



US006699871B2

(12) **United States Patent**  
**Edmondson et al.**

(10) **Patent No.:** **US 6,699,871 B2**  
(45) **Date of Patent:** **Mar. 2, 2004**

(54) **BETA-AMINO HETEROCYCLIC  
DIPEPTIDYL PEPTIDASE INHIBITORS FOR  
THE TREATMENT OR PREVENTION OF  
DIABETES**

WO	WO 98/19998	5/1998
WO	WO 00/34241	6/2000
WO	WO 01/34594 A1	5/2001
WO	WO 01/96295 A3	12/2001
WO	WO 01/96295 A2	12/2001
WO	WO 02/02560 A2	1/2002

(75) Inventors: **Scott D. Edmondson**, New York, NJ (US); **Michael H. Fisher**, Ringoes, NJ (US); **Dooseop Kim**, Westfield, NJ (US); **Malcolm Maccoss**, Freehold, NJ (US); **Emma R. Parmee**, Scotch Plains, NJ (US); **Ann E. Weber**, Scotch Plains, NJ (US); **Jinyou Xu**, Scotch Plains, NJ (US)

**OTHER PUBLICATIONS**

“Novel N-substituted-2-cyanopyrrolidines as potent inhibitors of dipeptidyl peptidase IV in the treatment of non-insulin-dependent diabetes mellitus”, Exp. Opin, Ther. Patents, 10(12), pp. 1937-1942 (2000).

(73) Assignee: **Merck & Co., Inc.**, Rahway, NJ (US)

Ashworth, et al., “2-Cyanopyrrolidides as Potent, Stable Inhibitors of Dipeptidyl Peptidase IV”, Bioorg. Med. Chem. Lett., vol. 6, No. 10, pp. 1163-1166, (1996).

(\* ) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 21 days.

Ashworth, et al., “4-Cyanothiazolidides as Very Potent, Stable Inhibitors of Dipeptidyl Peptidase IV”, Bioorg. Med. Chem. Lett., vol. 6, No. 22, pp. 2745-2748, (1996).

(21) Appl. No.: **10/189,603**

Deacon, et al., “Dipeptidyl peptidase IV inhibition as an approach to the treatment and prevention of type 2 diabetes: a historical perspective”, Biochem. Biophys. Res. Commun. vol. 294, pp. 1-4 (2002).

(22) Filed: **Jul. 5, 2002**

(65) **Prior Publication Data**

US 2003/0100563 A1 May 29, 2003

**Related U.S. Application Data**

(60) Provisional application No. 60/303,474, filed on Jul. 6, 2001.

(51) **Int. Cl.**<sup>7</sup> ..... **C07D 487/04**; A61K 31/4985; A61P 3/04; A61P 3/10

(52) **U.S. Cl.** ..... **514/249**; 544/350

(58) **Field of Search** ..... 544/350; 514/249

*Primary Examiner*—Mark L. Berch  
*Assistant Examiner*—Kahsay Habte  
(74) *Attorney, Agent, or Firm*—Philippe L. Durette; Melvin Winokur; J. Eric Thies

(57) **ABSTRACT**

The present invention is directed to compounds which are inhibitors of the dipeptidyl peptidase-IV enzyme (“DP-IV inhibitors”) and which are useful in the treatment or prevention of diseases in which the dipeptidyl peptidase-IV enzyme is involved, such as diabetes and particularly type 2 diabetes. The invention is also directed to pharmaceutical compositions comprising these compounds and the use of these compounds and compositions in the prevention or treatment of such diseases in which the dipeptidyl peptidase-IV enzyme is involved.

(56) **References Cited**

**U.S. PATENT DOCUMENTS**

5,939,560 A	8/1999	Jenkins et al.	548/535
6,166,063 A	12/2000	Villhauer	514/423
6,303,661 B1	10/2001	Demuth et al.	514/866

**FOREIGN PATENT DOCUMENTS**

WO WO 97/40832 11/1997

**26 Claims, No Drawings**

**BETA-AMINO HETEROCYCLIC  
DIPEPTIDYL PEPTIDASE INHIBITORS FOR  
THE TREATMENT OR PREVENTION OF  
DIABETES**

CROSS REFERENCE TO RELATED  
APPLICATIONS

This application claims priority under 35 U.S.C. §119(e) from Serial No. 60/303,474, filed Jul. 6, 2001.

