LQFNSHTLHQ WLKDKNKGEI YDAAIDLFTR SCAGYCVATF ILGIGDRHNS
1021 NIMVKDDGQL FHIDFGHFLD HKKKKFGYKR ERVPFVLTQD FLIVISKGAQ ECTKTREFER
1081 FQEMCYKAYL AIRQHANLFI NLFSMMLGSG MPELQSFDDI AYIRKTLALD KTEQEALEYF
1141 MKQMNDAHHG GWTTKMDWIF HTIKQHALNE LGGAHHHHHH (SEQ ID No. 4)

PI3Kβ constructs and proteins

PI3Kβ	BV949	p85iSH2(461-N58K-568)-GGGGGG-p110β(2-1070)-His

BV949: PCR products for the inter SH2 domain (iSH2) of the p85 PI3Kα, PI3Kβ and PI3Kδ subunit and for the full-length p110β subunit were generated and fused by overlapping PCR. The iSH2 PCR product was obtained from first strand cDNA generated by RT-PCR from commercial human RNA from placenta, testis and brain (Clontech), initially using primers gwG130-p01 (5'-CGAGAATATGATAGATTATATGAAGAAT-3') (SEQ ID No. 5) and gwG130-p02 (5'-TGGTTT-AATGCTGTTCATACGTTTGTCAAT-3') (SEQ ID No. 6). Subsequently, in a secondary PCR reaction Gateway recombination AttB1 sites and linker sequences were added at the 5'end and 3'end of the p85 iSH2 fragment respectively, using primers gwG130-p03 (5'-GGGACAAGTT-TGTACAAAAAAGCAGGCTACGAAGGAGATATACATATGCGAGAATATGATAGAT TATATGAAGAAT-3') (SEQ ID No. 7) and gwG130-p05 (5'-ACTGAAGCATCCTCCTC-CTCCTCCT-CCTGGTTTAATGCTGTTCATACGTTTGTC-3') (SEQ ID No. 8). The p110β fragment was obtained by PCR using as template a p110β clone (from unknown source that was sequence verified) using primers gwG130-p04 (5'-ATTAAACCAGGAGGAGGAGGAGGAGGATGCTT-CAGTTTCATAATGCCTCCTGCT -3') (SEQ ID No. 9) which contains linker sequences and the 5'end of p110β and gwG130-p06 (5'-AGCTCCGTGATGGTGATGGTGATGTGCTCCAGATC-TGTAGTCTTTCCGAA-

CTGTGTG-3') (SEQ ID No. 10) which contains sequences of the 3'end of p110-β fused to a Histidine tag. The p85-iSH2/ p110β fusion protein was assembled by an overlapping PCR a reaction of the linkers at the 3'end of the iSH2 fragment and the 5'end of the p110β fragment, using the above mentioned gwG130-p03 primer and a primer containing an overlapping Histidine tag and the AttB2 recombination sequences (5'-GGGACCACTTTGTACAAGAAAGCTGGGTTTAAGCTCCGTGATGGTGATGGTGA TGTGCTCC-3') (SEQ ID No. 11). This final product was recombined in a Gateway (Invitrogen) OR reaction into the donor vector pDONR201 (Invitrogen) to generate the ORF253 entry clone. This clone was verified by sequencing and used in a Gateway LR reaction (Invitrogen) to transfer the insert into the Gateway adapted pBlueBac4.5 (Invitrogen) vector for generation of the baculovirus expression vector LR280. This LR280 has an amino acid mutation in the p85 sequence.

Kinase domain.

