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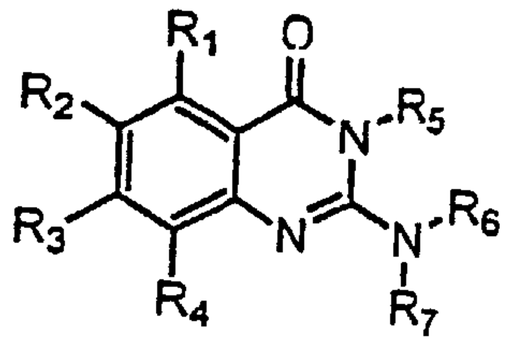
(71) Demandeur/Applicant:
PHENEX PHARMACEUTICALS AG, DE

(72) Inventeurs/Inventors:
DEUSCHLE, ULRICH, DE;
LOEBBERT, RALPH, DE;
BLUME, BEATRIX, DE;
KOEGL, MANFRED, DE;
KREMOSER, CLAUS, DE;
KOBER, INGO, DE;
BAUER, ULRIKE, DE;
HERMANN, KRISTINA, DE

(74) Agent: HEENAN BLAIKIE LLP

(54) Titre : 2-AMINO-4-QUINAZOLINONES EN TANT QUE COMPOSES SE FIXANT SUR LES RECEPTEURS NUCLEAIRES LXR

(54) Title: 2-AMINO-4-QUINAZOLINONES AS LXR NUCLEAR RECEPTOR BINDING COMPOUNDS



(1)

(57) Abrégé/Abstract:
The present invention relates to 2-amino-4-oxo-quinazolines according to the general formula (1), which bind to the LXR receptors and act as agonists and antagonists of the LXR receptors. The invention further relates to the treatment of diseases and/or conditions through binding of said nuclear receptor by said compounds and the production of medicaments using said compounds. In particular the compounds are useful in the treatment of hypercholesterolemia, obesity or other diseases associated with elevated lipoprotein (LDL) levels.

ABSTRACT

The present invention relates to 2-amino-4-oxo-quinazolines according according to the general formula (1), which bind to the LXR receptors and act as agonists and antagonists of the LXR receptors. The invention further relates to the treatment of diseases and/or conditions through binding of said nuclear receptor by said compounds and the production of medicaments using said compounds. In particular the compounds are useful in the treatment of hypercholesterolemia, obesity or other diseases associated with elevated lipoprotein (LDL) levels.

2-AMINO-4-QUINAZOLINONES AS LXR NUCLEAR RECEPTOR BINDING COMPOUNDS

The present invention relates to compounds according to the general formula (1), which bind to the LXR receptors and act as agonists and antagonists of the LXR receptors. The invention further relates to the treatment of diseases and/or conditions through binding of said nuclear receptor by said compounds and the production of medicaments using said compounds.

BACKGROUND OF THE INVENTION

Multicellular organisms are dependent on advanced mechanisms of information transfer between cells and body compartments. The information that is transmitted can be highly complex and can result in the alteration of genetic programs involved in cellular differentiation, proliferation, or reproduction. The signals, or hormones, are often simple molecules, such as peptides, fatty acid, or cholesterol derivatives.

Many of these signals produce their effects by ultimately changing the transcription of specific genes. One well-studied group of proteins that mediate a cells response to a variety of signals is the family of transcription factors known as nuclear receptors, hereinafter referred to often as "NR". Members of this group include receptors for steroid hormones, vitamin D, ecdysone, cis and trans retinoic acid, thyroid hormone, bile acids, cholesterol-derivatives, fatty acids (and other peroxisomal proliferators), as well as so-called orphan receptors, proteins that are structurally similar to other members of this group, but for which no ligands are known (Escriva, H. et al., Ligand binding was acquired during evolution of nuclear receptors, PNAS, 94, 6803 – 6808, 1997). Orphan receptors may be indicative of unknown signaling pathways in the cell or may be nuclear receptors that function without ligand activation. The activation of transcription by some of these orphan receptors may occur in the absence of an exogenous ligand and/or through signal transduction pathways originating from the cell surface (Mangelsdorf, D. J. et al., The nuclear receptor superfamily: the second decade, Cell 83, 835-839, 1995).

In general, three functional domains have been defined in NRs. An amino terminal domain is believed to have some regulatory function. A DNA-binding domain hereinafter referred to as "DBD" usually comprises two zinc finger elements and recognizes a specific Hormone Responsive Element hereinafter referred to as "HRE" within the promoters of responsive genes. Specific amino acid residues in the "DBD" have been shown to confer DNA sequence binding specificity (Schena, M. & Yamamoto, K.R., Mammalian Glucocorticoid Receptor Derivatives Enhance Transcription in Yeast, Science, 241:965-967, 1988). A Ligand-binding-domain hereinafter referred to as "LBD" is at the carboxy-terminal region of known NRs. In the absence of hormone, the LBD of some but not all NRs appears to interfere with the interaction of the DBD with its HRE. Hormone binding seems to result in a conformational change in the NR and thus opens this interference (Brzozowski et al., Molecular basis of agonism and antagonism in the oestrogen receptor, Nature, 389, 753 – 758, 1997; Wagner et al., A structural role for hormone in the thyroid hormone receptor, Nature, 378, 690 – 697. 1995). A NR without the HBD constitutively activates transcription but at a low level.

Coactivators or transcriptional activators are proposed to bridge between sequence specific transcription factors and the basal transcription machinery and in addition to influence the chromatin structure of a target cell. Several proteins like SRC-1, ACTR, and Grip1 interact with NRs in a ligand enhanced manner (Heery et al., A signature motif in transcriptional coactivators mediates binding to nuclear receptors, Nature, 387, 733 – 736; Heinzel et al., A complex containing N-CoR, mSin3 and histone deacetylase mediates transcriptional repression, Nature 387, 43 – 47, 1997). Furthermore, the physical interaction with repressing receptor-interacting proteins or corepressors has been demonstrated (Xu et al., Coactivator and Corepressor complexes in nuclear receptor function, Curr Opin Genet Dev, 9 (2), 140 – 147, 1999).

Nuclear receptor modulators like steroid hormones affect the growth and function of specific cells by binding to intracellular receptors and forming nuclear receptor-ligand complexes. Nuclear receptor-hormone complexes then interact with a hormone response element (HRE) in the control region of specific genes and alter specific gene expression.

The term LXR (Liver X Receptor) includes all subtypes of this receptor. Specifically LXR includes LXRa (also known as LXRalpha, RLD-1 and NR1H3) and LXRb (also known as LXRbeta, NER, NER1, UR, OR-1, R1P15 and NH1H2) and ligands of LXR should be under-

stood to include ligands of LXRa or LXRb. LXR is a prototypical type 2 nuclear receptor which activates genes upon binding to promoter region of target genes in a prototypical heterodimeric fashion with Retinoid X Receptor (hereinafter RXR, Forman et al., Cell, 81, 687-93, 1995). The relevant physiological ligands of LXR seem to be oxidized derivatives of cholesterol, including 22-hydroxycholesterol and 24,25(S)-epoxycholesterol (Lehmann, et al., Biol. Chem. 272(6), 3137-40, 1997). The oxysterol ligands bound to LXR were found to regulate the expression of several genes that participate in cholesterol metabolism (Janowski, et al., Nature, 383, 728-31, 1996).

LXR is proposed to be a hepatic oxysterol sensor. Upon activation (e.g. binding of oxysterols) it influences the conversion of dietary cholesterol into bile acids by upregulating the transcription of key genes which are involved in bile acid synthesis such as CYP7A1. Hence, activation of LXR in the liver could result in an increased synthesis of bile acids from cholesterol which could lead to decreased levels of hepatic cholesterol. This proposed LXR function in hepatic cholesterol metabolism was experimentally confirmed using knockout mice. Mice lacking the receptor LXRa lost their ability to respond normally to an increase in dietary cholesterol and did not induce transcription of the gene encoding CYP7A1. This resulted in accumulation of large quantities of cholesterol in the livers and impaired hepatic function. (Peet, et al., Cell, 93, 693-704, 1998).

Besides its important function in liver, LXR plays an important role in the regulation of cholesterol homeostasis in macrophages and intestinal mucosa cells where it upregulates cholesterol transporters from the ABC (=ATP binding cassette) family of membrane proteins (Repa, et al., J Biol Chem. 2002 May 24;277(21):18793-800). These transporters are believed to be crucially involved in the uptake of cholesterol from the diet since mutations in their genes leads to diseases such as sitosterolemia (Berge, et al., Science (2000);290(5497):1771-5.).

Other members of the ABC transporter family seem to be responsible for the efflux of cholesterol from loaded macrophages, a process which is thought to prevent the generation of atherosclerotic lesions. Stimulation of LXR by synthetic ligands might result in an increased cholesterol efflux from macrophages and a decreased deposition of atherosclerotic plaques (Venkateswaran, et al., PNAS (2000) 24;97(22):12097-102; Sparrow, et al., J Biol Chem (2002) 277(12):10021-7; Joseph, et al., PNAS (2002);99(11):7604-9).

However, in animal studies it was observed that activation of LXR in the liver by full agonists does not only increase bile acid synthesis but also stimulates the de novo synthesis of fatty acids and triglygerids through the upregulation of key enzymes such as Fatty Acid Synthase (FAS) or Stearyl-CoA Desaturase (SCD-1). (Schultz, et al., Genes Dev (2000) 14(22):2831-8.

Therefore, an ideal synthetic LXR binding compound should have properties that retain the agonistic activity on hepatic bile acid formation and ABC-transporter -mediated decrease in cholesterol uptake from the diet and increased cholesterol efflux from macrophages. In parallel such a compound should lack the hyperlipidemic potential which is exerted through increased fatty acid and triclyceride synthesis.

To date few compounds have been described which bind the LXR receptor and thus show utility for treating diseases or conditions which are due to or influenced by said nuclear receptor (Collins, et al., J Med Chem. (2002) 45(10):1963-6; Schultz, et al., Genes Dev (2000) 14(22):2831-8; Sparrow, et al., J Biol Chem (2002) 277(12):10021-7).

It was thus an object of the present invention to provide for compounds which by means of binding the LXR receptor act as agonist, antagonist or mixed agonist / antagonist of said receptor and thus show utility for treating diseases or conditions which are due to or influenced by said nuclear receptor.

It was further an object of the invention to provide for compounds that may be used for the manufacture of a medicament for the treatment of cholesterol associated conditions or diseases. In a preferred embodiment of the invention it was an object of the invention to provide for compounds that lower serum cholesterol and/or increase High Density lipoproteins (HDL) and/or decrease Low Density Lipoproteins (LDL). It was also an object of the invention to provide for compounds that may be used for the treatment of lipid disorders including hypercholesterolemia, atherosclerosis, Alzheimer´s disease, skin disorders, obesity and diabetes.

SUMMARY OF THE INVENTION

The present invention provides, *inter alia*, novel LXR nuclear receptor protein binding compounds according to the general formula (1) shown below. Said compounds are also binders of mammalian homologues of said receptor. Further the object of the invention was solved by

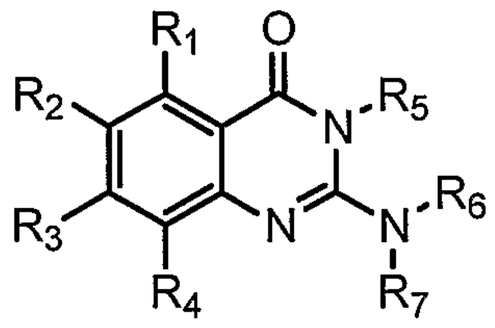
providing for amongst the LXR nuclear receptor protein binding compounds according to the general formula (1) such compounds which act as agonists, antagonists or mixed agonists / antagonists of the human LXR receptor or a mammalian homologue thereof.

The invention provides for LXR agonists that may be used for the manufacture of a medicament for the treatment of cholesterol associated conditions or diseases. In a preferred embodiment of the invention it was an object of the invention to provide for compounds that lower serum cholesterol and/or increase High Density lipoproteins (HDL) and/or decrease Low Density Lipoproteins (LDL). It was also an object of the invention to provide for compounds that may be used for the treatment of lipid disorders including hypercholesterolemia, atherosclerosis, Alzheimer´s disease, skin disorders, obesity and diabetes.

The foregoing merely summarizes certain aspects of the present invention and is not intended, nor should it be construed, to limit the invention in any manner. All patents and other publications recited herein are hereby incorporated by reference in their entirety.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

The invention provides for a compound according to the following formula (1), or pharmaceutical acceptable salts or solvates thereof, hereinafter also referred to as the "compounds according to the invention" including particular and preferred embodiments thereof, wherein



(formula (1))

R_1, R_2, R_3 and/or R_4, is independently from each other selected from H, halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 al-

kyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N- (phenylsulfonyl)amino or phenyl, wherein the phenyl is substituted or unsubstituted, such that, for example, a biphenyl results. R_5 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl, R_6 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl, R_7 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl, R_6 and R_7 may be taken together with nitrogen to form a heterocycle or substituted heterocycle or a heteroaryl or substituted heteroaryl ring.

The compounds of the invention can also exist as solvates and hydrates. Thus, these compounds may crystallize with, for example, waters of hydration, or one, a number of, or any fraction thereof of molecules of the mother liquor solvent. The solvates and hydrates of such compounds are included within the scope of this invention.

The symbol “H” denotes a hydrogen atom.

The term "C_1 to C_7 acyl" encompasses groups such as formyl, acetyl, propionyl, butyryl, pentanoyl, pivaloyl, hexanoyl, heptanoyl, benzoyl and the like. Preferred acyl groups are acetyl and benzoyl.

The term "C_1 to C_7 substituted acyl" denotes the acyl group substituted by one or more, and preferably one or two, halogen, hydroxy, protected hydroxy, oxo, protected oxo, cyclohexyl, naphthyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, guanidino, heterocyclic ring, substituted heterocyclic ring, imidazolyl, indolyl, pyrrolidinyl, C_1 to C_7 alkoxy, C_1 to C_7 acyl, C_1 to C_7 acyloxy, nitro, C_1 to C_6 alkyl ester, carboxy, protected carboxy, carbamoyl, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N,N-di(C_1 to C_6 alkyl)carboxamide, cyano, methylsulfonylamino, thiol, C_1 to C_4 alkylthio or C_1 to C_4 alkylsulfonyl groups. The substituted acyl groups may be substituted once or more, and preferably once or twice, with the same or with different substituents.

The term "substituted phenyl" specifies a phenyl group substituted with one or more, and preferably one or two, moieties chosen from the groups consisting of halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to

C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N- (phenylsulfonyl)amino or phenyl, wherein the phenyl is substituted or unsubstituted, such that, for example, a biphenyl results.

Examples of the term "substituted phenyl" includes a mono- or di(halo)phenyl group such as 2, 3 or 4-chlorophenyl, 2,6-dichlorophenyl, 2,5-dichlorophenyl, 3,4-dichlorophenyl, 2, 3 or 4-bromophenyl, 3,4-dibromophenyl, 3-chloro-4-fluorophenyl, 2, 3 or 4-fluorophenyl and the like; a mono or di(hydroxy)phenyl group such as 2, 3 or 4-hydroxyphenyl, 2,4-dihydroxyphenyl, the protected-hydroxy derivatives thereof and the like; a nitrophenyl group such as 2, 3 or 4-nitrophenyl; a cyanophenyl group, for example, 2, 3 or 4-cyanophenyl; a mono- or di(alkyl)phenyl group such as 2, 3 or 4-methylphenyl, 2,4-dimethylphenyl, 2, 3 or 4-(iso-propyl)phenyl, 2, 3 or 4-ethylphenyl, 2, 3 or 4-(n-propyl)phenyl and the like; a mono or di(alkoxyl)phenyl group, for example, 2,6-dimethoxyphenyl, 2, 3 or 4-methoxyphenyl, 2, 3 or 4-ethoxyphenyl, 2, 3 or 4-(isopropoxy)phenyl, 2, 3 or 4-(t-butoxy)phenyl, 3-ethoxy-4-methoxyphenyl and the like; 2, 3 or 4-trifluoromethylphenyl; a mono- or dicarboxyphenyl or (protected carboxy)phenyl group such as 2, 3 or 4-carboxyphenyl or 2,4-di(protected carboxy)phenyl; a mono-or di(hydroxymethyl)phenyl or (protected hydroxymethyl)phenyl such as 2, 3, or 4-(protected hydroxymethyl)phenyl or 3,4-di(hydroxymethyl)phenyl; a mono- or di(aminomethyl)phenyl or (protected aminomethyl)phenyl such as 2, 3 or 4-(aminomethyl)phenyl or 2,4-(protected aminomethyl)phenyl; or a mono- or di(N-(methylsulfonylamino))phenyl such as 2, 3 or 4-(N-(methylsulfonylamino))phenyl. Also, the term "substituted phenyl" represents disubstituted phenyl groups wherein the substituents are different, for example, 3-methyl-4-hydroxyphenyl, 3-chloro-4-hydroxyphenyl, 2-methoxy-4-bromophenyl, 4-ethyl-2-hydroxyphenyl, 3-hydroxy-4-nitrophenyl, 2-hydroxy 4-chlorophenyl and the like.