BACKGROUND OF THE INVENTION

Diabetes refers to a disease process derived from multiple causative factors and characterized by elevated levels of plasma glucose or hyperglycemia in the fasting state or after administration of glucose during an oral glucose tolerance test. Persistent or uncontrolled hyperglycemia is associated with increased and premature morbidity and mortality. Often abnormal glucose homeostasis is associated both directly and indirectly with alterations of the lipid, lipoprotein and apolipoprotein metabolism and other metabolic and hemodynamic disease. Therefore patients with Type 2 diabetes mellitus are at especially increased risk of macrovascular and microvascular complications, including coronary heart disease, stroke, peripheral vascular disease, hypertension, nephropathy, neuropathy, and retinopathy. Therefore, therapeutic control of glucose homeostasis, lipid metabolism and hypertension are critically important in the clinical management and treatment of diabetes mellitus.

There are two generally recognized forms of diabetes. In type 1 diabetes, or insulin-dependent diabetes mellitus (IDDM), patients produce little or no insulin, the hormone which regulates glucose utilization. In type 2 diabetes, or noninsulin dependent diabetes mellitus (NIDDM), patients often have plasma insulin levels that are the same or even elevated compared to nondiabetic subjects; however, these patients have developed a resistance to the insulin stimulating effect on glucose and lipid metabolism in the main insulin-sensitive tissues, which are muscle, liver and adipose tissues, and the plasma insulin levels, while elevated, are insufficient to overcome the pronounced insulin resistance.

Insulin resistance is not primarily due to a diminished number of insulin receptors but to a post-insulin receptor binding defect that is not yet understood. This resistance to insulin responsiveness results in insufficient insulin activation of glucose uptake, oxidation and storage in muscle and inadequate insulin repression of lipolysis in adipose tissue and of glucose production and secretion in the liver.

The available treatments for type 2 diabetes, which have not changed substantially in many years, have recognized limitations. While physical exercise and reductions in dietary intake of calories will dramatically improve the diabetic condition, compliance with this treatment is very poor because of well-entrenched sedentary lifestyles and excess food consumption, especially of foods containing high amounts of saturated fat. Increasing the plasma level of insulin by administration of sulfonylureas (e.g. tolbutamide and glipizide) or meglitinide, which stimulate the pancreatic  $\beta$ -cells to secrete more insulin, and/or by injection of insulin when sulfonylureas or meglitinide become ineffective, can result in insulin concentrations high enough to stimulate the very insulin-resistant tissues. However, dangerously low levels of plasma glucose can result from administration of insulin or insulin secretagogues (sulfonylureas or meglitinide), and an increased level of insulin resistance due to the even higher plasma insulin levels can occur. The biguanides increase insulin sensitivity resulting in some

correction of hyperglycemia. However, the two biguanides, phenformin and metformin, can induce lactic acidosis and nausea/diarrhea. Metformin has fewer side effects than phenformin and is often prescribed for the treatment of Type 2 diabetes.

The glitazones (i.e. 5-benzylthiazolidine-2,4-diones) are a more recently described class of compounds with potential for ameliorating many symptoms of type 2 diabetes. These agents substantially increase insulin sensitivity in muscle, liver and adipose tissue in several animal models of type 2 diabetes resulting in partial or complete correction of the elevated plasma levels of glucose without occurrence of hypoglycemia. The glitazones that are currently marketed are agonists of the peroxisome proliferator activated receptor (PPAR), primarily the PPAR-gamma subtype. PPAR-gamma agonism is generally believed to be responsible for the improved insulin sensitization that is observed with the glitazones. Newer PPAR agonists that are being tested for treatment of Type II diabetes are agonists of the alpha, gamma or delta subtype, or a combination of these, and in many cases are chemically different from the glitazones (i.e., they are not thiazolidinediones). Serious side effects (e.g. liver toxicity) have occurred with some of the glitazones, such as troglitazone.