Protein sequence of BV949:

```
   1 MREYDRLYEE YTRTSQEIQM KRTAIEAFNE TIKIFEEQCQ TQERYSKEYI EKFKREGKEK
  61 EIQRIMHNYD KLKSRISEII DSRRRLEEDL KKQAAEYREI DKRMNSIKPG GGGGGCFSFI
 121 MPPAMADILD IWAVDSQIAS DGSIPVDFLL PTGIYIQLEV PREATISYIK QMLWKQVHNY
 181 PMFNLLMDID SYMFACVNQT AVYEELEDET RRLCDVRPFL PVLKLVTRSC DPGEKLDSKI
 241 GVLIGKGLHE FDSLKDPEVN EFRRKMRKFS EEKILSLVGL SWMDWLKQTY PPEHEPSIPE
 301 NLEDKLYGGK LIVAVHFENC QDVFSFQVSP NMNPIKVNEL AIQKRLTIHG KEDEVSPYDY
 361 VLQVSGRVEY VFGDHPLIQF QYIRNCVMNR ALPHFILVEC CKIKKMYEQE MIAIEAAINR
 421 NSSNLPLPLP PKKTRIISHV WENNNPFQIV LVKGNKLNTE ETVKVHVRAG LFHGTELLCK
 481 TIVSSEVSGK NDHIWNEPLE FDINICDLPR MARLCFAVYA VLDKVKTKKS TKTINPSKYQ
 541 TIRKAGKVHY PVAWVNTMVF DFKGQLRTGD IILHSWSSFP DELEEMLNPM GTVQTNPYTE
 601 NATALHVKFP ENKKQPYYYP PFDKIIEKAA EIASSDSANV SSRGGKKFLP VLKEILDRDP
 661 LSQLCENEMD LIWTLRQDCR EIFPQSLPKL LLSIKWNKLE DVAQLQALLQ IWPKLPPREA
 721 LELLDFNYPD QYVREYAVGC LRQMSDEELS QYLLQLVQVL KYEPFLDCAL SRFLLERALG
 781 NRRIGQFLFW HLRSEVHIPA VSVQFGVILE AYCRGSVGHM KVLSKQVEAL NKLKTLNSLI
 841 KLNAVKLNRA KGKEAMHTCL KQSAYREALS DLQSPLNPCV ILSELYVEKC KYMDSKMKPL
 901 WLVYNNKVFG EDSVGVIFKN GDDLRQDMLT LQMLRLMDLL WKEAGLDLRM LPYGCLATGD
 961 RSGLIEVVST SETIADIQLN SSNVAAAAAF NKDALLNWLK EYNSGDDLDR AIEEFTLSCA
1021 GYCVASYVLG IGDRHSDNIM VKKTGQLFHI DFGHILGNFK SKFGIKRERV PFILTYDFIH
1081 VIQQGKTGNT EKFGRFRQCC EDAYLILRRH GNLFITLFAL MLTAGLPELT SVKDIQYLKD
1141 SLALGKSEEE ALKQFKQKFD EALRESWTTK VNWMAHTVRK DYRSGAHHHH HHGA
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(SEQ ID No. 12)

Kinase domain.

PI3Kγ construct and protein

PI3Kγ	BV950	p110γ(Δ143-[Met144-1102])-His

Construct obtained from Roger Williams lab, MRC Laboratory of Molecular Biology, Cambridge, UK (November, 2003). Description of the construct in (Pacold, Michael E.; Suire, Sabine; Perisic, Olga; Lara-Gonzalez, Samuel; Davis, Colin T.; Walker, Edward H.; Hawkins, Phillip T.; Stephens, Len; Eccleston, John F.; Williams, Roger L. Crystal structure and functional analysis of Ras binding to its effector phosphoinositide 3-kinase gamma. Cell (2000), 103(6), 931-943). Constructs lacking the N-terminal 144 aa.

Protein sequence of BV950:

```
   1 MSEESQAFQR QLTALIGYDV TDVSNVHDDE LEFTRRGLVT PRMAEVASRD PKLYAMHPWV
  61 TSKPLPEYLW KKIANNCIFI VIHRSTTSQT IKVSPDDTPG AILQSFFTKM AKKKSLMDIP
 121 ESQSEQDFVL RVCGRDEYLV GETPIKNFQW VRHCLKNGEE IHVVLDTPPD PALDEVRKEE
 181 WPLVDDCTGV TGYHEQLTIH GKDHESVFTV SLWDCDRKFR VKIRGIDIPV LPRNTDLTVF
 241 VEANIQHGQQ VLCQRRTSPK PFTEEVLWNV WLEFSIKIKD LPKGALLNLQ IYCGKAPALS
 301 SKASAESPSS ESKGKVRLLY YVNLLLIDHR FLLRRGEYVL HMWQISGKGE DQGSFNADKL
 361 TSATNPDKEN SMSISILLDN YCHPIALPKH QPTPDPEGDR VRAEMPNQLR KQLEAIIATD
 421 PLNPLTAEDK ELLWHFRYES LKHPKAYPKL FSSVKWGQQE IVAKTYQLLA RREVWDQSAL
 481 DVGLTMQLLD CNFSDENVRA IAVQKLESLE DDDVLHYLLQ LVQAVKFEPY HDSALARFLL
 541 KRGLRNKRIG HFLFWFLRSE IAQSRHYQQR FAVILEAYLR GCGTAMLHDF TQQVQVIEML
 601 QKVTLDIKSL SAEKYDVSSQ VISQLKQKLE NLQNSQLPES FRVPYDPGLK AGALAIEKCK
 661 VMASKKKPLW LEFKCADPTA LSNETIGIIF KHGDDLRQDM LILQILRIME SIWETESLDL
 721 CLLPYGCIST GDKIGMIEIV KDATTIAKIQ QSTVGNTGAF KDEVLNHWLK EKSPTEEKFQ
 781 AAVERFVYSC AGYCVATFVL GIGDRHNDNI MITETGNLFH IDFGHILGNY KSFLGINKER
 841 VPFVLTPDFL FVMGTSGKKT SPHFQKFQDI CVKAYLALRH HTNLLIILFS MMLMTGMPQL
 901 TSKEDIEYIR DALTVGKNEE DAKKYFLDQI EVCRDKGWTV QFNWFLHLVL GIKQGEKHSA
 961 HHHHHH
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(SEQ ID No. 13)