The term "heteroaryl" means a heterocyclic aromatic derivative which is a five-membered or six-membered ring system having from 1 to 4 heteroatoms, such as oxygen, sulfur and/or nitrogen, in particular nitrogen, either alone or in conjunction with sulfur or oxygen ring atoms.

Examples of heteroaryls include pyridinyl, pyrimidinyl, and pyrazinyl, pyridazinyl, pyrrolo, furano, thiopheno, oxazolo, isoxazolo, phthalimido, thiazolo and the like.

The term "substituted heteroaryl" means the above-described heteroaryl is substituted with, for example, one or more, and preferably one or two, substituents which are the same or different which substituents can be halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino or N-(phenylsulfonyl)amino groups.

The term "substituted naphthyl" specifies a naphthyl group substituted with one or more, and preferably one or two, moieties either on the same ring or on different rings chosen from the groups consisting of halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_7 alkoxy, C_1 to C_7 acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino or N-(phenylsulfonyl)amino.

Examples of the term "substituted naphthyl" includes a mono or di(halo)naphthyl group such as 1, 2, 3, 4, 5, 6, 7 or 8-chloronaphthyl, 2, 6-dichloronaphthyl, 2, 5-dichloronaphthyl, 3, 4-dichloronaphthyl, 1, 2, 3, 4, 5, 6, 7 or 8-bromonaphthyl, 3, 4-dibromonaphthyl, 3-chloro-4-fluoronaphthyl, 1, 2, 3, 4, 5, 6, 7 or 8-fluoronaphthyl and the like; a mono or di(hydroxy)naphthyl group such as 1, 2, 3, 4, 5, 6, 7 or 8-hydroxynaphthyl, 2, 4-dihydroxynaphthyl, the protected-hydroxy derivatives thereof and the like; a nitronaphthyl group such as 3- or 4-nitronaphthyl; a cyanonaphthyl group, for example, 1, 2, 3, 4, 5, 6, 7 or 8-cyanonaphthyl; a mono- or di(alkyl)naphthyl group such as 2, 3, 4, 5, 6, 7 or 8-methylnaphthyl, l, 2, 4-dimethylnaphthyl, l, 2, 3, 4, 5, 6, 7 or 8-(isopropyl)naphthyl, l, 2, 3, 4, 5, 6, 7 or 8-ethylnaphthyl, l, 2, 3, 4, 5, 6, 7 or 8-(n-propyl)naphthyl and the like; a mono or

di(alkoxy)naphthyl group, for example, 2, 6-dimethoxynaphthyl, 1, 2, 3, 4, 5, 6, 7 or 8-methoxynaphthyl, 1, 2, 3, 4, 5, 6, 7 or 8-ethoxynaphthyl, l, 2, 3, 4, 5, 6, 7 or 8-(isopropoxy)naphthyl, 1, 2, 3, 4, 5, 6, 7 or 8-(t-butoxy)naphthyl, 3-ethoxy-4-methoxynaphthyl and the like; 1, 2, 3, 4, 5, 6, 7 or 8-trifluoromethylnaphthyl; a mono- or di-carboxynaphthyl or (protected carboxy)naphthyl group such as 1, 2, 3, 4, 5, 6, 7 or 8-carboxynaphthyl or 2, 4-di(-protected carboxy)naphthyl; a mono-or di(hydroxymethyl)naphthyl or (protected hydroxymethyl)naphthyl such as 1, 2, 3, 4, 5, 6, 7 or 8-(protected hydroxymethyl)naphthyl or 3, 4-di(hydroxymethyl)naphthyl; a mono- or di(amino)naphthyl or (protected amino)naphthyl such as 1, 2, 3, 4, 5, 6, 7 or 8-(amino)naphthyl or 2, 4-(protected amino)-naphthyl, a mono- or di(aminomethyl)naphthyl or (protected aminomethyl)naphthyl such as 2, 3, or 4-(aminomethyl)naphthyl or 2, 4-(protected aminomethyl)-naphthyl; or a mono- or di-(N-methylsulfonylamino) naphthyl such as 1, 2, 3, 4, 5, 6, 7 or 8-(N-methylsulfonylamino)naphthyl. Also, the term "substituted naphthyl" represents disubstituted naphthyl groups wherein the substituents are different, for example, 3-methyl-4-hydroxynaphth-1-yl, 3-chloro-4-hydroxynaphth-2-yl, 2-methoxy-4-bromonaphth-1-yl, 4-ethyl-2-hydroxynaphth-1-yl, 3-hydroxy-4-nitronaphth-2-yl, 2-hydroxy-4-chloronaphth-1-yl, 2-methoxy-7-bromonaphth-1-yl, 4-ethyl-5-hydroxynaphth-2-yl, 3-hydroxy-8-nitronaphth-2-yl, 2-hydroxy-5-chloronaphth-1-yl and the like.

The term "C_1 to C_8 alkyl" denotes such radicals as methyl, ethyl, n-propyl, isopropyl, n-butyl, iso-butyl, sec-butyl, tert-butyl, amyl, tert-amyl, hexyl , n-heptyl, 2-heptyl, 3-heptyl, 4-heptyl, 2-methyl-1hexyl, 2-methyl-2hexyl, 2-methyl-3-hexyl, n-octyl and the like.

Examples of the above substituted alkyl groups include the 2-oxo-prop-1-yl, 3-oxo-but-1-yl, cyanomethyl, nitromethyl, chloromethyl, hydroxymethyl, tetrahydropyranyloxymethyl, trityloxymethyl, propionyloxymethyl, amino, methylamino, aminomethyl, dimethylamino, carboxymethyl, allyloxycarbonylmethyl, allyloxycarbonylaminomethyl, methoxymethyl, ethoxymethyl, t-butoxymethyl, acetoxymethyl, chloromethyl, bromomethyl, iodomethyl, trifluoromethyl, 6-hydroxyhexyl, 2,4-dichloro(n-butyl), 2-aminopropyl, 1-chloroethyl, 2-chloroethyl, 1- bromoethyl, 2-chloroethyl, 1-fluoroethyl, 2-fluoroethyl, 1- iodoethyl, 2-iodoethyl, 1-chloropropyl, 2-chloropropyl, 3- chloropropyl, 1-bromopropyl, 2-bromopropyl, 3-bromopropyl, 1-fluoropropyl, 2-fluoropropyl, 3-fluoropropyl, 1- iodopropyl, 2-iodopropyl, 3-iodopropyl, 2-aminoethyl, 1- aminoethyl, N-benzoyl-2-aminoethyl, N-acetyl-2-aminoethyl, N-benzoyl-1-aminoethyl, N-acetyl-1-aminoethyl and the like.

The term "C_1 to C_8 substituted alkyl" denotes that the above C_1 to C_8 alkyl groups are substituted by one or more, and preferably one or two, halogen, hydroxy, protected hydroxy, oxo, protected oxo, C_3 to C_7 cycloalkyl, naphthyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, guanidino, protected guanidino, heterocyclic ring, substituted heterocyclic ring, imidazolyl, indolyl, pyrrolidinyl, C_1 to C_7 alkoxy, C_1 to C_7 acyl, C_1 to C_7 acyloxy, nitro, carboxy, protected carboxy, carbamoyl, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N,N-di(C_1 to C_6 alkyl)carboxamide, cyano, methylsulfonylamino, thiol, C_1 to C_4 alkylthio or C_1 to C_4 alkylsulfonyl groups. The substituted alkyl groups may be substituted once or more, and preferably once or twice, with the same or with different substituents.

The term "C_7 to C_{12} phenylalkyl" denotes a C_1 to C_6 alkyl group substituted at any position by a phenyl, substituted phenyl, heteroaryl or substituted heteroaryl. Examples of such a group include benzyl, 2-phenylethyl, 3-phenyl(n-propyl), 4-phenylhexyl, 3-phenyl(n-amyl), 3-phenyl(sec-butyl) and the like. Preferred C_7 to C_{12} phenylalkyl groups are the benzyl and the phenylethyl groups.

The term "C_7 to C_{12} substituted phenylalkyl" denotes a C_7 to C_{12} phenylalkyl group substituted on the C_1 to C_6 alkyl portion with one or more, and preferably one or two, groups chosen from halogen, hydroxy, protected hydroxy, oxo, protected oxo, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, guanidino, protected guanidino, heterocyclic ring, substituted heterocyclic ring, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, nitro, carboxy, protected carboxy, carbamoyl, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-(C_1 to C_6 dialkyl)carboxamide, cyano, N-(C_1 to C_6 alkylsulfonyl)amino, thiol, C_1 to C_4 alkylthio, C_1 to C_4 alkylsulfonyl groups; and/or the phenyl group may be substituted with one or more, and preferably one or two, substituents chosen from halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino,

protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl) carboxamide, protected N-(C_1 to C_6 alkyl) carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N-(phenylsulfonyl)amino, cyclic C_2 to C_7 alkylene or a phenyl group, substituted or unsubstituted, for a resulting biphenyl group. The substituted alkyl or phenyl groups may be substituted with one or more, and preferably one or two, substituents which can be the same or different.

Examples of the term "C_7 to C_{12} substituted phenylalkyl" include groups such as 2-phenyl-1-chloroethyl, 2-(4-methoxyphenyl)ethyl, 4-(2,6-dihydroxy phenyl)n-hexyl, 2-(5-cyano-3-methoxyphenyl)n-pentyl, 3-(2,6-dimethylphenyl)n-propyl, 4-chloro-3-aminobenzyl, 6-(4-methoxyphenyl)-3-carboxy(n-hexyl), 5-(4-aminomethylphenyl)- 3-(aminomethyl)n-pentyl, 5-phenyl-3-oxo-n-pent-1-yl and the like.

As outlined above R_6 and R_7 may be taken together with nitrogen to form a heterocycle or substituted heterocycle of the following kind aziridine, azetidine, pyrrolidine, 3-methylpyrrolidine, 3-aminopyrrolidine, 3-hydroxypyrrolidine, pyrazolidine, imidazolidine, piperidine, 2-methylpiperidine, piperazine, morpholine, azepine, tetrahydroisoquinoline

The term "heterocycle" or "heterocyclic ring" denotes optionally substituted five-membered to eight-membered rings that have 1 to 4 heteroatoms, such as oxygen, sulfur and/or nitrogen, in particular nitrogen, either alone or in conjunction with sulfur or oxygen ring atoms. These five-membered to eight-membered rings may be saturated, fully unsaturated or partially unsaturated, with fully saturated rings being preferred. Preferred heterocyclic rings include morpholino, piperidinyl, piperazinyl, 2-amino-imidazoyl, tetrahydrofurano, pyrrolo, tetrahydrothiophen-yl, hexylmethyleneimino and heptylmethyleneimino.

The term "substituted heterocycle" or "substituted heterocyclic ring" means the above-described heterocyclic ring is substituted with, for example, one or more, and preferably one or two, substituents which are the same or different which substituents can be halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_{12} alkyl, C_1 to C_{12} alkoxy, C_1 to C_{12} substituted alkoxy, C_1 to C_{12} acyl, C_1 to C_{12} acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino carbox-

amide, protected carboxamide, N-(C_1 to C_{12} alkyl)carboxamide, protected N-(C_1 to C_{12} alkyl)carboxamide, N, N-di(C_1 to C_{12} alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_{12} alkyl)sulfonyl)amino, N-(phenylsulfonyl)amino, heterocycle or substituted heterocycle groups.

The term "C_1 to C_8 alkoxy" as used herein denotes groups such as methoxy, ethoxy, n-propoxy, isopropoxy, n-butoxy, t-butoxy and like groups. A preferred alkoxy is methoxy. The term "C_1 to C_8 substituted alkoxy" means the alkyl portion of the alkoxy can be substituted in the same manner as in relation to C_1 to C_8 substituted alkyl.

The term "C_1 to C_8 aminoacyl" encompasses groups such as formyl, acetyl, propionyl, butyryl, pentanoyl, pivaloyl, hexanoyl, heptanoyl, octanoyl, benzoyl and the like.

The term "C_1 to C_8 substituted aminoacyl" denotes the acyl group substituted by one or more, and preferably one or two, halogen, hydroxy, protected hydroxy, oxo, protected oxo, cyclohexyl, naphthyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, guanidino, heterocyclic ring, substituted heterocyclic ring, imidazolyl, indolyl, pyrrolidinyl, C_1 to C_{12} alkoxy, C_1 to C_{12} acyl, C_1 to C_{12} acyloxy, nitro, C_1 to C_{12} alkyl ester, carboxy, protected carboxy, carbamoyl, carboxamide, protected carboxamide, N-(C_1 to C_{12} alkyl)carboxamide, protected N-(C_1 to C_{12} alkyl)carboxamide, N,N-di(C_1 to C_{12} alkyl)carboxamide, cyano, methylsulfonylamino, thiol, C_1 to C_{10} alkylthio or C_1 to C_{10} alkylsulfonyl groups. The substituted acyl groups may be substituted once or more, and preferably once or twice, with the same or with different substituents.

Examples of C_1 to C_8 substituted acyl groups include 4-phenylbutyroyl, 3-phenylbutyroyl, 3-phenylpropanoyl, 2- cyclohexanylacetyl, cyclohexanecarbonyl, 2-furanoyl and 3-dimethylaminobenzoyl.

This invention provides a pharmaceutical composition comprising an effective amount of a compound according to the invention. Such compounds can be administered by various routes, for example oral, subcutaneous, intramuscular, intravenous or intracerebral. The preferred route of administration would be oral at daily doses of the compound for adult human treatment of about 0.01 -5000 mg, preferably 1-1500 mg per day. The appropriate dose may be administered in a single dose or as divided doses presented at appropriate intervals for example as two, three four or more subdoses per day.

For preparing pharmaceutical compositions containing compounds of the invention, inert, pharmaceutically acceptable carriers are used. The pharmaceutical carrier can be either solid or liquid. Solid form preparations include, for example, powders, tablets, dispersible granules, capsules, cachets, and suppositories.

A solid carrier can be one or more substances which can also act as diluents, flavoring agents, solubilizers, lubricants, suspending agents, binders, or tablet disintegrating agents; it can also be an encapsulating material.

In powders, the carrier is generally a finely divided solid which is in a mixture with the finely divided active component. In tablets, the active compound is mixed with the carrier having the necessary binding properties in suitable proportions and compacted in the shape and size desired.

For preparing pharmaceutical composition in the form of suppositories, a low-melting wax such as a mixture of fatty acid glycerides and cocoa butter is first melted and the active ingredient is dispersed therein by, for example, stirring. The molten homogeneous mixture is then poured into convenient-sized molds and allowed to cool and solidify.

Powders and tablets preferably contain between about 5% to about 70% by weight of the active ingredient. Suitable carriers include, for example, magnesium carbonate, magnesium stearate, talc, lactose, sugar, pectin, dextrin, starch, tragacanth, methyl cellulose, sodium carboxymethyl cellulose, a low-melting wax, cocoa butter and the like.

The pharmaceutical compositions can include the formulation of the active compound with encapsulating material as a carrier providing a capsule in which the active component (with or without other carriers) is surrounded by a carrier, which is thus in association with it. In a similar manner, cachets are also included. Tablets, powders, cachets, and capsules can be used as solid dosage forms suitable for oral administration.

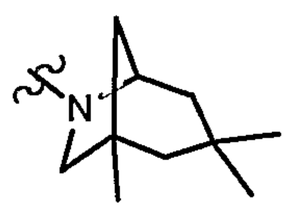
Liquid pharmaceutical compositions include, for example, solutions suitable for oral or parenteral administration, or suspensions, and emulsions suitable for oral administration. Sterile water solutions of the active component or sterile solutions of the active component in sol-

vents comprising water, ethanol, or propylene glycol are examples of liquid compositions suitable for parenteral administration.

Sterile solutions can be prepared by dissolving the active component in the desired solvent system, and then passing the resulting solution through a membrane filter to sterilize it or, alternatively, by dissolving the sterile compound in a previously sterilized solvent under sterile conditions.

In one embodiment of the present invention a compound is claimed according to formula (1) above, or pharmaceutical acceptable salts or solvates thereof, wherein R_1, R_2, R_3, R_4, is H, halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N- (phenylsulfonyl)amino or phenyl, R_5 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl, R_6 and R_7 may be taken together with nitrogen to form the heterocycle according to formula (2),

formula (2)

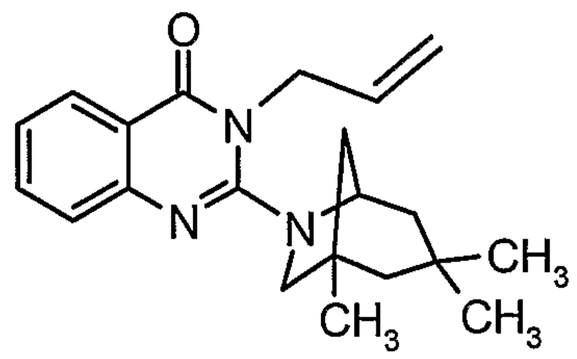


In a preferred embodiment of the invention a compound is provided, or pharmaceutical acceptable salts or solvates thereof, wherein R_1, R_2, R_3, R_4, is H, halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, pro-

tected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N- (phenylsulfonyl)amino or phenyl, R_5 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, R_6 and R_7 may be taken together with nitrogen to form the heterocycle according to formula (2) shown above.