Additional methods of treating the disease are still under investigation. New biochemical approaches that have been recently introduced or are still under development include treatment with alpha-glucosidase inhibitors (e.g. acarbose) and protein tyrosine phosphatase-1B (PTP-1B) inhibitors.

Compounds that are inhibitors of the dipeptidyl peptidase-IV ("DP-IV" or "DPP-IV") enzyme are also under investigation as drugs that may be useful in the treatment of diabetes, and particularly type 2 diabetes. See for example WO 97/40832, WO 98/19998, U.S. Pat. No. 5,939,560, *Bioorg. Med. Chem. Lett.*, 6(10), 1163-1166 (1996); and *Bioorg. Med. Chem. Lett.*, 6(22), 2745-2748 (1996). The usefulness of DP-IV inhibitors in the treatment of type 2 diabetes is based on the fact that DP-IV in vivo readily inactivates glucagon like peptide-1 (GLP-1) and gastric inhibitory peptide (GIP). GLP-1 and GIP are incretins and are produced when food is consumed. The incretins stimulate production of insulin. Inhibition of DP-IV leads to decreased inactivation of the incretins, and this in turn results in increased effectiveness of the incretins in stimulating production of insulin by the pancreas. DP-IV inhibition therefore results in an increased level of serum insulin. Advantageously, since the incretins are produced by the body only when food is consumed, DP-IV inhibition is not expected to increase the level of insulin at inappropriate times, such as between meals, which can lead to excessively low blood sugar (hypoglycemia). Inhibition of DP-IV is therefore expected to increase insulin without increasing the risk of hypoglycemia, which is a dangerous side effect associated with the use of insulin secretagogues.

DP-IV inhibitors also have other therapeutic utilities, as discussed herein. DP-IV inhibitors have not been studied extensively to date, especially for utilities other than diabetes. New compounds are needed so that improved DP-IV inhibitors can be found for the treatment of diabetes and potentially other diseases and conditions.

SUMMARY OF THE INVENTION

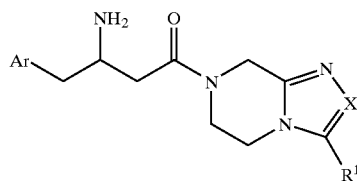
The present invention is directed to compounds which are inhibitors of the dipeptidyl peptidase-IV enzyme ("DP-IV inhibitors") and which are useful in the treatment or prevention of diseases in which the dipeptidyl peptidase-IV

3

enzyme is involved, such as diabetes and particularly type 2 diabetes. The invention is also directed to pharmaceutical compositions comprising these compounds and the use of these compounds and compositions in the prevention or treatment of such diseases in which the dipeptidyl peptidase-IV enzyme is involved.

### DETAILED DESCRIPTION OF THE INVENTION

The present invention is directed to compounds of the formula I:



wherein:

Ar is phenyl which is unsubstituted or substituted with 1-5 of  $R^3$ , wherein  $R^3$  is independently selected from the group consisting of:

- (1) halogen,
- (2)  $C_{1-6}$ alkyl, which is linear or branched and is unsubstituted or substituted with 1-5 halogens,
- (3)  $OC_{1-6}$ alkyl, which is linear or branched and is unsubstituted or substituted with 1-5 halogens, and
- (4) CN;

X is selected from the group consisting of:

- (1) N, and
- (2)  $CR^2$ ;

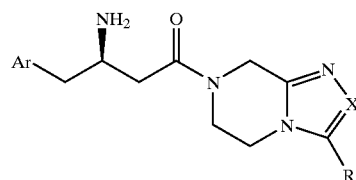
$R^1$  and  $R^2$  are independently selected from the group consisting of:

- (1) hydrogen,
- (2) CN,
- (3)  $C_{1-10}$ alkyl, which is linear or branched and which is unsubstituted or substituted with 1-5 halogens or phenyl, which is unsubstituted or substituted with 1-5 substituents independently selected from halogen, CN, OH,  $R^4$ ,  $OR^4$ ,  $NHSO_2R^4$ ,  $SO_2R^4$ ,  $CO_2H$ , and  $CO_2C_{1-6}$ alkyl, wherein the  $CO_2C_{1-6}$ alkyl is linear or branched,
- (4) phenyl which is unsubstituted or substituted with 1-5 substituents independently selected from halogen, CN, OH,  $R^4$ ,  $OR^4$ ,  $NHSO_2R^4$ ,  $SO_2R^4$ ,  $CO_2H$ , and  $CO_2C_{1-6}$ alkyl, wherein the  $CO_2C_{1-6}$ alkyl is linear or branched, and
- (6) a 5- or 6-membered heterocycle which may be saturated or unsaturated comprising 1-4 heteroatoms independently selected from N, S and O, the heterocycle being unsubstituted or substituted with 1-3 substituents independently selected from oxo, OH, halogen,  $C_{1-6}$ alkyl, and  $OC_{1-6}$ alkyl, wherein the  $C_{1-6}$ alkyl and  $OC_{1-6}$ alkyl are linear or branched and optionally substituted with 1-5 halogens;

$R^4$  is  $C_{1-6}$ alkyl, which is linear or branched and which is unsubstituted or substituted with 1-5 groups independently selected from halogen,  $CO_2H$ , and  $CO_2C_{1-6}$ alkyl, wherein the  $CO_2C_{1-6}$ alkyl is linear or branched; and pharmaceutically acceptable salts thereof and individual diastereomers thereof.

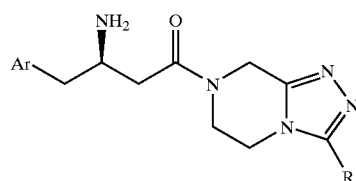
4

An embodiment of the present invention includes compounds of the formula Ia:



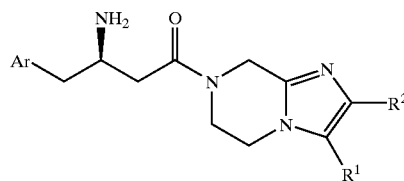
wherein X, Ar and  $R^1$  are defined herein; and pharmaceutically acceptable salts and individual diastereomers thereof.

Another embodiment of the present invention includes compounds of the formula Ib:



wherein Ar and  $R^1$  are defined herein; and pharmaceutically acceptable salts and individual diastereomers thereof.

Another embodiment of the present invention includes compounds of the formula Ic:



wherein Ar,  $R^1$  and  $R^2$  are defined herein; and pharmaceutically acceptable salts thereof and individual diastereomers thereof.

In the present invention it is preferred that Ar is phenyl which is unsubstituted or substituted with 1-5 substituents which are independently selected from the group consisting of:

- (1) fluoro,
- (2) bromo, and
- (3)  $CF_3$ .

In the present invention it is more preferred that Ar is selected from the group consisting of:

- (1) phenyl,
- (2) 2-fluorophenyl,
- (3) 3,4-difluorophenyl,
- (4) 2,5-difluorophenyl,
- (5) 2,4,5-trifluorophenyl,
- (6) 2-fluoro-4-(trifluoromethyl)phenyl, and
- (7) 4-bromo-2,5-difluorophenyl.

In the present invention it is preferred that  $R^1$  is selected from the group consisting of:

- (1) hydrogen, and
- (2)  $C_{1-6}$ alkyl, which is linear or branched and which is unsubstituted or substituted with phenyl or 1-5 fluoro

5

In the present invention it is more preferred that R<sup>1</sup> is selected from the group consisting of:

- (1) hydrogen,
- (2) methyl,
- (3) ethyl,
- (4) CF<sub>3</sub>,
- (5) CH<sub>2</sub>CF<sub>3</sub>,
- (5) CF<sub>2</sub>CF<sub>3</sub>
- (6) phenyl, and
- (7) benzyl.

In the present invention it is more preferred that R<sup>1</sup> is selected from the group consisting of:

- (1) hydrogen,
- (2) methyl,
- (3) ethyl,
- (4) CF<sub>3</sub>, and
- (5) CH<sub>2</sub>CF<sub>3</sub>.