PI3Kδ construct and protein

PI3Kδ	BV1060	p85iSH2(461-568)-GGGGGG-p110δ(2-1044)-His

BV1060: PCR products for the inter SH2 domain (iSH2) of the p85 subunit and for the full-length p110δ subunit were generated and fused by overlapping PCR. The iSH2 PCR product was generated by using as a template the ORF318 (see above)

and the primers gwG130-p03 (5'- GGGACAAG-TTTGTACAAAAAAGCAGGCTACGAAGGAGATATACATATGC-GAGAATATGATAGATTATATGAAGAAT-3') (SEQ ID No. 7) and gwG154-p04 (5'-TCCTCCTCCT-CCTCCTCCTGGTTTAATGCTGTTCATACGTTTGTC-3') (SEQ ID No. 14). The p110δ fragment was obtained from first strand cDNA generated by RT-PCR from commercial human RNA from placenta, testis and brain (Clontech), using initially primers gwG154-p01 (5'- ATGCCCCCTGGGGTGGACTGCCCCAT-3') (SEQ ID No. 15) and gwG154-p02 (5'-CTACTGCCTGT-TGTCTTTGGACACGT-3') (SEQ ID No. 16). In a subsequent PCR reaction linker sequences and a Histidine tag was added at the 5'end and 3'end of the p110δ fragment respectively, using primers gw154-p03 (5'-ATTAAACCAGGAGGAGGAGGAGGAGGACCCCCTGGGGTGGAC-TGCCCCATGGA-3') (SEQ ID No. 17) and gwG154-p06 (5'-AGCTCCGTGATGGTGATGGTGAT-GTGCT-CCCTGCCTGTTGTCTTTGGACACGTTGT-3') (SEQ ID No. 18).The p85-iSH2/p110δ fusion protein was assembled in a third PCR reaction by the overlapping linkers at the 3'end of the iSH2 fragment and the 5'end of the p110δ fragment, using the above mentioned gwG130-p03 primer and a primer containing an overlapping Histidine tag and the Gateway (Invitrogen) AttB2 recombination sequences (5'-GGG-ACCACTTTGTACAAGAAAGCTGGGTTTAA-GCTCCGTGATGGTGATGGTGAGTGCTCC-3') (SEQ ID No. 19). This final product was recombined in a Gateway OR reaction into the donor vector pDONR201 (Invitrogen) to generate the ORF319 entry clone. This clone was verified by sequencing and used in a Gateway LR reaction (Invitrogen) to transfer the insert into the Gateway adapted pBlueBac4.5 (Invitrogen) vector for generation of the baculovirus expression vector LR415.

Protein sequence of BV1060:

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  1 MREYDRLYEE YTRTSQEIQM KRTAIEAFNE TIKIFEEQCQ TQERYSKEYI EKFKREGNEK
 61 EIQRIMHNYD KLKSRISEII DSRRRLEEDL KKQAAEYREI DKRMNSIKPG GGGGGPPGVD
121 CPMEFWTKEE NQSVVVDFLL PTGVYLNFPV SRNANLSTIK QLLWHRAQYE PLFHMLSGPE
181 AYVFTCINQT AEQQELEDEQ RRLCDVQPFL PVLRLVAREG DRVKKLINSQ ISLLIGKGLH
241 EFDSLCDPEV NDFRAKMCQF CEEAAARRQQ LGWEAWLQYS FPLQLEPSAQ TWGPGTLRLP
301 NRALLVNVKF EGSEESFTFQ VSTKDVPLAL MACALRKKAT VFRQPLVEQP EDYTLQVNGR
361 HEYLYGSYPL CQFQYICSCL HSGLTPHLTM VHSSSILAMR DEQSNPAPQV QKPRAKPPPI
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421 PAKKPSSVSL WSLEQPFRIE LIQGSKVNAD ERMKLVVQAG LFHGNEMLCK TVSSSEVSVC
481 SEPVWKQRLE FDINICDLPR MARLCFALYA VIEKAKKARS TKKKSKKADC PIAWANLMLF
541 DYKDQLKTGE RCLYMWPSVP DEKGELLNPT GTVRSNPNTD SAAALLICLP EVAPHPVYYP
601 ALEKILELGR HSECVHVTEE EQLQLREILE RRGSGELYEH EKDLVWKLRH EVQEHFPEAL
661 ARLLLVTKWN KHEDVAQMLY LLCSWPELPV LSALELLDFS FPDCHVGSFA IKSLRKLTDD
721 ELFQYLLQLV QVLKYESYLD CELTKFLLDR ALANRKIGHF LFWHLRSEMH VPSVALRFGL
781 ILEAYCRGST HHMKVLMKQG EALSKLKALN DFVKLSSQKT PKPQTKELMH LCMRQEAYLE
841 ALSHLQSPLD PSTLLAEVCV EQCTFMDSKM KPLWIMYSNE EAGSGGSVGI IFKNGDDLRQ
901 DMLTLQMIQL MDVLWKQEGL DLRMTPYGCL PTGDRTGLIE VVLRSDTIAN IQLNKSNMAA
961 TAAFNKDALL NWLKSKNPGE ALDRAIEEFT LSCAGYCVAT YVLGIGDRHS DNIMIRESGQ
1021 LFHIDFGHFL GNFKTKFGIN RERVPFILTY DFVHVIQQGK TNNSEKFERF RGYCERAYTI
1081 LRRHGLLFLH LFALMRAAGL PELSCSKDIQ YLKDSLALGK TEEEALKHFR VKFNEALRES
1141 WKTKVNWLAH NVSKDNRQEL GGAHHHHHH (SEQ ID No. 20)

Purification of PI3Kα, PI3Kβ and PI3Kγ constructs

PI3Kα, PI3Kβ and PI3Kγ were purified in two chromatographic steps: immobilized metal affinity chromatography (IMAC) on a Ni sepharose resin (GE Healthcare) and gel filtration utilizing a Superdex 200 26/60 column (GE Healthcare). All buffers were chilled to 4°C and lysis was performed chilled on ice. Column fractionation was performed at room temperature. All buffers used to purify PI3Kβ contained 0.05% Triton X100 in addition to what is described below.

Typically frozen cells from 10 L of Tn5 cell culture were resuspended in "Lysis Buffer" 20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 5% glycerol, 5 mM imidazole, 1 mM NaF, 0.1ug/mL okadaic acid (OAA), 5 mM BME, 1 x Complete protease inhibitor cocktail – EDTA-free (20 tablets/1 L buffer, Roche Applied Sciences), benzonase (25U/mL buffer, EMD Biosciences) at a ratio of 1:6 v/v pellet to Lysis Buffer ratio, and mechanically lysed by douncing 20 strokes using a tight-fitting pestle. The lysate was centrifuged at 45,000 g for 30 minutes, and the supernatant was loaded onto a pre-equilibrated IMAC column (3 mL resin/100 mL lysate). The column was washed with 3-5 column volumes of Lysis Buffer, followed by a second wash of 3-5 column volumes with 20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 5% glycerol, 45 mM imidazole, 1 mM NaF, 0.1µg/mL OAA, 5 mM BME, 1x Complete protease inhibitor cocktail – EDTA-free. Protein was eluted with 20 mM Tris-Cl, pH 7.5, 0.5 M NaCl, 5% glycerol, 250 mM imidazole, 1 mM NaF, 0.1µg/mL OAA, 5 mM BME, 1x Complete protease inhibitor cocktail – EDTA-free. Pertinent fractions were analyzed by SDS-PAGE and pooled accordingly. The protein was further purified by gel filtration on a Superdex 200 26/60 column equilibrated in 20 mM Tris-Cl, pH 7.5, 0.5 M NaCl, 5% glycerol, 1 mM NaF, 5 mM

DTT, 1x Complete protease inhibitor cocktail – EDTA-free. Pertinent fractions were analyzed by SDS-PAGE and pooled accordingly. An equal volume of Dialysis Buffer (20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 50% glycerol, 5 mM NaF, 5 mM DTT) was added to the pool and than dialyzed against Dialysis Buffer two changes (one change overnight). Protein was stored at -20°C.