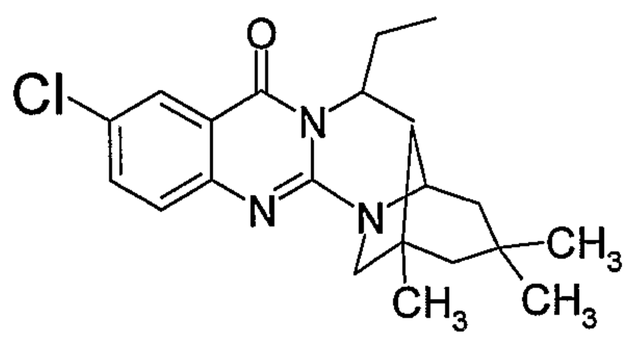
A particularly preferred compound which may act as agonist of LXR is shown in formula (6) below. The inventors have been able to demonstrate that the compound according to formula (3) has a low effective concentration at LXR with an EC_{50} of 0.5 µM wherein the EC_{50} reflects the half-maximal effective concentration, and which is higher than the EC_{50} of 0.015 µM for the published LXR agonist TO901317 (J. Schultz et al., Genes Dev. 14, 2831-2838, 2000)

formula (3) (MOLNAME 3252)

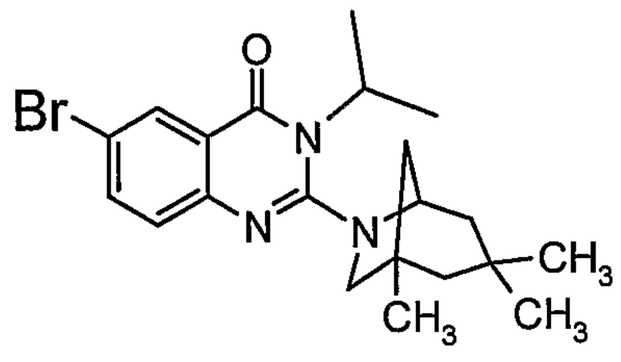


The inventors have also found the compounds according to formula (4, 5 and 6) (shown below) to be active as agonist of the LXR human nuclear receptor (see figures for details).

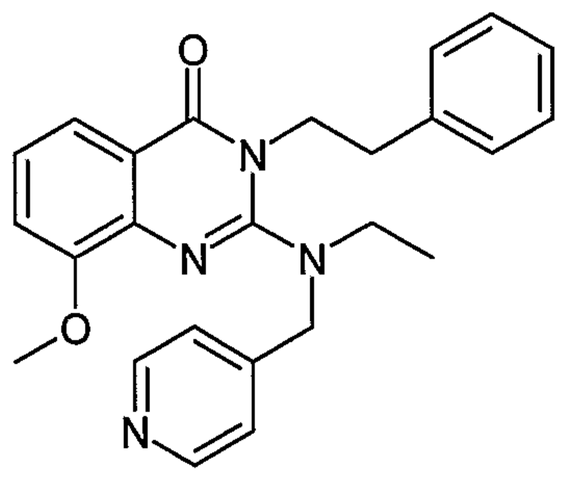
formula (4) (MOLNAME 7459)



formula (5) (MOLNAME 6584)



formula (6)) (MOLNAME 7364)



In particular the invention relates to a compound as described above wherein said compounds is capable of binding the LXR receptor protein or a portion thereof according to SEQ ID NO. 1 (Fig. 3 A to F) or a mammalian homologue thereof. The claimed compound can bind to the LXR receptor protein or a portion thereof in a mixture comprising 10-200 ng of LXR receptor protein, a fusion protein containing LXR or a portion thereof, preferably the ligand binding domain, fused to a Tag, 5-100 mM Tris /HCl at pH 6,8-8,3 ; 60-1000 mM KCl; 0-20 mM MgCl2; 100-1000ng/µl BSA in a total volume of preferably about 25 µl.).

A mammalian receptor protein homologue of the protein according to SEQ ID NO. 1 as used herein is a protein that performs substantially the same task as LXR does in humans and shares at least 40% sequence identity at the amino acid level, preferably over 50 % sequence identity at the amino acid level more preferably over 65 % sequence identity at the amino acid level, even more preferably over 75 % sequence identity at the amino acid level and most preferably over 85 % sequence identity at the amino acid level.

The invention in particular concerns a method for prevention or treatment of a LXR receptor protein or LXR receptor protein homologue mediated disease or condition in a mammal comprising administration of a therapeutically effective amount of a compound according to the invention wherein the prevention or treatment is directly or indirectly accomplished through

the binding of a compound according to the invention to the LXR receptor protein or to the LXR receptor protein homologue.

The term mediated herein means that the physiological pathway in which the LXR receptor protein acts is either directly or indirectly involved in the disease or condition to be treated or prevented. In the case where it is indirectly involved it could be that, e.g. modulating the activity of LXR by a compound according to the invention influences a parameter which has a beneficial effect on a disease or a condition. One such example is that modulation of LXR activity leads to decreased levels of serum cholesterol or certain lipoproteins which in turn have a beneficial effect on the prevention and treatment of atherosclerosis. Herein a condition is a physiological or phenotypic state which is desirably altered. One such example would be obesity which is not necessarily medically harmful but nonetheless a non desirable phenotypic condition. In a preferred embodiment of the invention the method for prevention or treatment of a LXR receptor protein mediated disease or condition is applied to a human. This may be male or female.

Pharmaceutical compositions generally are administered in an amount effective for treatment or prophylaxis of a specific condition or conditions. Initial dosing in human is accompanied by clinical monitoring of symptoms, such symptoms for the selected condition. In general, the compositions are administered in an amount of active agent of at least about 100 μg/kg body weight. In most cases they will be administered in one or more doses in an amount not in excess of about 20 mg/kg body weight per day. Preferably, in most cases, doses is from about 100 μg/kg to about 5 mg/kg body weight, daily.

For administration particularly to mammals, and particularly humans, it is expected that the daily dosage level of active agent will be 0,1 mg/kg to 10 mg/kg and typically around 1 mg/kg.

By "therapeutically effective amount" is meant a symptom- alleviating or symptom -reducing amount, a cholesterol-reducing amount, a cholesterol absorption blocking amount, a protein and/or carbohydrate digestion-blocking amount and/or a de novo cholesterol biosynthesis-blocking amount of a compound according to the invention.

Likewise, the invention concerns a method of treating in mammal a disease which is correlated with abnormal cholesterol, triglyceride, or bile acid levels or deposits comprising administering to a mammal in need of such treatment a therapeutically effective amount of a compound according to the invention.

Accordingly, the compounds according to the invention may also be used as a method of prevention or treatment of mammalian atherosclerosis, gallstone disease, lipid disorders, Alzheimer´s disease, skin disorders, obesity or cardiovascular disorders such as coronary heart disease or stroke.

The invention further concerns a method of blocking in a mammal the cholesterol absorption in the intestine in need of such blocking comprising administering to a mammal in need of such treatment a therapeutically effective amount of a compound according to the invention. The invention may also be used to treat obesity in humans.

The Liver X Receptor alpha is a prototypical type 2 nuclear receptor meaning that it activates genes upon binding to the promoter region of target genes in a heterodimeric fashion with Retinoid X Receptor. The relevant physiological ligands of LXR are oxysterols The present compounds according to the invention have been demonstrated to have a high binding efficacy (binding coefficients measured as EC50 in the range 100 nM to 1500 nM) as well as agonistic and / or antagonistic properties. Consequently they may be applied to regulate genes that participate in bile acid, cholesterol and fatty acid homeostasis as well as other downstream regulated genes. Examples of such genes are but are not limited to lipid absorption, cholesterol biosynthesis, cholesterol transport or binding, bile acid transport or binding, proteolysis, amino acid metabolism, glucose biosynthesis, protein translation, electron transport, and hepatic fatty acid metabolism. LXR often functions in vivo as a heterodimer with the Retinoid X Receptor. Published LXR agonists such as the Tularik compound "TO901317" (See figure 5) are known to influence the regulation of various liver genes. Genes found to be regulated by TO901317 can be found in figure 6. Thus, the invention also concerns a method of modulating a gene whose expression is regulated by the LXR receptor in a mammal comprising administration of a therapeutically effective amount of a compound according to the invention to said mammal.

A number of direct and indirect LXR target genes have been described whose regulated expression contribute to cholesterol homeostasis and lipogenesis. In this respect the direct regulation of Cyp7A, which was shown to be a direct target gene of LXR at least in the rodent lineage is an important aspect of cholesterol removal by increased metabolism of bile acids (Lehmann et al., J Biol.Chem. 272 (6) 3137-3140; 1007). Gupta et al. (Biochem. Biophys Res.Com, 293; 338-343, 2002) showed that LXR α regulation of Cyp7A is dominant over FXR inhibitory effects on Cyp7A transcription.

A key transcription factor that was also shown to be a direct target gene for the LXR receptor is SREBP-1C (Repa et al., Genes and Development, 14:2819-2830; 2000: Yoshikawa et al.; Mol.Cell.Biol.21 (9) 2991-3000, 2001). SREBP-1C itself activates transcription of genes involved in cholesterol and fatty acid synthesis in liver but also other mammalian tissues. Some of the SREBP1c target genes involved in lipogenesis like FAS and SCD have shown to be additionally direct targets of the LXR receptors (Joseph et al.; J Biol Chem. 2002 Mar 29;277(13):11019-25; Liang et al., J Biol Chem. 2002 Mar 15;277(11):9520-8.).

Another gene that has been shown to be directly regulated by LXRs is the LPL gene, that codes for a key enzyme that is responsible for the hydrolysis of triglycerides in circulating lipoprotein, releasing free fatty acids to peripheral tissues. (Zhang et al. J Biol Chem. 2001 Nov 16;276(46):43018-24.) This enzyme is believed to promote uptake of HDL cholesterol in liver, thereby promoting reverse cholesterol transport. A similar functional involvement in HDL clearance is described for the CETP gene product that facilitated the transfer of HDL cholesterol esters from plasma to the liver. LXR response elements were found in the CETP promoter and direct activation of this gene by LXR was demonstrated (Luo and Tall; J Clin Invest. 2000 Feb;105(4):513-20.).

The regulated transport of cholesterol through biological membranes is an important mechanism in order to maintain cholesterol homeostasis. A pivotal role in these processes in multiple tissues like e.g. macrophages and intestinal mucosa cells is maintained by the ATP-binding cassette transporter proteins (ABC). ABCA1 and ABCG1 were identified as direct LXR target genes (Costet et al.; J Biol Chem. 2000 Sep 8;275(36):28240-5) that mediate cholesterol efflux and prevent thereby e.g. generation of artherogenic plaques in macrophages (Singaraja et al. J Clin Invest. 2002 Jul;110(1):35-42). Other ABC transporters like ABCG5

and ABCG8 , primarily expressed in hepatocytes and enterocytes have also been reported to be directly responsive to LXR agonists (Repa et al., J Biol Chem. 2002 May 24;277(21):18793-800. Kennedy et al., J Biol Chem. 2001 Oct 19;276(42):39438-47) and mediate the secretion of sterols from the liver and efflux of dietary sterols from the gut .

Apolipoproteins E, C-I, C-II, and C-IV, that fulfill important roles in lipoprotein/lipid homeostasis have also been shown to be direct targets of the LXR receptor (Laffitte et al., Proc Natl Acad Sci U S A. 2001 Jan 16;98(2):507-12; Mak et al.; J Biol Chem. 2002 May 24 [epub ahead of print]). These proteins have been found to be crucial components of chylomicrons, VLDL, IDL, and HDL and are among other things associated with hypertriglyceridemia and arteriosclerosis.

Recently the LXRα itself was shown to be regulated by both LXR receptors in human cell types including macrophages suggesting an autoregulatory amplification event in the response to LXR ligands which could e.g. lead to an enhanced stimulation of LXR target genes like e.g. ABCA1 (Bolten et al.; Mol Endocrinol. 2002 Mar;16(3):506-14.; Laffitte et al., Mol Cell Biol. 2001 Nov;21(22):7558-68; Whitney et al.; J Biol Chem. 2001 Nov 23;276(47):43509-15).

Besides the important function of LXR receptors in tissues like liver and macrophages it has recently been reported that that stimulation of epidermal differentiation is mediated by Liver X receptors in murine epidermis. Differentiation maker genes like involucrin, loricin and profilaggrin have been shown to be upregulated upon LXR ligand treatment (Kömüves et al.; J Invest Dermatol. 2002 Jan;118(1):25-34.).

Another recent report describes the regulation of cholesterol homeostasis (primarily the regulation of ABCA1, ABCG1 and SREBP-1C) by the LXR receptors in the central nervous system suggesting that LXRs may prove beneficial in the treatment of CNS diseases such as Alzheimer's and Niemann-Pick disease that are known to be accompanied by dysregulation of cholesterol balance (Whitney et al.; Mol Endocrinol. 2002 Jun;16(6):1378-85).

Therefore one important embodiment the invention concerns are methods that enhances or suppresses amongst other today yet unknown LXR target genes the above mentioned genes

and the associated biological processes and pathways through LXR compounds that are subject of this invention.

The compounds according to the invention may be used as medicaments, in particular for the manufacture of a medicament for the prevention or treatment of a LXR receptor protein or LXR receptor protein homologue mediated disease or condition in a mammal wherein the prevention or treatment is directly or indirectly accomplished through the binding of the compound according to the invention to the LXR receptor protein or LXR receptor protein homologue. These pharmaceutical compositions contain 0,1 % to 99,5 % of the compound according to the invention, more particularly 0,5 % to 90 % of the compound according to the invention in combination with a pharmaceutically acceptable carrier.

The invention concerns also the use of a compound according to the invention for the manufacture of a medicament for the prevention or treatment of a LXR receptor protein mediated disease or condition wherein the mammal described above is a human. The medicament may be used for regulating the cholesterol transport system , for regulating levels of cholesterol, triglyceride, and/or bile acid in a mammal preferentially a human by activating the LXR receptor. The medicament may be used for the treatment of atherosclerosis, gallstone disease, lipid disorders, Alzherimer´s disease, skin disorders, obesity or a cardiovascular disorder.

The further concerns the use of a compound according to the invention for the manufacture of a medicament capable for blocking in a mammal, preferentially a human the cholesterol absorption in the intestine. Further the claimed compound may be used for the manufacture of a medicament for treating obesity in humans and for modulating a gene whose expression is regulated by the LXR receptor (see details above and figures).

The present invention shall now be further illustrated based on the following examples without being limited thereto. In the accompanying sequence protocol and the figures:

SEQ ID NO. 1 shows protein sequence of the LRX alpha protein a portion of which was used for cloning as described in the examples,

SEQ ID NO. 2 shows the mRNA sequence of the LRX alpha protein,

SEQ ID NO. 3 shows the protein sequence of TIF2 (Acc. No: XM_011633 RefSeq DB),

SEQ ID NO. 4 shows the respective mRNA sequence corresponding to the TIF2 protein,

SEQ ID NO 5 shows the protein sequence of the LXR beta protein a portion of which was used for cloning as described in examples,

SEQ ID NO 6 shows the mRNA sequence of the LXR beta protein,

SEQ ID NO 7 shows the sequence of primer (a) used in Example 1

SEQ ID NO 8 shows the sequence of primer (b) used in Example 1.

Fig. 1 shows the synthesis of the compounds according to the invention as also described in Example 2.

Fig. 2 shows the measurement parameters employed by the Wallace VICTOR2V™ Multilabel Counter which was used for measuring the EC_{50} values

Fig. 3 A shows SEQ ID NO. 1 which is the protein sequence of the LRX alpha protein a portion of which was used for cloning as described in the examples . Figure 3 B shows SEQ ID NO. 2 which is the mRNA sequence of the LRX alpha protein. Figure 3 C shows SEQ ID NO. 3 which is the protein sequence of TIF2 (Acc. No: XM_011633 RefSeq DB), Figure 3 D shows SEQ ID NO. 4 which is the respective mRNA sequence corresponding to the TIF2 protein. Figure 3 E shows SEQ ID NO 5 which is the protein sequence of the LXR beta protein a portion of which was used for cloning as described in examples. Figure 3 F shows SEQ ID NO 6 which is the mRNA sequence of the LXR beta protein.

Fig. 4 shows the internal molecular name used by the applicant (MOLNAME) as well as the corresponding structures of preferred compounds according to the invention. The figure further shows their respective EC_{50} values (EC50 AVG) as established according to the experiment 1 in multiple experiments (see above), as well as their respective average efficacy (% activity relative to 22-(R)-hydroxycholesterol control agonist).

Figure 5 shows various known LXR ligands. It is apparent from their structures that the inventors have identified novel compounds which are structurally not related to these known ligands.

Figure 6 shows various genes that have been found to be regulated through binding of an LXR agonist to the LXR protein.

Figure 7 shows a dose-dependent transactivation (EC50 ~ 3 μM) by LN0000007465 of the luciferase reporter gene via LXR alpha.