In the present invention it is even more preferred that R<sup>1</sup> is hydrogen or CF<sub>3</sub>.

In the present invention it is preferred that R<sup>2</sup> is selected from:

- (1) hydrogen,
- (2) C<sub>1-6</sub>alkyl, which is linear or branched and which is unsubstituted or substituted with 1-5 fluoro,
- (3) phenyl, which is unsubstituted or substituted with 1-3 substituents independently selected from fluoro, OCH<sub>3</sub>, and OCF<sub>3</sub>.

In the present invention it is more preferred that R<sup>2</sup> is selected from the group consisting of:

- (1) hydrogen,
- (2) methyl,
- (3) ethyl,
- (4) CF<sub>3</sub>,
- (5) CH<sub>2</sub>CF<sub>3</sub>,
- (5) CF<sub>2</sub>CF<sub>3</sub>
- (6) phenyl,
- (7) (4-methoxy)phenyl,
- (8) (4-trifluoromethoxy)phenyl,
- (9) 4-fluorophenyl, and
- (10) 3,4-difluorophenyl.

In the present invention it is even more preferred that R<sup>2</sup> is CF<sub>3</sub> or CF<sub>2</sub>F<sub>3</sub>.

In the present invention it is preferred that R<sup>3</sup> is F, Br or CF<sub>3</sub>.

The compounds of the present invention may contain one or more asymmetric centers and can thus occur as racemates and racemic mixtures, single enantiomers, diastereomeric mixtures and individual diastereomers. The compounds of the instant invention have one asymmetric center at the beta carbon atom. Additional asymmetric centers may be present depending upon the nature of the various substituents on the molecule. Each such asymmetric center will independently produce two optical isomers and it is intended that all of the possible optical isomers and diastereomers in mixtures and as pure or partially purified compounds are included within the ambit of this invention. The present invention is meant to comprehend all such isomeric forms of these compounds.

Some of the compounds described herein contain olefinic double bonds, and unless specified otherwise, are meant to include both E and Z geometric isomers.

Some of the compounds described herein may exist as tautomers which have different points of attachment of

6

hydrogen accompanied by one or more double bond shifts. For example, a ketone and its enol form are keto-enol tautomers. The individual tautomers as well as mixtures thereof are encompassed with compounds of the present invention.

Formula I shows the structure of the class of compounds without preferred stereochemistry. Formula Ia shows the preferred stereochemistry at the carbon atom that is attached to the amine group of the beta amino acid from which these compounds are prepared.

The independent syntheses of these diastereomers or their chromatographic separations may be achieved as known in the art by appropriate modification of the methodology disclosed herein. Their absolute stereochemistry may be determined by the x-ray crystallography of crystalline products or crystalline intermediates which are derivatized, if necessary, with a reagent containing an asymmetric center of known absolute configuration.

If desired, racemic mixtures of the compounds may be separated so that the individual enantiomers are isolated. The separation can be carried out by methods well known in the art, such as the coupling of a racemic mixture of compounds to an enantiomerically pure compound to form a diastereomeric mixture, followed by separation of the individual diastereomers by standard methods, such as fractional crystallization or chromatography. The coupling reaction is often the formation of salts using an enantiomerically pure acid or base. The diastereomeric derivatives may then be converted to the pure enantiomers by cleavage of the added chiral residue. The racemic mixture of the compounds can also be separated directly by chromatographic methods utilizing chiral stationary phases, which methods are well known in the art.

Alternatively, any enantiomer of a compound may be obtained by stereoselective synthesis using optically pure starting materials or reagents of known configuration by methods well known in the art.