Purification of PI3Kδ

PI3Kδ was purified in three chromatographic steps: immobilized metal affinity chromatography on a Ni Sepharose resin (GE Healthcare), gel filtration utilizing a Superdex 200 26/60 column (GE Healthcare), and finally a ion exchange step on a Q-HP column (GE Healthcare). All buffers were chilled to 4°C and lysis was performed chilled on ice. Column fractionation was performed at room temperature.

Typically frozen cells from 10 L of Tn5 cell culture were resuspended in "Lysis Buffer" 20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 5% glycerol, 5 mM imidazole, 1 mM NaF, 0.1µg/mL okadaic acid (OAA), 5 mM BME, 1 x Complete protease inhibitor cocktail – EDTA-free (20 tablets/1 L buffer, Roche Applied Sciences), benzonase (25U/mL lysis buffer, EMD Biosciences) at a ratio of 1:10 v/v pellet to Lysis Buffer ratio, and mechanically lysed by douncing 20 strokes using a tight-fitting pestle. The lysate was centrifuged at 45,000 g for 30 minutes, and the supernatant was loaded onto a pre-equilibrated IMAC column (5 mL resin/100 mL lysate). The column was washed with 3-5 column volumes of Lysis Buffer, followed by a second wash of 3-5 column volumes with 20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 5% glycerol, 40 mM imidazole, 1 mM NaF, 0.1ug/mL OAA, 5 mM BME, 1 x Complete protease inhibitor cocktail – EDTA-free. Protein was eluted with 20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 5% glycerol, 250 mM imidazole, 1 mM NaF, 0.1µg/mL OAA, 5 mM BME, 1 x Complete protease inhibitor cocktail – EDTA-free. Pertinent fractions were analyzed by SDS-PAGE and pooled accordingly. The protein was further purified by gel filtration on a Superdex 200 equilibrated in 20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 5% glycerol, 1 mM NaF, 0.1ug/mL OAA, 5 mM DTT, 1 x Complete protease inhibitor cocktail – EDTA-free. Pertinent fractions were analyzed by SDS-PAGE and pooled accordingly. These fractions were diluted 1:10 v/v pool volume to buffer ratio with "Buffer A" 20 mM Tris-Cl, pH 8.2, 5% glycerol, 1 mM NaF, 0.1µg/mL OAA, 5 mM DTT and loaded onto a prepared Q-HP column. After sample loading is completed we wash with Buffer A and 5% "Buffer B" 20 mM Tris-Cl, pH 8.2, 1 M NaCl, 5% glycerol, 1 mM NaF, 0.1ug/mL OAA, 5 mM DTT for 3-5 column volumes. We elute the protein using a 5%-30% gradient of Buffer B. Typically the protein elutes at ~200 mM NaCl.

Pertinent fractions were analyzed by SDS-PAGE and pooled accordingly. An equal volume of Dialysis Buffer (20 mM Tris-Cl, pH 7.5, 500 mM NaCl, 50% glycerol, 1 mM NaF, 0.1µg/mL OAA, 5 mM DTT) was added to the pool and then dialyzed against Dialysis Buffer two changes (one change overnight). Protein was stored at -20°C.

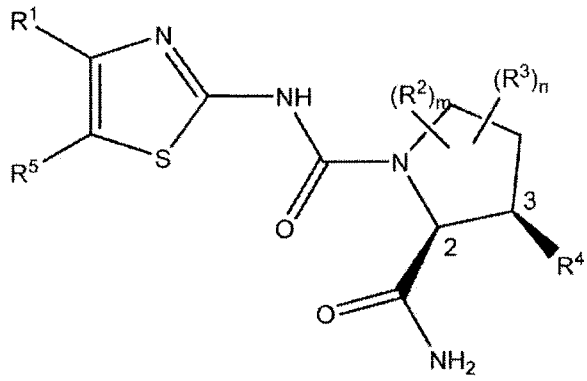
The following results were obtained using the above described assays. The selectivity factors for the PI3K beta, gamma and delta isoforms were calculated by dividing the respective IC_{50} value by the PI3Kalpha IC_{50} value.