Figure 8 shows (A) Analysis of mRNA content of the indicated genes in total RNA isolated from THP-1 cells treated for 24 hours with 2, 10 or 25μM of LN0000006500 or 10 μM of the Tularik compound (T0901317). (B) Analysis of mRNA content fo the indicated genes in total RNA from HepG2 cells treated for 24 hours with 2, 10 or 25 μM of LN0000006500 or 10 μM of the Tularik compound (T0901317).

Figure 9 shows the dose dependent transactivation by LN0000006500 of the pFR-luc reporter gene in CHO cells via Gal4 LBD-fusion constructs derived from LXRa- or LXRb. Concentrations of the compound administered (μM) and RLU´s determined from extracts of cells are indicated.

Figure 10 shows the analysis of total cholesterol from supernatants of cultivated THP-1 cells incubated without or with ApoA1 and ApoA1 plus 10μM of the compounds Tularik (T0901317) or LN0000006500, LN0000006662, LN0000006671 or LN0000006672 as indicated.

EXAMPLES

EXAMPLE 1:

In vitro screening for compounds which influence LXR binding to coactivators.

For screening purposes a GST and 6 x His fusion of the LBD (from amino acids 155 of hLXRalpha to 447) of human LXRalpha was constructed by first cloning a Gateway cassette (Invitrogen) in frame into the Sma I site of the pAGGHLT Polylinker (Pharmingen). Then a PCR fragment specifically amplified from human liver cDNA was cloned into the resulting

pACGHLT-GW following the manufacturers instructions for Gateway cloning (Invitrogen) to yield pACGHLT-GW-hLXRalphaLBD.

Primers used for amplification were: primer (a) GGGGACAAGTTTGTACAAAAAAGCAGGCTCGCTTCGCAAATGCCGTCAG (SEQ ID NO 7), and primer (b) GGGGACCACTTTGTACAAGAAAGCTGGGTCCCCTTCTCAGTCTGTTCCACTT (SEQ ID NO 8).

100 % sequence integrity of all recombinant products was verified by sequencing. Recombinant Baculovirus was constructed from pACGHLT-GW-hLXRalphaLBD using the Pharmingen Baculovirus Expression vector system according to instructions of the manufacturer. Monolayer cultures of SF9 cells were infected by the virus as recommended by Pharmingen or 200ml cultures of 1 x10^6 cells/ml grown in 2 liter Erlenmeyer flasks on an orbital shaker at 30 rpm were infected by 10ml of same virus stock. In both cases cells were harvested 3 days after infection. All cell growth was performed in Gibco SF900 II with Glutamine (Invitrogen) medium without serum supplementation at 28°C. Since SF9 cells contain significant amounts of endogenous GST, purification was performed via His and not via GST affinity chromatography. To this end instructions of Pharmingen for purification of recombinant His tagged proteins from SF9 cells were followed with the following modifications: All detergents were omitted from the buffers and cells were lysed on ice by 5 subsequent sonication pulses using a sonicator needle at maximum power.

All eluates were dialyzed against 20 mM Tris/HCl pH 6,8, 300 mM KCl; 5 mM $MgCl_2$; 1 mM DTT; 0,2 mM PMSF; 10% Glycerol. A typical dialyzed eluate fraction contained the fusion protein at a purity of more than 80%. Total protein concentration was 0,1-0,3 mg/ml.

For E. coli expression of a NR coactivator, pDest17-hTif2BD expressing a NR interaction domain from amino acids 548-878 of human Tif2 (Acc. No: XM_011633 RefSeq) tagged by 6 N-terminal His residues was constructed. Therefore, a PCR fragment specifically amplified from human liver cDNA was subcloned into pDest 17 (Invitrogen) following the manufacturers instructions for Gateway cloning (Invitrogen).Primers used for Amplification were: primer (a)
GGGGACAAGTTTGTACAAAAAAGCAGGCTCGTTAGGGTCATCGTTGGCTTCACC

and primer (b)
GGGGACCACTTTGTACAAGAAAGCTGGGTCTCAAAGTTGCCCTGGTCGTGGGTTA
.

For E. coli expression plasmid DNA was transformed into chemically competent E. coli BL21 (Invitrogen, USA) and cells were grown to an OD600 of 0.4-0.7 before expression was induced by addition of 0,5 mM IPTG according instructions of the manufacturer (Invitrogen). After induction for 8 hours at 30°C cells were harvested by centrifugation for 10 minutes at 5000 x g. Fusion proteins were affinity purified using Ni-NTA Agarose (QIAGEN) according to the instructions of the manufacturer. Recombinant Tif2 construct was dialyzed against 20 mM Tris/HCL pH 7.9; 60 mM KCl; 5 mM $MgCl_2$; 1 mM DTT, 0,2 mM PMSF; 10% glycerol. A typical dialyzed eluate fraction contained the fusion protein at a purity of more than 80%. Total protein concentration was 0,1-0,3 mg/ml.

The TIF2 fragment was subsequently biotinylated by addition of 5-40µl/ml Tif2 fraction of a Biotinamidocaproate N-Hydroxysuccinimide-ester (Sigma) solution (20 mg/ml in DMSO). Overhead rotating samples were incubated for 2 hours at room temperature. Unincorporated label was then separated using G25 Gel filtration chromatography (Pharmacia Biotech, Sweden). Protein containing fractions from the column were pooled and tested for activity in the assay as described below.

For screening of compound libraries as provided for by the methods shown below in the examples for substances which influence the LXR/Tif 2 interaction, the Perkin Elmer LANCE technology was applied. This method relies on the binding dependent energy transfer from a donor to an acceptor fluorophore attached to the binding partners of interest. For ease of handling and reduction of background from compound fluorescence LANCE technology makes use of generic fluorophore labels and time resoved detection (for detailed description see Hemmilä I, Blomberg K and Hurskainen P, Time-resolved resonance energy transfer (TR-FRET) principle in LANCE, Abstract of Papers Presented at the 3 rd Annual Conference of the Society for Biomolecular Screening, Sep., California (1997))

For screening, 20-200 ng of biotinylated Tif 2 fragment and 10-200 ng of GST-LXR fragment were combined with 0.5-2 nM LANCE Eu-(W1024) labelled anti-GST antibody (Perkin Elmer) and 0,1-0,5µg of highly fluorescent APC-labelled streptavidin (Perkin Elmer,

AD0059) in the presence of 50µM of individual compounds to be screened in a total volume of 25 µl of 20 mM Tris /HCl pH 6,8; 300 mM KCl; 5 mM MgCl2; 100-1000 ng/µl/ BSA DMSO content of the samples was kept below 4%. Samples were incubated for a minimum of 60 minutes in the dark at room temperature in FIA-Plates black 384well med. binding (Greiner).

The LANCE signal was detected by a Perkin Elmer VICTOR2V™ Multilabel Counter applying the detection parameters listed in Fig. 2. The results were visualized by plotting the ratio between the emitted light at 665 nm and at 615 nm. For every batch of recombinant proteins amount of proteins, including BSA and labeling reagents giving the most sensitive detection of hits was determined individually by analysis of dose response curves for 22R Hydroxycholesterol and TO 901317

EXAMPLE 2:

Experimental procedure for the preparation of the compounds according to the invention.

o-AZIDOBENZOIC ACID SYNTHESIS (2)

The anthranilic acid (1, 1 eq., 0.5-1 M) was suspended in 6 M HCl, containing enough AcOH (0-20% dependent upon the anthranilic acid) to facilitate dissolution of the anthranilic acid and/or the intermediate diazonium salt, and cooled to 0 °C. $NaNO_2$ (1.1 eq., 1.3-2.5 M) dissolved in H_2O was added to the anthranilic acid solution at a rate such that the temperature of the reaction solution remained below 5 °C. The resulting homogeneous solution of the diazonium salt was slowly filtered through a sintered glass funnel into a solution of NaN_3 (1.1 eq., 0.7-1.1 M) and NaOAc (12 eq.) in H_2O. The reaction mixture was stirred/shaken for 30-60 min following cessation of vigorous N_2 evolution. Following acidification of the reaction mixture to pH 1 with concentrated HCl, the mixture was cooled to 0 °C to encourage complete precipitation of the *o*-azidobenzoic acid. The precipitate was collected by filtration and washed with 6 M HCl (2x) and H_2O (2x). The *o*-azidobenzoic acid product (2) was dried *in vacuo* (500 mtorr, 30 °C).

ACYLATION OF HYDROXYMETHYL RESIN (4)

To hydroxymethyl resin (1.0 eq., 1.3 mmol/g) and the *o*-azidobenzoic acid (1, 2.5 eq.) was added DMF (to give 400 mM *o*-azidobenzoic acid ,1), $CsCO_3$ (2.0 eq.) and KI (2.0 eq.). Following agitation of the reaction mixture for 36-48 h, the resin-bound *o*-azidobenzoic acid (4)

was washed with MeOH (2 cycles), CH_2Cl_2 (3 cycles), MeOH (3 cycles), DMF (3 cycles), MeOH (3 cycles) and CH_2Cl_2 (3 cycles), and dried *in vacuo*.

AZA-WITTIG FORMATION (5)

To the resin-bound *o*-azidobenzoic acid (4,1.0 eq.) was added a solution of PPh_3 (THF, 500 mM, 5.0 eq.). After 6 h, the resin was washed with 3 cycles of the following: THF (3 cycles), toluene (3 cycles), CH_2Cl_2 (3 cycles) and hexanes (3 cycles). Followed by drying *in vacuo* to afford resin bound iminophosphorane (5)

CARBODIIMIDE FORMATION (6)

To the resin-bound iminophosphorane (5, 1 eq.) was added isocyanate (9 , 5 eq., 450 mM) dissolved in $ClCH_2CH_2Cl$. The compounds were shaken at ambient temperature for 16 h, washed with 3 cycles of the following: THF (3 cycles), toluene (3 cycles), CH_2Cl_2 (3 cycles) and hexanes (3 cycles), and dried *in vacuo* to afford carbodiimide (6).

GUANIDINE FORMATION / CYCLIZATION

To the carbodiimide functionalized resin (6) was added secondary amine (10, 0.6 eq., 500 mM) dissolved in $ClCH_2CH_2Cl$. The reaction mixture was heated to 50 °C in an incubator for 12-72 h to afford 2-aminoquinazoline (8).

All of the final products were analyzed by HPLC using mass and an Evaporative Light Scattering Detector (ELSD) detection to determine purity and identity.

One skilled in the art will be able to arrive at the compounds claimed herein making use of said protocol.

EXAMPLE 3:

This example illustrates that a compound according to the invention (experiments shown were done with MOLSTRUCTURE LN 0000007465 (see figures 4 for structural formula)) can mediate transactivation of LXR mediated transcription in HEK293 cells.

HEK293 cells were grown in 48 well plates and co-transfected with the pTRexDest30 (Invitrogen) derivatives pTRexDest30-hLXRa, pTRexDest30-hRXR□ and the pGL2promoter (Promega) derivative pGL2promoter-LXRRE (each 300 ng of plasmid DNA). The full length

human LXR (accession U68233) and the full length human RXRα (accession P19793) were cloned into the pTRexDest30 applying the manufacturer protocols for the Gateway™ system (Invitrogen).

The LXR response elements (LXRRE) were (upper case and underlined) 5'
Ccctt**TGGTCActcaAGTTCA**agtgatgatagaattcggatcctt**TGGTCActcaAGTTCA**agtgA 3'
(SEQ ID NO. 5) derived from the rat Cyp7a promoter (Laffite et al., 2001, PNAS 98,pp 507).
Luciferase reporter activity was measured in triplicates from extracts of cells after incubating cells in culture medium (DMEM [Gibco-BRL] + 10% FCS [PAA laboratories]) for 16 hours (5% CO_2, 37^0C) containing 0,5% DMSO (control) or 0,5% DMSO with increasing concentrations of LN0000007465.

A dose-dependent transactivation (EC50 ~ 3 μM) of the reporter gene by LXRa was observed (Fig. 7).

Example 4:
This example shows that described compounds can increase the abundance of mRNA of target genes for the LXR proteins in THP-1 cells treated with TPA.

THP-1 ($3x10^5$ cells per dish) cells were seeded in 24 well dishes in 3 ml modified RPMI-1640 medium (ATTC, Cat.No. 30-2001) containing 10%FCS (GIBCO) and 100nM TPA and cultivated at 37^0C in 5% CO_2 for 48 hours. The medium was then removed and replaced with medium containing 10% charcoal/stripped FBS (Hyclone) and incubated with LN0000006500 at 2, 10 or 25 μM concentration or Tularik (T0901317) at 10 μM for 24 hours as indicated in Fig 8A as an example. HepG2 ($4,5x10^5$ cells per dish) were seeded in 24 well dishes in 3 ml DMEM Medium containing 10%FCS (GIBCO) and cultivated at 37^0C in 5% CO_2 for 48 hours as indicated in Fig 8B.

After incubation for 24 hours in presence of compound, total RNA was isolated from the cells using a Quiagen RNAeasy kit (Quiagen) according to the manufacturers protocol. The RNA was then reverse transcribed and analyzed by TaqMan Analysis using kits and equipment from Perkin-Elmer known to those knowledgeable in the field.

The fold change of mRNA abundancy of compound treated versus DMSO treated as a control is shown in Figure 8A and B for several analyzed target genes indicated in Figure 8Aand B.

Example 5:

This example shows that described compounds can selectively enhance transcription mediated by the LBD´s of the respective nuclear receptors LXRa and LXRb.

CHO cells ($1x10^5$ cells 96well plate) were co-transfected (Lipofectamine 2000 GIBCO) with pFR-luc (Stratagene) as a reporter gene construct and pCMV-AD derivatives containing the LXRa or LXRb ligand binding domains, which were cloned via the gateway system (GIBCO) described in Example 1, in order to express Gal4DBD-LXRa or Gal4DBD-LXRb fusion proteins.

Cells were grown in DMEM containing 10%FCS at 37^0C in 5% humidified CO2 for 16h in presence of 0,05% DMSO vehicle or 0,032 to 50 µM LN0000006500 in vehicle (as indicated in Fig.9). Luciferase activity was determined from aliquots of extracts prepared from cells following standard luciferase assay kits and protocols from Promega.

Example 6:

This example shows that described compounds at 10 µM concentration for 24 hours can increase the reverse cholesterol transport in THP-1 cells that were treated with TPA.

THP-1 ($1x10^6$ cells per dish) cells were seeded in 6 well dishes in 3ml modified RPMI1640 medium (ATTC, Cat.No. 30-2001) containing 10%FCS (GIBCO) and 100nM TPA and cultivated at 37^0C in 5% CO_2 for 72 hours. The medium was then removed and replaced with fresh medium containing 100 nM TPA and 0,15 % BSA. After 24 h incubation the cells were washed in PBS and 1,5 ml of fresh medium containing either 0,1% DMSO alone or 0,1% DMSO together with 40µg/ml ApoA1 (Calbiochem) or 40µg/ml ApoA1 plus the in Fig.10 as an example indicated compounds Tularik (T0901317), LN0000006500, LN0000006662, LN0000006671, LN0000006674 at 10 µM.

After incubation for 24 hours, total cholesterol was determined from cell supernatant in each of the wells using an enzymatic assay with fluorescence read-out for the determination of cholesterol (Amplex Red Cholesterol Assay Kit (A-12213). The fluorescence readout per mg

of total protein content as determined from cells that were present in the respective well are shown in Figure 9 as an example.

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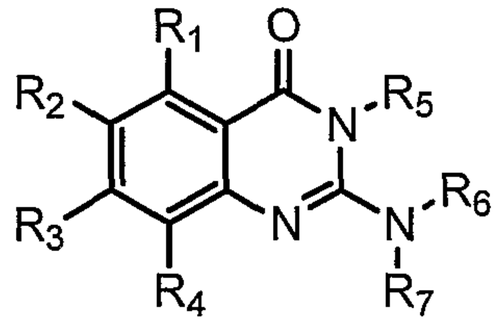
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Claims:

1. A compound of the formula (1), or pharmaceutical acceptable salts or solvates thereof according to formula (1)



wherein:

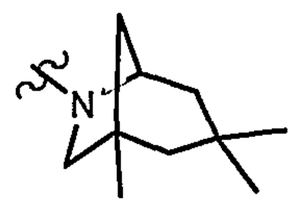
R_1, R_2, R_3 and/or R_4, is independently from each other H, halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N-(phenylsulfonyl)amino or phenyl, wherein the phenyl is substituted or unsubstituted,

R_5 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl,

R_6 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl, and

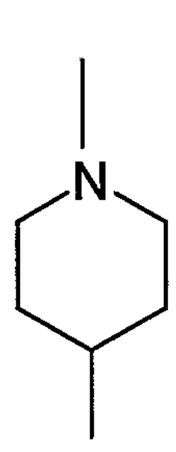
R_7 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl, C_7 to C_{12} alkylphenyl or C_7 to C_{12} substituted phenylalkyl.

2. A compound according to claim 1 wherein R_6 and R_7 are taken together with nitrogen to form a heterocycle or substituted heterocycle or a heteroaryl or substituted heteroaryl according to the following formula (2).