The term "pharmaceutically acceptable salts" refers to salts prepared from pharmaceutically acceptable non-toxic bases or acids including inorganic or organic bases and inorganic or organic acids. Salts derived from inorganic bases include aluminum, ammonium, calcium, copper, ferric, ferrous, lithium, magnesium, manganic salts, manganous, potassium, sodium, zinc, and the like. Particularly preferred are the ammonium, calcium, magnesium, potassium, and sodium salts. Salts in the solid form may exist in more than one crystal structure, and may also be in the form of hydrates. Salts derived from pharmaceutically acceptable organic non-toxic bases include salts of primary, secondary, and tertiary amines, substituted amines including naturally occurring substituted amines, cyclic amines, and basic ion exchange resins, such as arginine, betaine, caffeine, choline, N,N'-dibenzylethylene-diamine, diethylamine, 2-diethylaminoethanol, 2-dimethylaminoethanol, ethanolamine, ethylenediamine, N-ethyl-morpholine, N-ethylpiperidine, glucamine, glucosamine, histidine, hydrabamine, isopropylamine, lysine, methylglucamine, morpholine, piperazine, piperidine, polyamine resins, procaine, purines, theobromine, triethylamine, trimethylamine, tripropylamine, tromethamine, and the like.

When the compound of the present invention is basic, salts may be prepared from pharmaceutically acceptable non-toxic acids, including inorganic and organic acids. Such acids include acetic, benzenesulfonic, benzoic, camphorsulfonic, citric, ethanesulfonic, fumaric, gluconic, glutamic, hydrobromic, hydrochloric, isethionic, lactic, maleic, malic, mandelic, methanesulfonic, mucic, nitric

pamoic, pantothenic, phosphoric, succinic, sulfuric, tartaric, p-toluenesulfonic acid, and the like. Particularly preferred are citric, hydrobromic, hydrochloric, maleic, phosphoric, sulfuric, fumaric, and tartaric acids.

It will be understood that, as used herein, references to the compounds of Formula I are meant to also include the pharmaceutically acceptable salts.

As appreciated by those of skill in the art, halo or halogen as used herein are intended to include fluoro, chloro, bromo and iodo. Similarly, C<sub>1-8</sub>, as in C<sub>1-8</sub>alkyl is defined to identify the group as having 1, 2, 3, 4, 5, 6, 7 or 8 carbons in a linear or branched arrangement, such that C<sub>1-8</sub>alkyl specifically includes methyl, ethyl, n-propyl, iso-propyl, n-butyl, iso-butyl, tert-butyl, pentyl, hexyl, heptyl and octyl. Likewise, C<sub>0</sub>, as in C<sub>0</sub>alkyl is defined to identify the presence of a direct covalent bond. A group which is designated as being independently substituted with substituents may be independently substituted with multiple numbers of such substituents. The term "heterocycle" as used herein is intended to include 5- or 6-membered ring systems which are within the following listing: benzimidazolyl, benzodioxanyl, benzofuranyl, benzopyrazolyl, benzothiadiazolyl, benzotriazolyl, benzothiophenyl, benzoxadiazolyl, benzoxazolyl, carbazolyl, carbolinyl, chromanyl, cinnolinyl, furanyl, imidazolyl, indolinyl, indolyl, indolaziny, indazolyl, isobenzofuranyl, isoindolyl, isoquinolyl, isothiazolyl, isoxazolyl, naphthyridinyl, oxadiazolyl, oxazolyl, pyrazinyl, pyrazolyl, pyridopyridinyl, pyridazinyl, pyridyl, pyrimidyl, pyrrolyl, quinazoliny, quinolinyl, quinoxaliny, tetrazolyl, thiadiazolyl, thiazolidinyl, thiazolyl, thienyl, triazolyl, azetidiny, 1,4-dioxanyl, hexahydroazepinyl, piperazinyl, piperidinyl, pyrrolidinyl, morpholinyl, thiomorpholinyl, dihydrobenzimidazolyl, dihydrobenzofuranyl, dihydrobenzothiophenyl, dihydrobenzoxazolyl, dihydrofuranyl, dihydroimidazolyl, dihydroindolyl, dihydroisooxazolyl, dihydroisothiazolyl, dihydrooxadiazolyl, dihydrooxazolyl, dihydropyrazinyl, dihydropyrazolyl, dihydropyridinyl, dihydropyrimidinyl, dihydropyrrolyl, dihydroquinoliny, dihydrotetrazolyl, dihydrothiadiazolyl, dihydrothiazolyl, dihydrothienyl, dihydrotriazolyl, dihydroazetidiny, methylenedioxybenzoyl, tetrahydrofuranyl, tetrahydroimidazolyl, tetrahydroisoquinoliny, and tetrahydrothienyl.