Example no.	PI3Kalpha IC_{50} [umol l-1]	PI3Kbeta IC_{50} [umol l-1] (selectivity)	PI3Kgamma IC_{50} [umol l-1] (selectivity)	PI3Kdelta IC_{50} [umol l-1] (selectivity)
1	0.007	2.596 (370)	0.175 (25)	0.168 (24)
2	0.027	1.204 (44)	0.104 (3)	0.059 (2)
3	0.053	>9.1 (>171)	1.078 (20)	0.8 (15)
4	0.091	6.956 (76)	1.68 (18)	0.437 (4)
5	0.018	3.218 (178)	n.d.	0.24 (13)
6	0.013	0.99 (76)	n.d.	0.684 (52)
7	0.038	5.154 (136)	n.d.	0.19 (5)
8	0.006	2.285 (380)	n.d.	0.049 (8)
9	0.010	>9.1 (910)	0.2 (20)	0.586 (58)
10	0.043	>9.1 (211)	0.29 (6)	1.09 (25)
11	0.015	4.618 (307)	0.194 (12)	0.225 (15)
12	0.0045	1.589 (353)	0.076 (16)	0.033 (7)
13	0.004	0.938 (234)	0.418 (104)	0.049 (12)
14	0.012	1.465 (122)	n.d.	0.110 (9)
15	0.017	3.265 (192)	n.d.	0.093 (5)
16	0.009	2.169 (241)	n.d.	0.113 (13)

n.d. = not done.

CLAIMS

1. A compound of formula I



(I)

wherein,

m is 0 or 1;

n is 0 or 1;

R^1 represents H, halogen, unsubstituted C_1-C_4-alkyl or substituted C_1-C_4-alkyl;

R^2 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, unsubstituted or substituted amino, halogen or hydroxy;

R^3 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, unsubstituted or substituted amino, halogen or hydroxy;

R^4 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, halogen or hydroxy; or

R^3 and R^4 form, together with the same or different carbon atom to which they are attached, C_3-C_8-cycloalkyl or heterocyclyl;

R^5 is unsubstituted or substituted heteroaryl;

or a salt thereof; and

excluding the compound (1*R*,2*S*,5*S*)-3-aza-bicyclo[3.1.0]hexane-2,3-dicarboxylic acid 2-amide 3-{[5-(2-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide}.

2. A compound according to Claim 1, wherein

R^2 represents C_1-C_4-alkyl, C_1-C_4-alkoxy, di-C_1-C_4-alkyl-amino, halogen or hydroxy, or a salt thereof.

3. A compound according to to any preceding claim, wherein

R^3 represents C_1-C_4-alkyl, C_1-C_4-alkoxy, di-C_1-C_4-alkyl-amino, halogen or hydroxy, or a salt thereof.

4. A compound according to any preceding claim, wherein

R^4 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, halogen or hydroxy; or

R^3 and R^4 form, together with the same carbon atom to which they are attached, C_3-C_8-cycloalkyl; or

R^3 and R^4 form, together with the same or different carbon atom to which they are attached, heterocyclyl.

5. A compound according to any preceding claim, wherein

R^4 is independently selected from the group consisting of unsubstituted or substituted C_1-C_8-alkyl, unsubstituted or substituted C_1-C_8-alkoxy, halogen or hydroxy; or

R^3 and R^4 form, together with the same carbon atom to which they are attached, C_3-C_8-cycloalkyl or heterocyclyl.