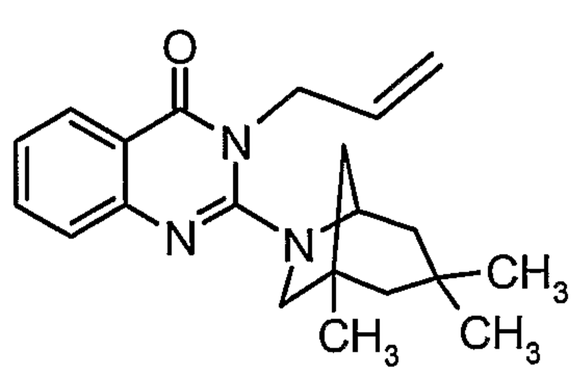
3. A compound according to claim 2, or pharmaceutical acceptable salts or solvates thereof, wherein:
R_1, R_2, R_3, R_4, is H, halogen, hydroxy, protected hydroxy, cyano, nitro, C_1 to C_6 alkyl, C_1 to C_6 substituted alkyl, C_1 to C_7 alkoxy, C_1 to C_7 substituted alkoxy, C_1 to C_7 acyl, C_1 to C_7 substituted acyl, C_1 to C_7 acyloxy, carboxy, protected carboxy, carboxymethyl, protected carboxymethyl, hydroxymethyl, protected hydroxymethyl, amino, protected amino, (monosubstituted)amino, protected (monosubstituted)amino, (disubstituted)amino, carboxamide, protected carboxamide, N-(C_1 to C_6 alkyl)carboxamide, protected N-(C_1 to C_6 alkyl)carboxamide, N, N-di(C_1 to C_6 alkyl)carboxamide, trifluoromethyl, N-((C_1 to C_6 alkyl)sulfonyl)amino, N- (phenylsulfonyl)amino or phenyl, and
R_5 is H, C_1 to C_8 alkyl, C_1 to C_8 substituted alkyl.

4. A compound according to claim 1 wherein R_6 and R_7 are taken together with nitrogen to form the heterocycle according to the following formula (3)



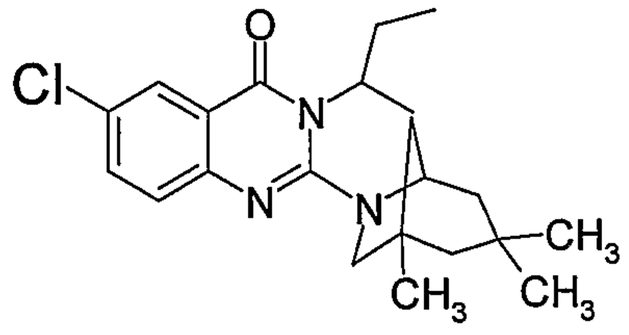
5. A compound according to any of claims 1 to 3 of the following formula (4)

formula (4)



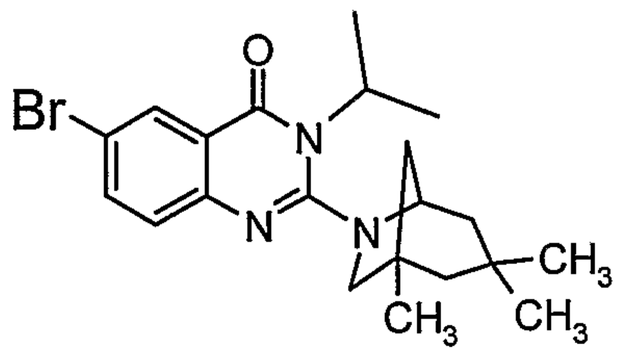
6. A compound according to any of claims 1 to 3 of the following formula (5)

formula (5)



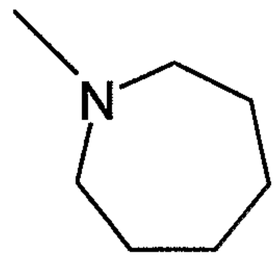
7. A compound according to any of claims 1 to 3 of the following formula (6)

formula (6)



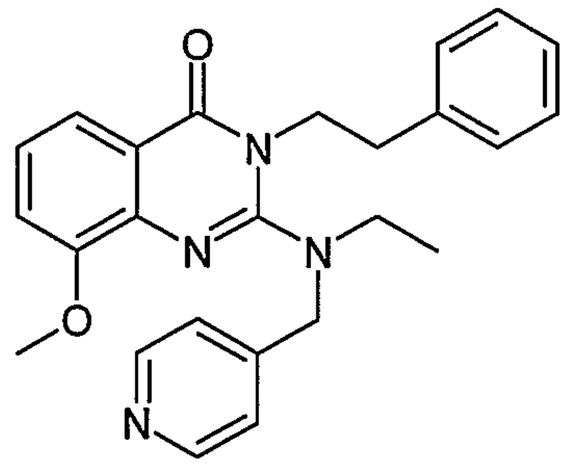
8. A compound according to any of claims 1 and 4 wherein R_6 and R_7 are taken together with nitrogen to form the heterocycle according to the following formula (7)

formula (7)



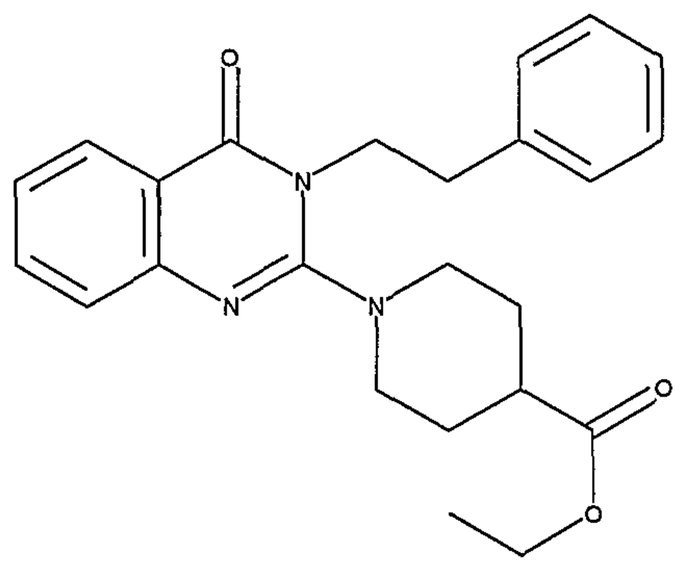
9. A compound according to claim1 according to the following formula (8)

formula (8)



10. A compound according to claims 1 and 4 according to the following formula (8)

formula (8)



11. A compound according to any of claims 1 to 10 wherein said compound is capable of binding the NR1H3 receptor protein or a portion thereof according to SEQ ID NO. 1 or a mammalian homologue thereof.

12. A compound according to any of claims 1 to 10 wherein said compound is capable of binding the NR1H2 receptor protein or a portion thereof or a mammalian homologue thereof.

13. Use of a compound according to any of claims 1 to 12 as a medicament

14. A method for prevention or treatment of a NR1H3 and/or NR1H2 receptor protein mediated disease or NR1H3 and/or NR1H2 receptor protein homologue mediated disease or condition in a mammal comprising administering a therapeutically effective amount of a compound according to any of claims 1 to 12, wherein the prevention or treatment is directly or indirectly accomplished through the binding of the compound according claims 1 to 13 to the NR1H3 and/or NR1H2 receptor proteins or to the NR1H3 and/or NR1H2 receptor protein homologues.

15. A method for prevention or treatment of a NR1H3 receptor protein and/or NR1H2 receptor protein mediated disease or condition according to claim 14, wherein said mammal is a human.

16. A method for regulating the cholesterol synthesis and/or transport in a mammal which comprises activating the NR1H3 and/or NR1H2 receptors with a therapeutically effective amount of a compound according to claims 1 to 12.

17. A method of treating in a mammal a disease which is affected by cholesterol, triglyceride, or bile acid levels comprising administering to a mammal in need of such treatment a therapeutically effective amount of a compound according to claims 1 to 12.

18. A method of treating atherosclerosis, alzheimers disease, lipid disorders, obesity or a cardiovascular disorder in a mammal, in particular a human, comprising administering to a mammal in need of such treatment a therapeutically effective amount of a compound according to claims 1 to 12.

19. A method according to any of claims 14 to 18 wherein the expression of ABCA1 and/or ABCG1 and/or ABCG5 and/or ABCG8 is increased.

20. A method according to any of claims claim 14 to 19 wherein the expression of the cholesterol 7 α hydroxylase and/or the activity of the cholesteryl ester transfer protein is increased.

21. A method of blocking in a mammal the cholesterol or fatty acid absorption in the intestine of a mammal in need of such blocking comprising administering to a mammal in need of such treatment a therapeutically effective amount of a compound according to claims 1 to 12.

22. A method for treating obesity in a mammal comprising administering a therapeutically effective amount of a compound according to any of claims 1 to 12.

23. A method of modulating a gene whose expression is regulated by the NR1H3 and/or NR1H2 receptor in a mammal comprising administering a therapeutically effective amount of a compound according to claims 1 to 10.

24. A method according to any of claims claim 14 to 19 wherein the expression of the cholesterol 7 α hydroxylase and/or the activity of the cholesteryl ester transfer protein is enhanced.

25. Use of a compound according to any of claims 1 to 12 wherein the mammal is a human

26. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament for the prevention or treatment of a NR1H3 and/or NR1H2 receptor protein or NR1H3 and/or NR1H2 receptor protein homologue mediated disease or condition in a mammal wherein the prevention or treatment is directly or indirectly accomplished through the binding of the compound according claims 1 to 8 to the NR1H3 and/or NR1H2 receptor protein or NR1H3 and/or NR1H2 receptor protein homologue.

27. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament for prevention or treatment of a NR1H3 and/or NR1H2 receptor protein mediated disease or condition according to claim 26, wherein the mammal is a human.

28. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament for regulating the cholesterol transport system in a mammal by activating the NR1H3 and/or NR1H2 receptor.

29. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament for regulating levels of cholesterol, triglyceride, and/or bile acid.

30. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament for treating in a mammal atherosclerosis, alzheimer disease, gallstone disease, lipid disorders, obesity or a cardiovascular disorder.

31. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament capable for blocking in a mammal the cholesterol and/or fatty acid absorption in the intestine.

32. Use of the compound according to any of claims 1 to 12 for the manufacture of a medicament for treating obesity in a mammal.

33. Use of a compound according to any of claims 1 to 12 for the manufacture of a medicament for modulating a gene whose expression is regulated by the NR1H3 and/or NR1H2 receptor.

34. Use of a compound according to any of claims 1 to 12 in a mammal for the selective up-regulation of one or more genes selected from the group consisting of ABCA1, ABCG1, ABCG5 and ABCG8 and a down-regulation of one or more of the genes selected from the group comprising FAS and SREBP-1c, said compound showing a larger difference in regulation of the two groups of genes when compared with the regulatory behavior of T0901317 on both groups of genes.

35. Use of a compound according to claims 28, 30, 31, 32, and 34 wherein the mammal is a human.

Fig.1:

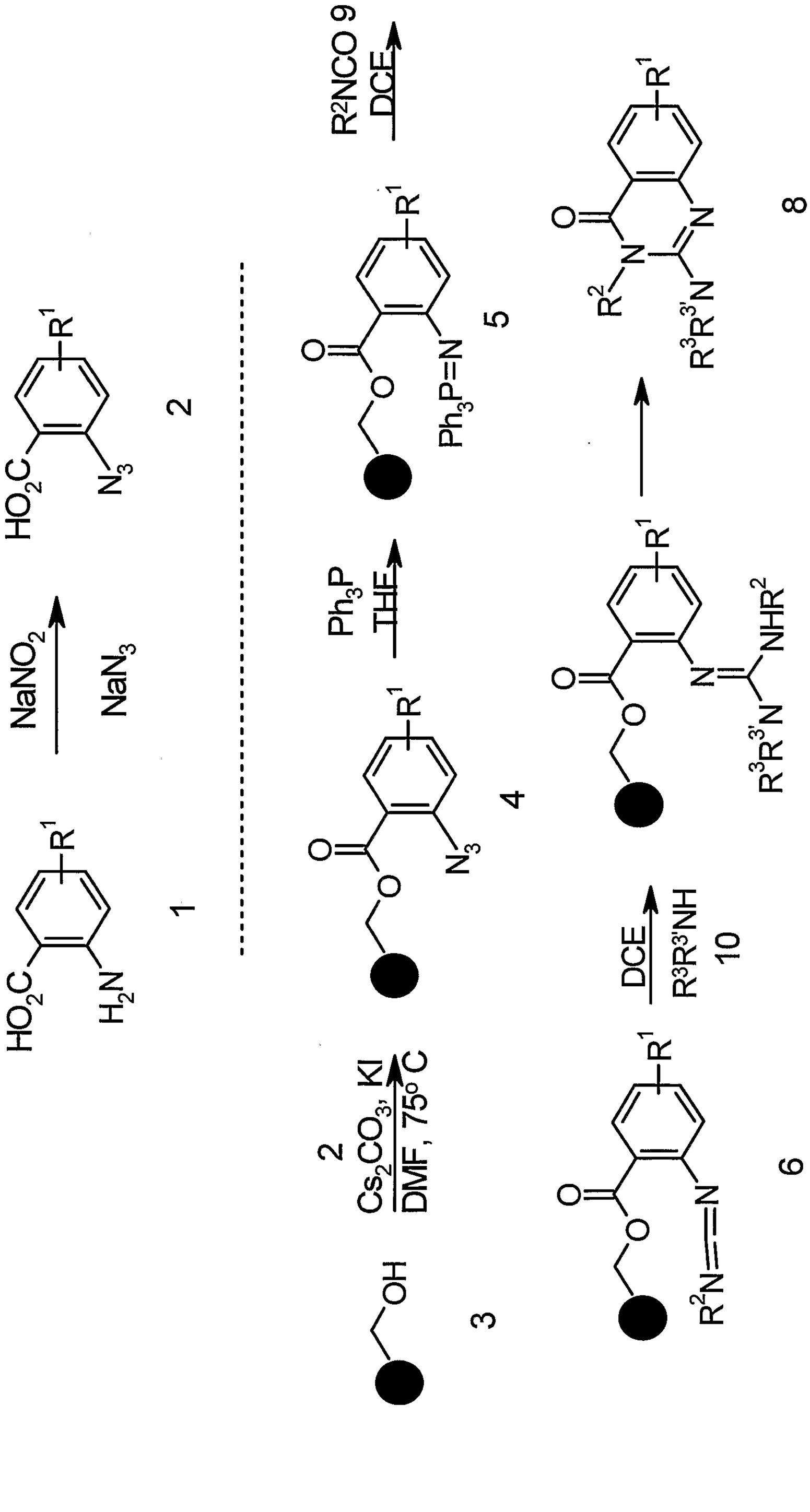

NaNO2
NaN3
Ph3P
THF
R2NCO 9
DCE
DCE
R3R3'NH
10
Cs2CO3, KI
DMF, 75°C
1
2
3
4
5
6
7
8

Fig. 2 : Measurement parameters employed by the Wallace VICTOR2V™ Multilabel Counter:

Number of repeats 1
plate: GREINER FIA-Plate black 384 well med. binding
Measurement height ..... 3.50 mm
Label technology ..... TR-F Lance
Emission filter name D615
Emission filter slot A1
Emission aperture ..... Normal
Excitation filter D340
Delay ..... 50 µs
Window time ..... 400 µs
Cycle ..... 1000 µs
Light integrator capacitors 1
Light integrator ref. level 95
Flash energy area ..... High
Flash energy level 223
Flash absorbance measurement ..... No
Beam ..... Normal
Label technology ..... TR-F Lance
Emission filter name D665
Emission filter slot A8
Emission aperture ..... Normal
Excitation filter D340
Delay ..... 50 µs
Window time ..... 400 µs
Cycle ..... 1000 µs
Light integrator capacitors 1
Light integrator ref. level 95
Flash energy area ..... High
Flash energy level 223
Flash absorbance measurement ..... No
Beam ..... Normal

Fig. 3 A and B:

A

```
   1 mslwlgapvp dippdsavel wkpgaqdass qaqggsscil reearmphsa ggtagvglea
  61 aeptalltra eppsepteir pqkrkkgpap kmlgnelcsv cgdkasgfhy nvlscegckg
 121 ffrrsvikga hyichsgghc pmdtymrrkc qecrlrkcrq agmreecvls eeqirlkklk
 181 rqeeeqahat slpprrsspp qilpqlspeq lgmieklvaa qqqcnrrsfs drlrvtpwpm
 241 apdphsrear qqrfahftel aivsvqeivd fakqlpgflq lsredqiall ktsaievmll
 301 etsrrynpgs esitflkdfs ynredfakag lqvefinpif efsramnelq lndaefalli
 361 aisifsadrp nvqdqlqver lqhtyvealh ayvsihhphd rlmfprmlmk lvslrtlssv
 421 hseqvfalrl qdkklpplls eiwdvhe
```