6. A compound according to any preceding claim, wherein,

R^5 represents unsubstituted or substituted heteroaryl,

substituted by one or more moiety independently selected from the group consisting of halogen, hydroxy, cyano, nitro, C_1-C_7-alkyl, per-deutero C_1-C_7-alkyl, C_3-C_{12}-cycloalkyl, (C_1-C_7-alkyl)-C_3-C_{12}-cycloalkyl, (halo-C_1-C_7-alkyl)-C_3-C_{12}-cycloalkyl, amino-C_1-C_7-alkyl, halo-C_1-C_7-alkyl, N-C_1-C_7-alkanoylamino-C_1-C_7-alkyl, N-C_1-C_7-alkanesulfonyl-amino-C_1-C_7-alkyl, pyrrolidino-C_1-C_7-alkyl, oxo- pyrrolidino-C_1-C_7-alkyl, C_1-C_7-alkanesulfinyl, C_1-C_7-alkanesulfonyl, C_1-C_7-alkoxy, amino, N-mono- or N,N-di-(C_1-C_7-alkyl)-amino, N-mono- or N,N-di-(per-deutero C_1-C_7-alkyl)-amino, N-mono- or N,N-di-(C_1-C_7-cycloalkyl)-amino C_1-C_7-alkanoylamino, pyrrolidino, oxo-pyrrolidino, piperidino, piperazin-1-yl, 4-(C_1-C_7-alkyl, C_1-C_7-alkoxy-C_1-C_7-alkyl, halo-C_1-C_7-alkyl or C_3-C_{10}-cycloalkyl)-piperazin-1-yl, 4-(amino-C_1-C_7-alkyl)-piperazin-1-yl, 4-[N-mono- or N,N-di-(C_1-C_7-alkylamino)-C_1-C_7-alkyl]-piperazin-1-yl, morpholino, thiomorpholino, S-oxo- or S,S-dioxothiomorpholino, C_1-C_7-alkanesulfonylamino, carbamoyl, N-mono- or N,N-di-

(C_1-C_7-alkyl, C_1-C_7-alkoxy-C_1-C_7-alkyl, amino-C_1-C_7-alkyl and/or (N'-mono- or N',N'-di-(C_1-C_7-alkyl)-amino-C_1-C_7-alkyl)-carbamoyl, pyrrolidin-1-carbonyl, piperidin-1-carbonyl, piperazin-1-carbonyl, 4-(C_1-C_7-alkyl)piperazin-1-carbonyl, morpholin-1-carbonyl, thiomorpholin-1-carbonyl, S-oxo- or S,S-dioxothiomorpholin-1-carbonyl, sulfo, C_1-C_7-alkanesulfonyl, C_1-C_7-alkanesulfinyl, sulfamoyl, N-mono- or N,N-di-(C_1-C_7-alkyl)-sulfamoyl, morpholinosulfonyl, thiomorpholinosulfonyl, thiazolyl.

7. A compound according to any preceding claim, wherein,
R^5 represents heteroaryl selected from the group consisting of pyridyl, pyrimidinyl, pyrazinyl and thiazolyl, each independently substituted by one substituent selected from the group consisting of C_1-C_4-alkyl, per-deutero C_1-C_4-alkyl, halo-C_1-C_4-alkyl, 1-(C_1-C_4-alkyl)-C_3-C_6-cycloalkyl, (halo-C_1-C_4-alkyl)-C_3-C_6-cycloalkyl, di-C_1-C_4-alkylamino, di-(per-deutero C_1-C_4-alkyl)amino.

8. A compound according to any preceding claim, wherein,
m is 0 or 1 and n is 1.

9. A compound according to claim 1 selected from: (2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(rac)-3,3-Dimethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(rac)-3,3-Dimethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3S)-3-(Acetylamino-methyl)-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3S)-3-(Acetylamino-methyl)-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3S)-3-Morpholin-4-ylmethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(1-methyl-cyclopropyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3S)-3-Morpholin-4-ylmethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({5-[2-(1-fluoro-1-methyl-ethyl)-pyrimidin-4-yl]-4-methyl-thiazol-2-yl}-amide);

(2S,3R)-3-Hydroxy-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide}

(2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[4'-methyl-2-(1-trifluoromethyl-cyclopropyl)-[4,5']bithiazolyl-2'-yl]-amide};

(2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[5-(2-d_9-tert-butyl-pyrimidin-4-yl)-4-methyl-thiazol-2-yl]-amide};

(2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[5-(6- d_{10}-diethylamino-pyrazin-2-yl)-4-methyl-thiazol-2-yl]-amide};

(2S,3R)-3-Methoxymethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3S)-3-Dimethylaminomethyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-({4-methyl-5-[2-(2,2,2-trifluoro-1,1-dimethyl-ethyl)-pyridin-4-yl]-thiazol-2-yl}-amide);

(2S,3R)-3-Methyl-pyrrolidine-1,2-dicarboxylic acid 2-amide 1-{[5-(2-tert-butyl-pyridin-4-yl)-4-methyl-thiazol-2-yl]-amide}.

10. A pharmaceutical composition comprising a compound of formula (I), according to any of Claims 1 to 9, or a pharmaceutically acceptable salt thereof, and optionally a further therapeutic agent, together with a pharmaceutically acceptable carrier.