B

```
   1 atgtccttgt ggctgggggc ccctgtgcct gacattcctc ctgactctgc ggtggagctg
  61 tggaagccag gcgcacagga tgcaagcagc caggcccagg gaggcagcag ctgcatcctc
 121 agagaggaag ccaggatgcc ccactctgct gggggtactg caggggtggg gctggaggct
 181 gcagagccca cagccctgct caccagggca gagccccctt cagaacccac agagatccgt
 241 ccacaaaagc ggaaaaaggg gccagccccc aaaatgctgg ggaacgagct atgcagcgtg
 301 tgtggggaca aggcctcggg cttccactac aatgttctga gctgcgaggg ctgcaaggga
 361 ttcttccgcc gcagcgtcat caagggagcg cactacatct gccacagtgg cggccactgc
 421 cccatggaca cctacatgcg tcgcaagtgc caggagtgtc ggcttcgcaa atgccgtcag
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 541 cggcaagagg aggaacaggc tcatgccaca tccttgcccc ccaggcgttc ctcacccccc
 601 caaatcctgc cccagctcag cccggaacaa ctgggcatga tcgagaagct cgtcgctgcc
 661 cagcaacagt gtaaccggcg ctccttttct gaccggcttc gagtcacgcc ttggcccatg
 721 gcaccagatc cccatagccg ggaggcccgt cagcagcgct ttgcccactt cactgagctg
 781 gccatcgtct ctgtgcagga gatagttgac tttgctaaac agctacccgg cttcctgcag
 841 ctcagccggg aggaccagat tgccctgctg aagacctctg cgatcgaggt gatgcttctg
 901 gagacatctc ggaggtacaa ccctgggagt gagagtatca ccttcctcaa ggatttcagt
 961 tataaccggg aagactttgc caaagcaggg ctgcaagtgg aattcatcaa ccccatcttc
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1141 ctgcagcaca catatgtgga agccctgcat gcctacgtct ccatccacca tccccatgac
1201 cgactgatgt tcccacggat gctaatgaaa ctggtgagcc tccggaccct gagcagcgtc
1261 cactcagagc aagtgtttgc actgcgtctg caggacaaaa agctcccacc gctgctctct
1321 gagatctggg atgtgcacga atga
1431
```

Fig. 3 C and D:

C

```
MLVKPLPDSE EEGHDNQEAH QKYETMQCFA VSQPKSIKEE GEDLQSCLIC VARRVPMKER      60
PVLPSSESFT TRQDLQGKIT SLDTSTMRAA MKPGWEDLVR RCIQKFHAQH EGESVSYAKR     120
HHHEVLRQGL AFSQIYRFSL SDGTLVAAQT KSKLIRSQTT NEPQLVISLH MLHREQNVCV     180
MNPDLTGQTM GKPLNPISSN SPAHQALCSG NPGQDMTLSS NINFPINGPK EQMGMPMGRF     240
GGSGGMNHVS GMQATTPQGS NYALKMNSPS QSSPGMNPGQ PTSMLSPRHR MSPGVAGSPR     300
IPPSQFSPAG SLHSPVGVCS STGNSHSYTN SSLNALQALS EGHGVSLGSS LASPDLKMGN     360
LQNSPVNMNP PPLSKMGSLD SKDCFGLYGE PSEGTTGQAE SSCHPGEQKE TNDPNLPPAV     420
SSERADGQSR LHDSKGQTKL LQLLTTKSDQ MEPSPLASSL SDTNKDSTGS LPGSGSTHGT     480
SLKEKHKILH RLLQDSSSPV DLAKLTAEAT GKDLSQESSS TAPGSEVTIK QEPVSPKKKE     540
NALLRYLLDK DDTKDIGLPE ITPKLERLDS KTDPASNTKL IAMKTEKEEM SFEPGDQPGS     600
ELDNLEEILD DLQNSQLPQL FPDTRPGAPA GSVDKQAIIN DLMQLTAENS PVTPVGAQKT     660
ALRISQSTFN NPRPGQLGRL LPNQNLPLDI TLQSPTGAGP FPPIRNSSPY SVIPQPGMMG     720
NQGMIGNQGN LGNSSTGMIG NSASRPTMPS GEWAPQSSAV RVTCAATTSA MNRPVQGGMI     780
RNPAASIPMR PSSQPGQRQT LQSQVMNIGP SELEMNMGGP QYSQQQAPPN QTAPWPESIL     840
PIDQASFASQ NRQPFGSSPD DLLCPHPAAE SPSDEGALLD QLYLALRNFD GLEEIDRALG     900
IPELVSQSQA VDPEQFSSQD SNIMLEQKAP VFPQQYASQA QMAQGSYSPM QDPNFHTMGQ     960
RPSYATLRMQ PRPGLRPTGL VQNQPNQLRL QLQHRLQAQQ NRQPLMNQIS NVSNVNLTLR    1020
PGVPTQAPIN AQMLAQRQRE ILNQHLRQRQ MHQQQQVQQR TLMMRGQGLN MTPSMVAPSG    1080
MPATMSNPRI PQANAQQFPF PPNYGISQQP DPGFTGATTP QSPLMSPRMA HTQSPMMQQS    1140
QANPAYQAPS DINGWAQGNM GGNSMFSQQS PPHFGQQANT SMYSNNMNIN VSMATNTGGM    1200
SSMNQMTGQI SMTSVTSVPT SGLSSMGPEQ VNDPALRGGN LFPNQLPGMD MIKQEGDTTR    1260
KYC                                                                  1263
```

D

```
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  61 cctgacggcg tgaccgaccc gagccgattt ctcttggatt tggctacaca cttatagatc
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 181 aatacctctg acccctccag ggcagagaca agaaagcgca aggaatgtcc tgaccaactt
 241 ggacccagcc ccaaaaggaa cactgaaaaa cgtaatcgtg aacaggaaaa taaatatata
 301 gaagaacttg cagagttgat ttttgcaaat tttaatgata tagacaactt taacttcaaa
 361 cctgacaaat gtgcaatctt aaaagaaact gtgaagcaaa ttcgtcagat caaagaacaa
 421 gagaaagcag cagctgccaa catagatgaa gtgcagaagt cagatgtatc ctctacaggg
 481 cagggtgtca tcgacaagga tgcgctgggg cctatgatgc ttgaggccct tgatgggttc
 541 ttctttgtag tgaacctgga aggcaacgtt gtgtttgtgt cagagaatgt gacacagtat
 601 ctaaggtata accaagaaga gctgatgaac aaaagtgtat atagcatctt gcatgttggg
 661 gaccacacgg aatttgtcaa aaacctgctg ccaaagtcta taggtaaatg ggggatcttg
 721 gtctggcgaa cctccgaggc ggaacagcca taccttcaat tgtcggatgc tggtaaaacc
 781 tttacctgat tcagaagagg agggtcatga taaccaggaa gctcatcaga aatatgaaac
 841 tatgcagtgc ttcgctgtct ctcaaccaaa gtccatcaaa gaagaaggag aagatttgca
 901 gtcctgcttg atttgcgtgg caagaagagt tcccatgaag gaaagaccag ttcttccctc
 961 atcagaaagt tttactactc gccaggatct ccaaggcaag atcacgtctc tggataccag
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1081 gttccatgcg cagcatgaag gagaatctgt gtcctatgct aagaggcatc atcatgaagt
1141 actgagacaa ggattggcat tcagtcaaat ctatcgtttt tccttgtctg atggcactct
1201 tgttgctgca caaacgaaga gcaaactcat ccgttctcag actactaatg aacctcaact
1261 tgtaatatct ttacatatgc ttcacagaga gcagaatgtg tgtgtgatga atccggatct
1321 gactggacaa acgatgggga agccactgaa tccaattagc tctaacagcc ctgcccatca
1381 ggccctgtgc agtgggaacc caggtcagga catgaccctc agtagcaata taaattttcc
1441 cataaatggc ccaaaggaac aaatgggcat gcccatgggc aggtttggtg gttctggggg
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```

Fig. 3 D (continued):

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2881 aaacagtagt ccctactcag tgatacctca gccaggaatg atgggtaatc aagggatgat
2941 aggaaaccaa ggaaatttag ggaacagtag cacaggaatg attggtaaca gtgcttctcg
3001 gcctactatg ccatctggag aatgggcacc gcagagttcg gctgtgagag tcacctgtgc
3061 tgctaccacc agtgccatga accggccagt ccaaggaggt atgattcgga acccagcagc
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3181 catgaatata gggccatctg aattagagat gaacatgggg ggacctcagt atagccaaca
3241 acaagctcct ccaaatcaga ctgccccatg gcctgaaagc atcctgccta tagaccaggc
3301 gtcttttgcc agccaaaaca ggcagccatt tggcagttct ccagatgact tgctatgtcc
3361 acatcctgca gctgagtctc cgagtgatga gggagctctc ctggaccagc tgtatctggc
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3481 cagccagagc caagcagtag atccagaaca gttctcaagt caggattcca acatcatgct
3541 ggagcagaag gcgcccgttt tcccacagca gtatgcatct caggcacaaa tggcccaggg
3601 tagctattct cccatgcaag atccaaactt tcacaccatg ggacagcggc ctagttatgc
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3721 aaatcaacta agacttcaac ttcagcatcg cctccaagca cagcagaatc gccagccact
3781 tatgaatcaa atcagcaatg tttccaatgt gaacttgact ctgaggcctg gagtaccaac
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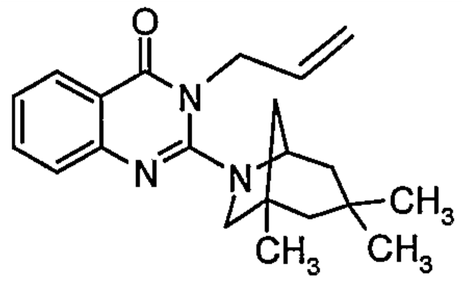
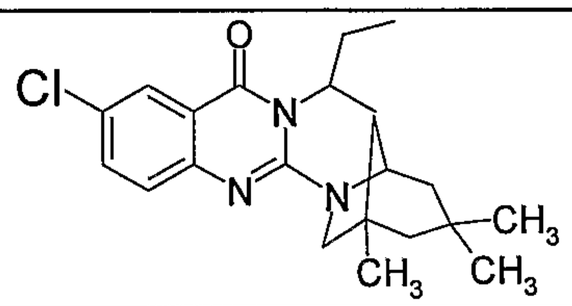
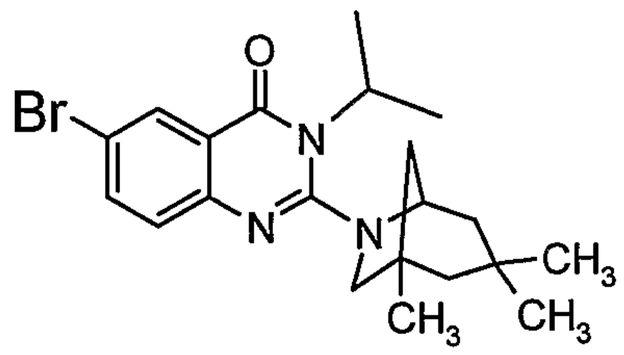
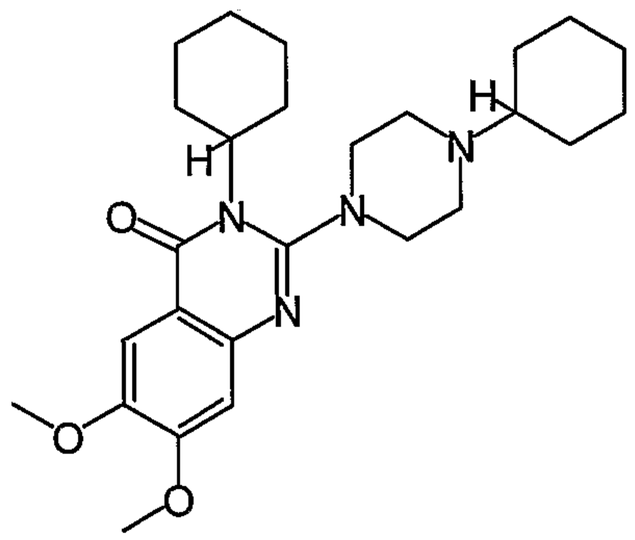
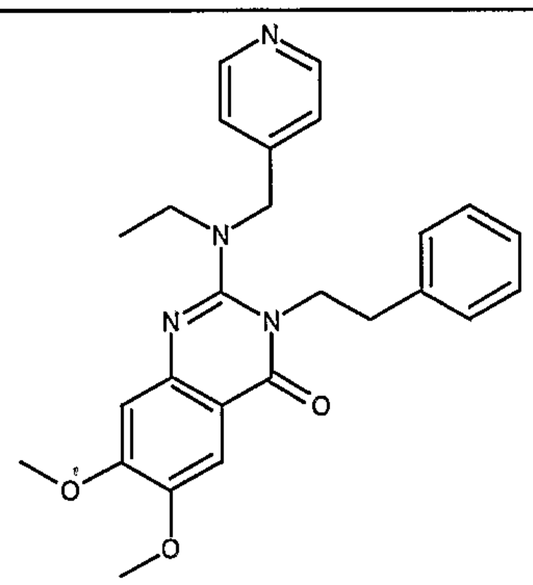
Fig. 3 E:

```
   1 MSSPTTSSLD TPLPGNGPPQ PGAPSSSPTV KEEGPEPWPG GPDPDVPGTD EASSACSTDW
  61 VIPDPEEEPE RKRKKGPAPK MLGHELCRVC GDKASGFHYN VLSCEGCKGF FRRSVVRGGA
 121 RRYACRGGGT CQMDAFMRRK CQQCRLRKCK EAGMREQCVL SEEQIRKKKI RKQQQESQSQ
 181 SQSPVGPQGS SSSASGPGAS PGGSEAGSQG SGEGEGVQLT AAQELMIQQL VAAQLQCNKR
 241 SFSDQPKVTP WPLGADPQSR DARQQRFAHF TELAIISVQE IVDFAKQVPG FLQLGREDQI
 301 ALLKASTIEI MLLETARRYN HETECITFLK DFTYSKDDFH RAGLQVEFIN PIFEFSRAMR
 361 RLGLDDAEYA LLIAINIFSA DRPNVQEPGR VEALQQPYVE ALLSYTRIKR PQDQLRFPRM
 421 LMKLVSLRTL SSVHSEQVFA LRLQDKKLPP LLSEIWDVHE
```

Fig. 3 F:

```
   1 atgtcctctc ctaccacgag ttccctggat acccccctgc ctggaaatgg ccccctcag
  61 cctggcgccc cttcttcttc acccactgta aaggaggagg gtccggagcc gtggcccggg
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1081 cggctgggcc tggacgacgc tgagtacgcc ctgctcatcg ccatcaacat cttctcggcc
1141 gaccggccca acgtgcagga gccgggccgc gtggaggcgt tgcagcagcc ctacgtggag
1201 gcgctgctgt cctacacgcg catcaagagg ccgcaggacc agctgcgctt cccgcgcatg
1261 ctcatgaagc tggtgagcct gcgcacgctg agctctgtgc actcggagca ggtcttcgcc
1321 ttgcggctcc aggacaagaa gctgccgcct ctgctgtcgg agatctggga cgtccacgag
1381 tga
```

Fig. 4 A:

MOLNAME	MOLECULE STRUCTURE	EC50 AVG	EFFIC AVG
LN0000003252		0.59	110
LN0000007459		0.12	129
LN0000011283		0.18	137
TR1040007465		1.2	114
LN0000007460		2,6	111
LN0000006500		0,34	133

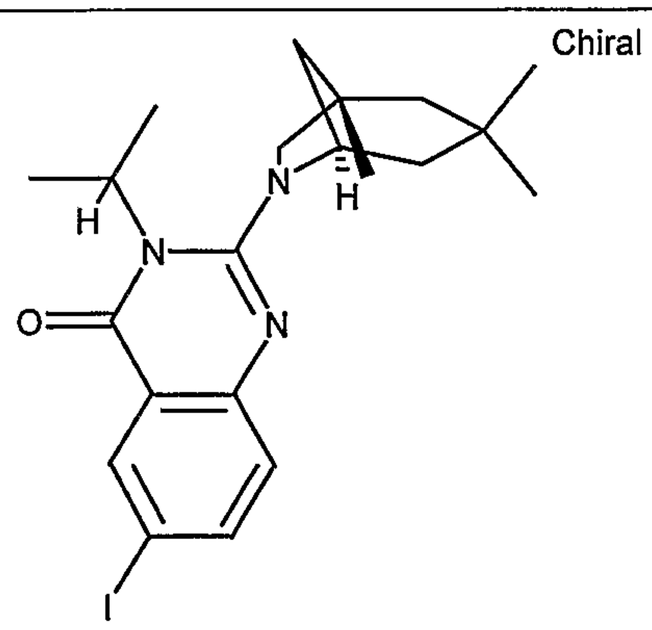
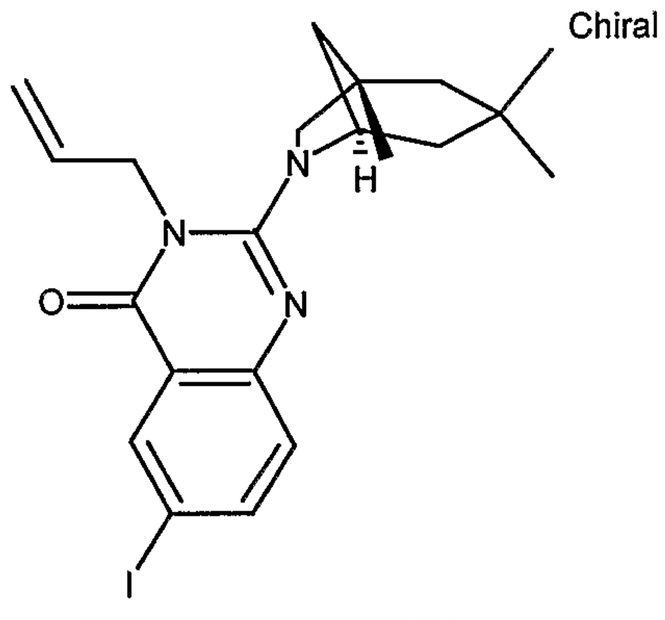
			
LN0000006494		0,23	112

Fig. 4 A (cont.)

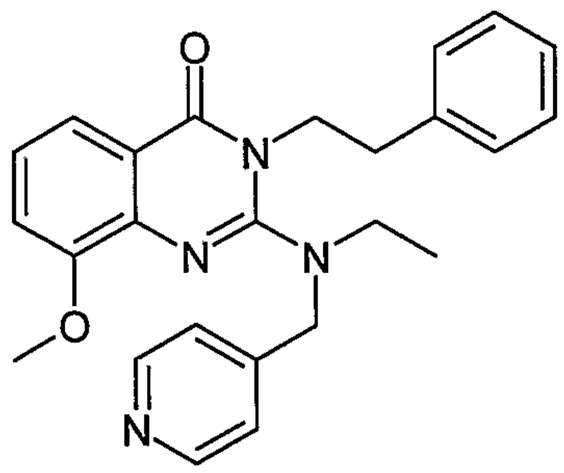
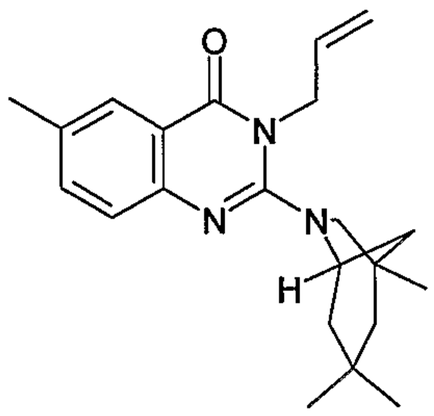
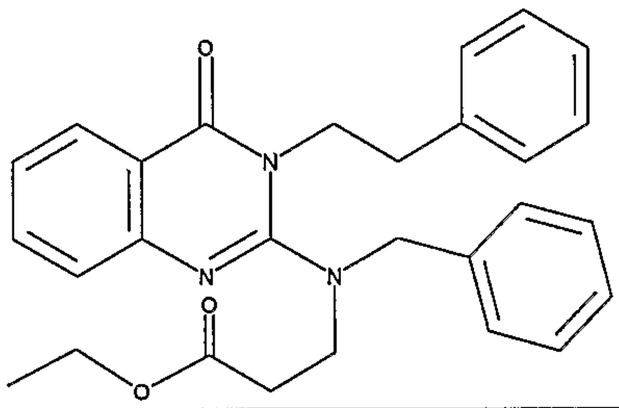
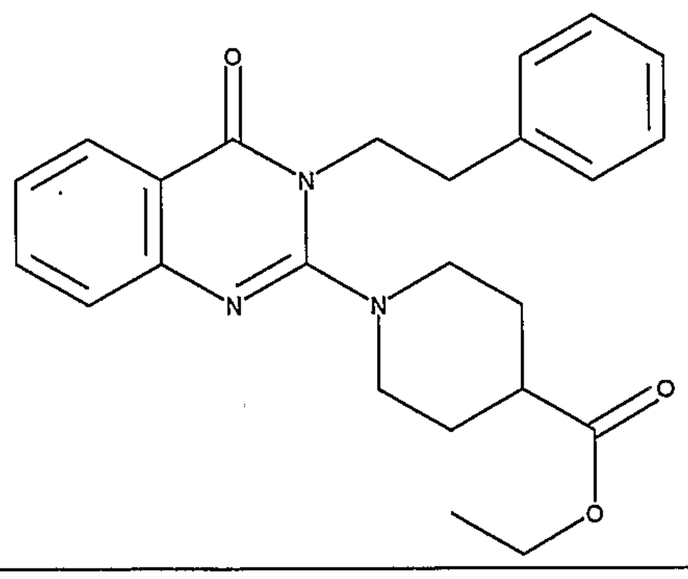
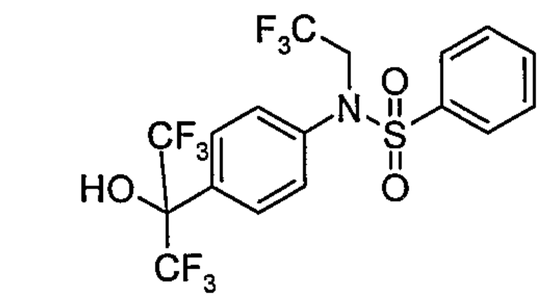
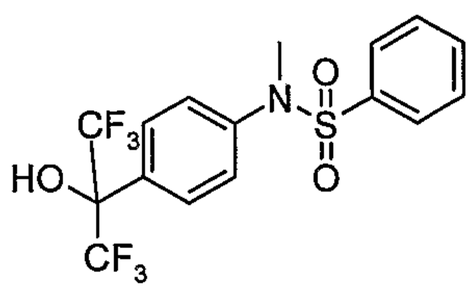
MOLNAME	MOLECULE STRUCTURE	EC50 AVG	EFFIC AVG
LN0000007364		1.5	101
LN0000003492		0.11	115
LN0000007180		0.15	128
LN0000007179		0.78	127

Fig. 4 A (cont.):

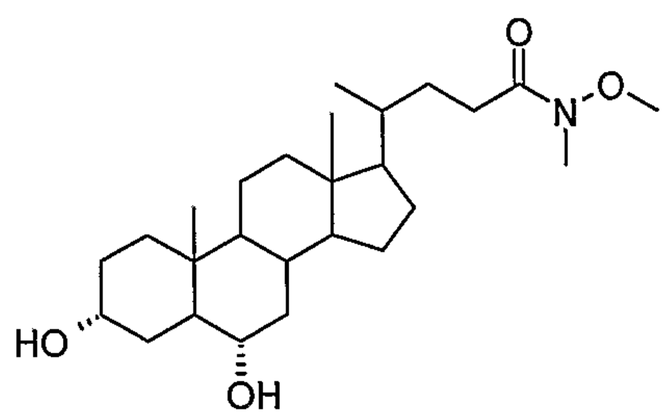
Fig. 5:



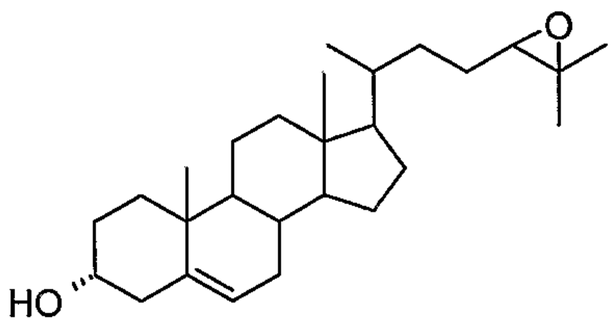
TO901317



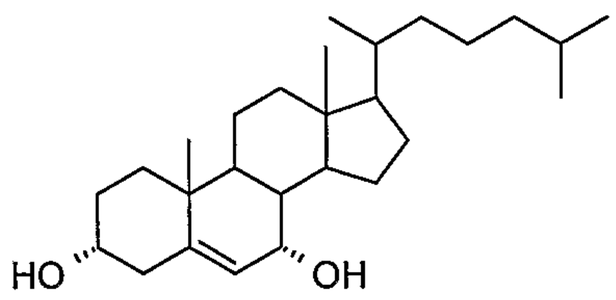
TO314407



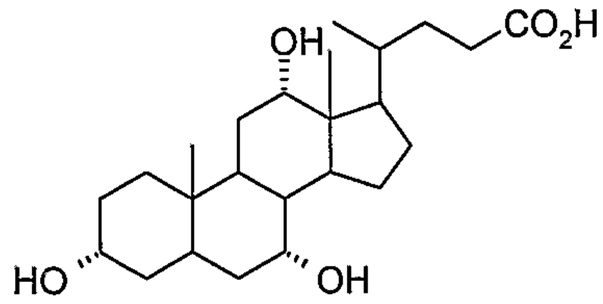
Hypocholamide



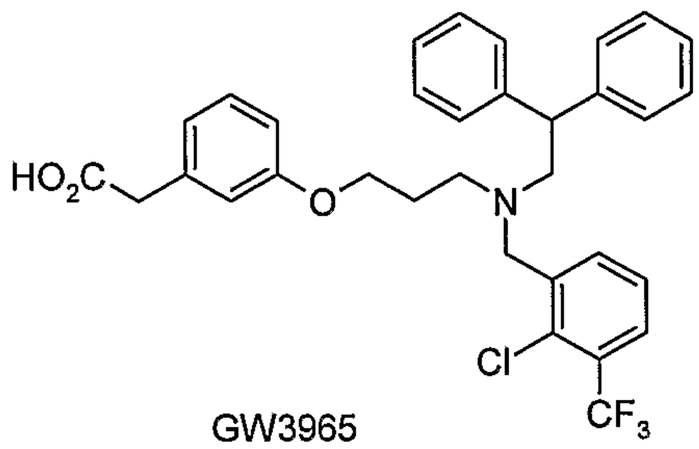
24(S),25-Epoxycholesterol



7a-Hydroxycholesterol



Cholic acid



GW3965

Fig. 6

Liver X receptor alpha, LXRα (NM_005693)