11. A compound of the formula (I), according to any one of Claims 1 to 9, or pharmaceutically acceptable salt thereof, for use in the treatment of a lipid and/or protein kinase dependent disease.

12. Use of a compound of formula (I), according to any one of Claims 1 to 9, or a pharmaceutically acceptable salt thereof, for the preparation of a pharmaceutical composition for use in the treatment of a lipid and/or protein kinase dependent disease.

13. A method of treatment of a disease that responds to inhibition of a lipid and/or protein kinase, which comprises administering a prophylactically or therapeutically effective amount of a compound of formula (I) according to any one of Claims 1-9, or a

pharmaceutically acceptable salt thereof, to a warm-blooded animal, in need of such treatment.

14. A compound for use according to Claim 11, or the use of a compound according to claim 12, or a method of treatment according to claim 13, wherein the disease is a lipid kinase dependent disease dependent on a Class I PI3K.

15. A compound for use according to Claim 11, or the use of a compound according to claim 12, or a method of treatment according to claim 13, wherein the disease is a lipid kinase dependent disease dependent on a Class I PI3K selected from the group consisting of PI3Kalpha, PI3Kbeta, PI3Kdelta, PI3Kgamma.

16. A compound for use according to Claim 11, or the use of a compound according to claim 12, or a method of treatment according to claim 13, wherein the disease is a proliferative disease; a benign or malignant tumor; a cancer selected from sarcoma; lung; bronchus; prostate; breast (including sporadic breast cancers and sufferers of Cowden disease); pancreas; gastrointestinal cancer; colon; rectum; colon carcinoma; colorectal adenoma; thyroid; liver; intrahepatic bile duct; hepatocellular; adrenal gland; stomach; gastric; glioma; glioblastoma; endometrial; melanoma; kidney; renal pelvis; urinary bladder; uterine corpus; uterine cervix; vagina; ovary; multiple myeloma; esophagus; a leukaemia; acute myelogenous leukemia; chronic myelogenous leukemia; lymphocytic leukemia; myeloid leukemia; brain; a carcinoma of the brain; oral cavity and pharynx; larynx; small intestine; non-Hodgkin lymphoma; melanoma; villous colon adenoma; a neoplasia; a neoplasia of epithelial character; lymphomas; a mammary carcinoma; basal cell carcinoma; squamous cell carcinoma; actinic keratosis; tumor diseases, including solid tumors; a tumor of the neck or head; polycythemia vera; essential thrombocythemia; and myelofibrosis with myeloid metaplasia.

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2010/059352

A. CLASSIFICATION OF SUBJECT MATTER
INV. C07D417/14 A61K31/427 A61K31/4439 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

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, BIOSIS, CHEM ABS Data, EMBASE, PAJ, WPI Data, BEILSTEIN Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2009/163469 A1 (CARAVATTI GIORGIO [CH] ET AL) 25 June 2009 (2009-06-25) Formula I; paragraphs [0007], [0019], [0120] - [0140]; claims 1,11,12,15; examples 9,10,11,13-15,20,21,29-31,35,37,61,86-91,146,147,1	1-16
X	WO 2004/096797 A1 (NOVARTIS AG [CH]; NOVARTIS PHARMA GMBH [AT]; BRUCE IAN [GB]; CUENOUD B) 11 November 2004 (2004-11-11) Formula I; page 1, lines 15-17; claims 1,11,12; example 133 page 5, line 24 - page 6, line 3 page 21, lines 1-13 page 22, line 30 - page 25, line 22	1-16

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[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

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"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	Date of mailing of the international search report
24 September 2010	04/10/2010

Name and mailing address of the ISA/	Authorized officer
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INTERNATIONAL SEARCH REPORT

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C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

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X,P	WO 2009/080705 A2 (NOVARTIS AG [CH]; FAIRHURST ROBIN ALEC [GB]; IMBACH PATRICIA [CH]) 2 July 2009 (2009-07-02) page 2, line 33 - page 3, line 6; claims 1,12; examples 9,11,40,41,44 page 23, line 28 - page 24, line 12	1,4-8, 10-16
A	WO 2007/115930 A1 (BOEHRINGER INGELHEIM INT [DE]; BOEHRINGER INGELHEIM PHARMA [DE]; MAIER) 18 October 2007 (2007-10-18) Formula I; page 2, lines 13-20; examples 186,240,241,245,249,268,271 page 16, line 23 - page 18, line 14; claims 1,8	1-16

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Information on patent family members

International application No
PCT/EP2010/059352

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