Cholesterol 7 α hydroxylase, Cyp7A1 (NM_000780)
FAS (NM_004104)
Stearyl CoA desaturase, SCD (XM_030447)
Sterol Response Element Binding Protein 1C, SREBP-1C (NM_004176)
ATP binding cassette transporter A1; ABCA1 (NM_005502)
ATP binding cassette transporter G1; ABCG1 (XM_032950)
ATP binding cassette transporter A1; ABCG5 (NM_031884)
ATP binding cassette transporter A1; ABCG8 AF324494
Apolipoprotein E, apoE (NM_000041)
Apolipoprotein C-I, apoC-I (NM_001645)
Apolipoprotein C-II apoC-II (NM_000483)
Apoliporprotein C-IV, apoC-IV (U32576)
Lipoprotein Lipase, LPL (M15856)
Cholesteryl Ester Transfer Protein, CETP (NM_000078)

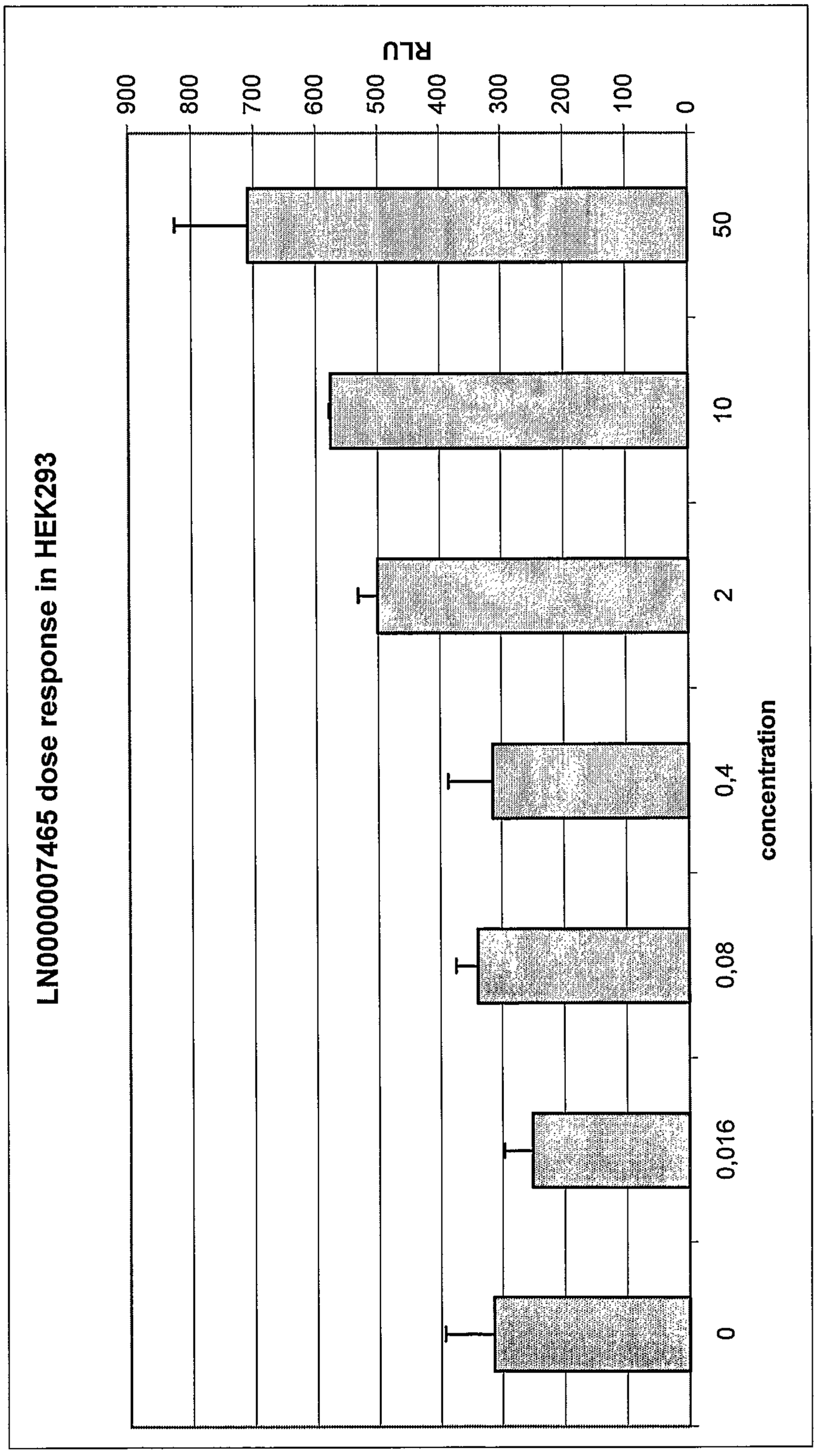

LN000007465 dose response in HEK293
RLU
900
800
700
600
500
400
300
200
100
0
50
10
2
0,4
0,08
0,016
0
concentration

Fig. 7:

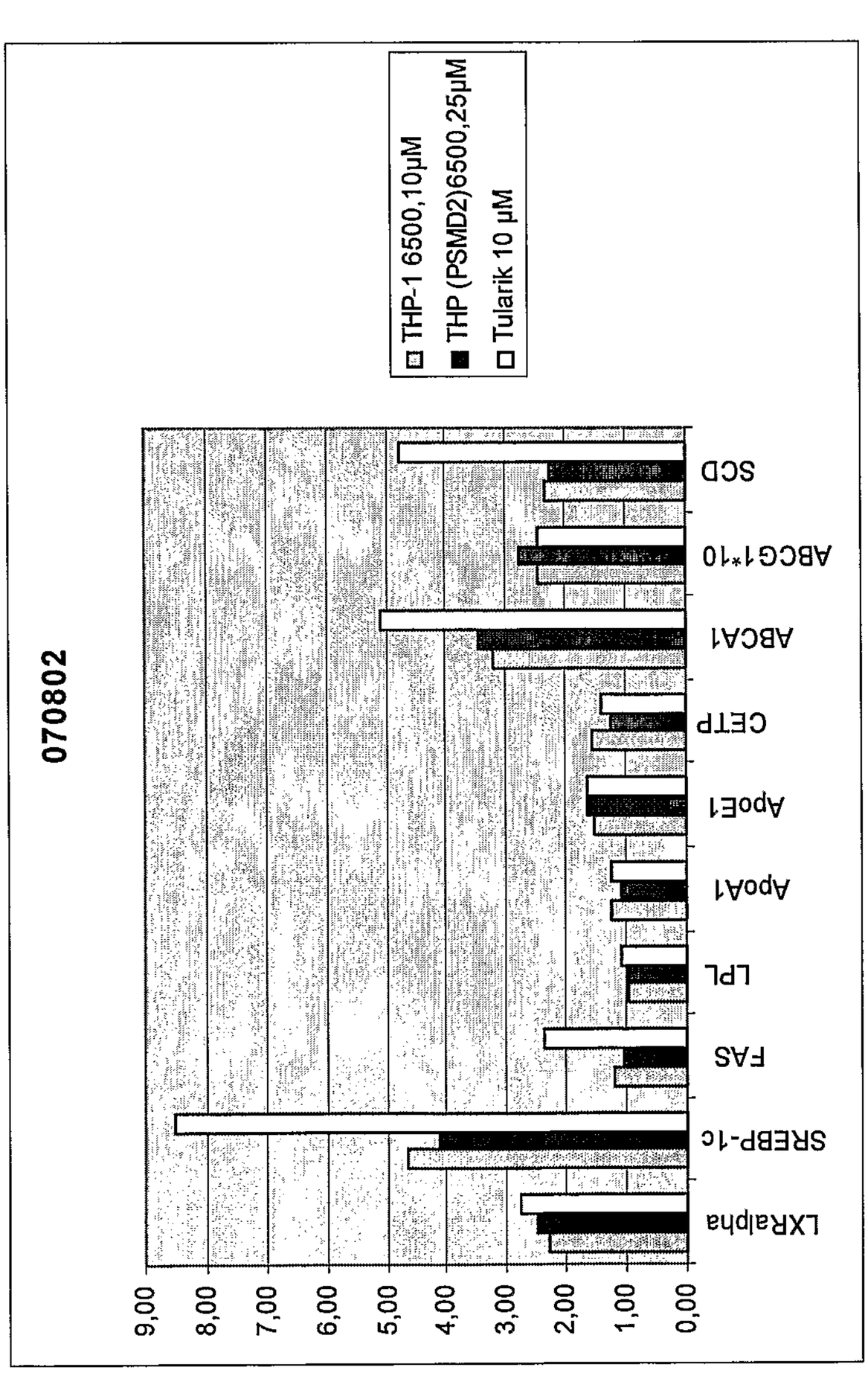

THP-1 6500,10µM
THP (PSMD2)6500,25µM
Tularik 10 µM
070802
SCD
ABCG1*10
ABCA1
CETP
ApoE1
ApoA1
LPL
FAS
SREBP-1c
LXRalpha
9,00
8,00
7,00
6,00
5,00
4,00
3,00
2,00
1,00
0,00

Fig. 8A:

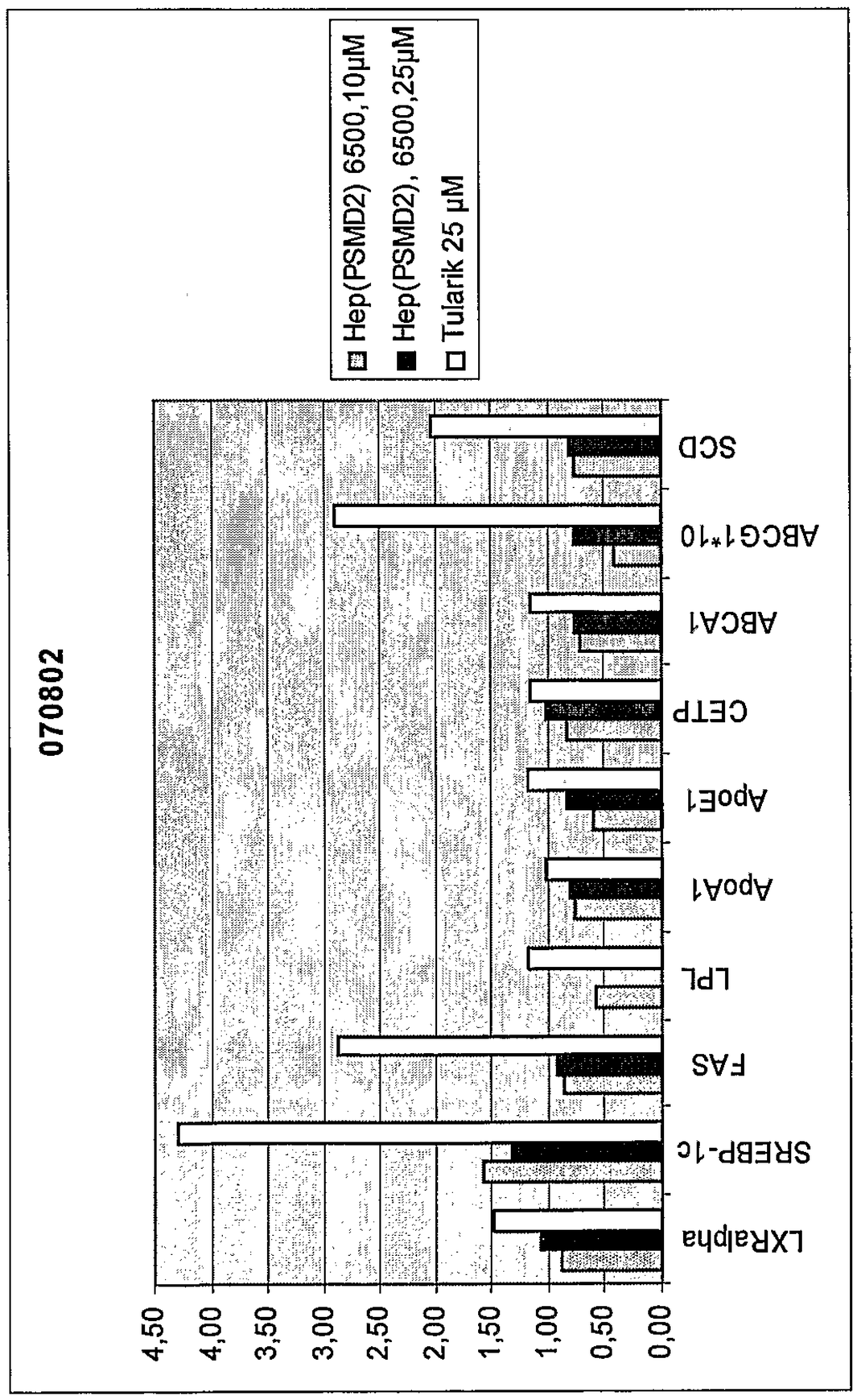

Hep(PSMD2) 6500,10µM
Hep(PSMD2), 6500,25µM
Tularik 25 µM
070802
SCD
ABCG1*10
ABCA1
CETP
ApoE1
ApoA1
LPL
FAS
SREBP-1c
LXRalpha
4,50
4,00
3,50
3,00
2,50
2,00
1,50
1,00
0,50
0,00

Fig. 8B

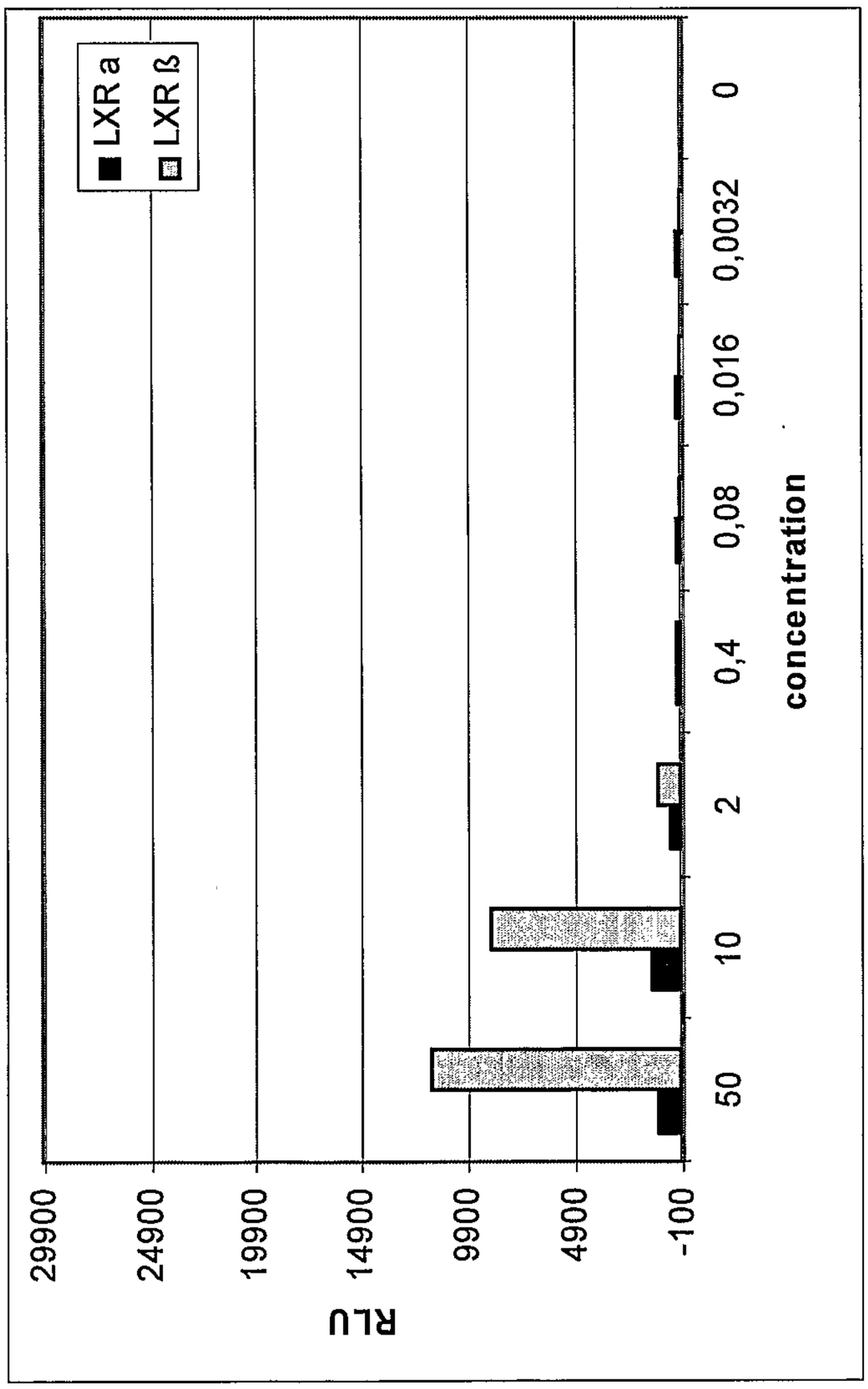

LXR a
LXR ß
0
0,0032
0,016
0,08
0,4
2
10
50
concentration
RLU
29900
24900
19900
14900
9900
4900
-100

Fig. 9

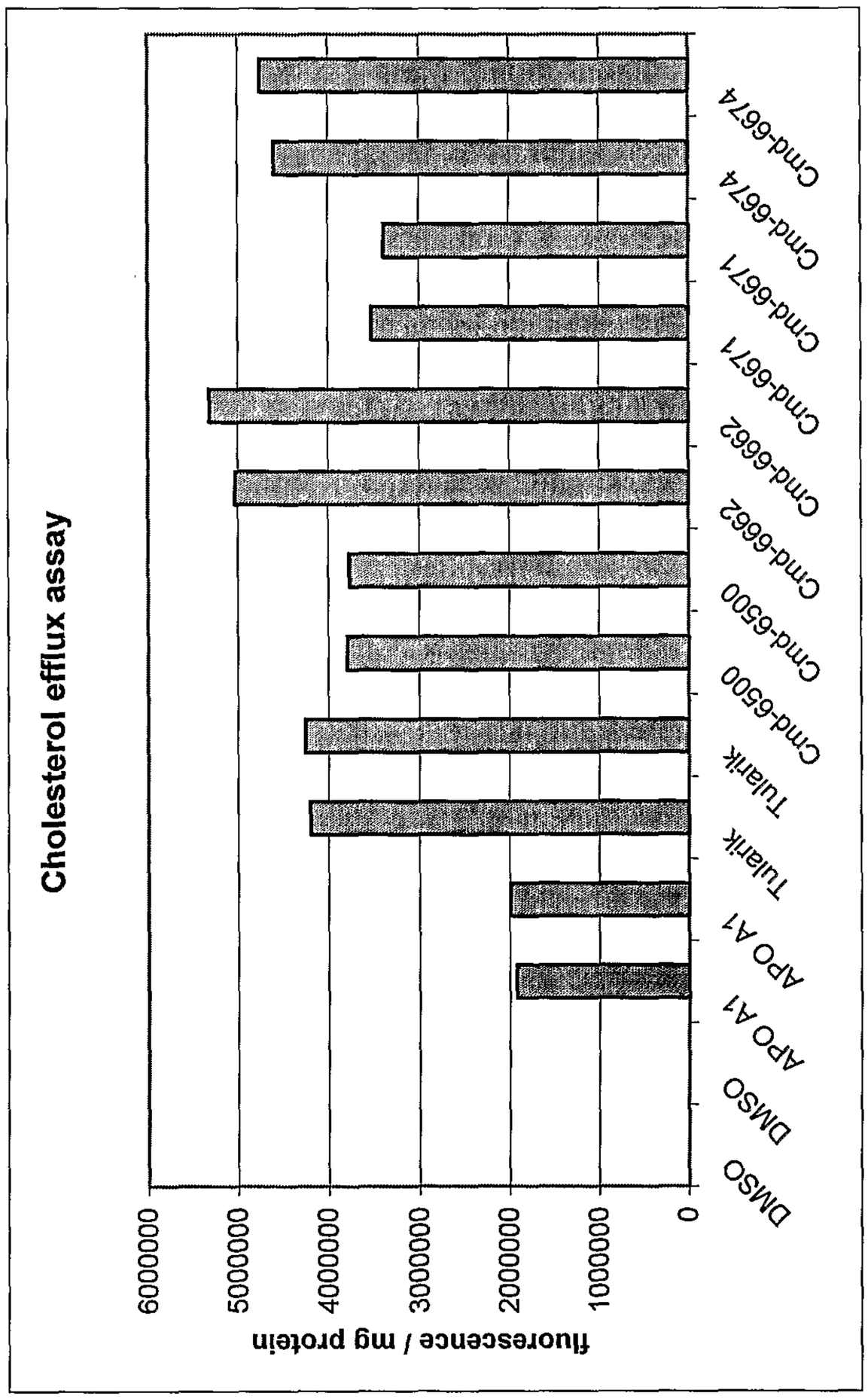
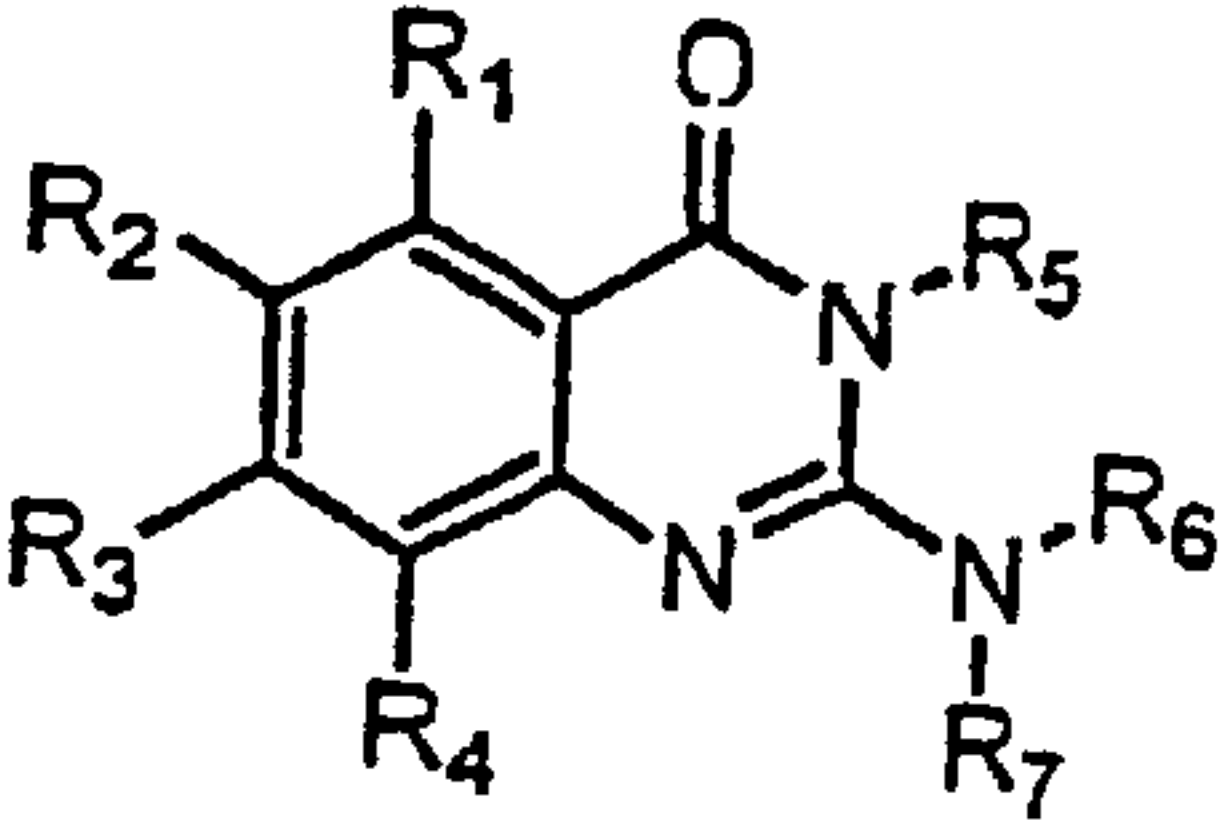

Cholesterol efflux assay
fluorescence / mg protein
6000000
5000000
4000000
3000000
2000000
1000000
0
DMSO
DMSO
APO A1
APO A1
Tularik
Tularik
Cmd-6500
Cmd-6500
Cmd-6662
Cmd-6662
Cmd-6671
Cmd-6671
Cmd-6674
Cmd-6674

Fig. 10:



(1)