Exemplifying the invention is the use of the compounds disclosed in the Examples and herein.

Specific compounds within the present invention include a compound which selected from the group consisting of the compounds disclosed in the following Examples and pharmaceutically acceptable salts thereof and individual diastereomers thereof.

The subject compounds are useful in a method of inhibiting the dipeptidyl peptidase-IV enzyme in a patient such as a mammal in need of such inhibition comprising the administration of an effective amount of the compound. The present invention is directed to the use of the compounds disclosed herein as inhibitors of dipeptidyl peptidase-IV enzyme activity.

In addition to primates, such as humans, a variety of other mammals can be treated according to the method of the present invention. For instance, mammals including, but not limited to, cows, sheep, goats, horses, dogs, cats, guinea pigs, rats or other bovine, ovine, equine, canine, feline, rodent or murine species can be treated. However, the method can also be practiced in other species, such as avian species (e.g., chickens).

The present invention is further directed to a method for the manufacture of a medicament for inhibiting dipeptidyl peptidase-IV enzyme activity in humans and animals comprising combining a compound of the present invention with a pharmaceutical carrier or diluent.

The subject treated in the present methods is generally a mammal, preferably a human being, male or female, in whom inhibition of dipeptidyl peptidase-IV enzyme activity is desired. The term "therapeutically effective amount" means the amount of the subject compound that will elicit the biological or medical response of a tissue, system, animal or human that is being sought by the researcher, veterinarian, medical doctor or other clinician.

The term "composition" as used herein is intended to encompass a product comprising the specified ingredients in the specified amounts, as well as any product which results, directly or indirectly, from combination of the specified ingredients in the specified amounts. Such term in relation to pharmaceutical composition, is intended to encompass a product comprising the active ingredient(s), and the inert ingredient(s) that make up the carrier, as well as any product which results, directly or indirectly, from combination, complexation or aggregation of any two or more of the ingredients, or from dissociation of one or more of the ingredients, or from other types of reactions or interactions of one or more of the ingredients. Accordingly, the pharmaceutical compositions of the present invention encompass any composition made by admixing a compound of the present invention and a pharmaceutically acceptable carrier. By "pharmaceutically acceptable" it is meant the carrier, diluent or excipient must be compatible with the other ingredients of the formulation and not deleterious to the recipient thereof.

The terms "administration of" and or "administering a" compound should be understood to mean providing a compound of the invention or a prodrug of a compound of the invention to the individual in need of treatment.

The utility of the compounds in accordance with the present invention as inhibitors of dipeptidyl peptidase-IV enzyme activity may be demonstrated by methodology known in the art. Inhibition constants are determined as follows. A continuous fluorometric assay is employed with the substrate Gly-Pro-AMC, which is cleaved by DP-IV to release the fluorescent AMC leaving group. The kinetic parameters that describe this reaction are as follows:  $K_m=50 \mu\text{M}$ ;  $k_{cat}=75 \text{ s}^{-1}$ ;  $k_{cat}/K_m=1.5 \times 10^6 \text{ M}^{-1}\text{s}^{-1}$ . A typical reaction contains approximately 50 pM enzyme, 50  $\mu\text{M}$  Gly-Pro-AMC, and buffer (100 mM HEPES, pH 7.5, 0.1 mg/ml BSA) in a total reaction volume of 100  $\mu\text{l}$ . Liberation of AMC is monitored continuously in a 96-well plate fluorometer using an excitation wavelength of 360 nm and an emission wavelength of 460 nm. Under these conditions, approximately 0.8  $\mu\text{M}$  AMC is produced in 30 minutes at 25 degrees C. The enzyme used in these studies was soluble (transmembrane domain and cytoplasmic extension excluded) human protein produced in a baculovirus expression system (Bac-To-Bac, Gibco BRL). The kinetic constants for hydrolysis of Gly-Pro-AMC and GLP-1 were found to be in accord with literature values for the native enzyme. To measure the dissociation constants for compounds, solutions of inhibitor in DMSO were added to reactions containing enzyme and substrate (final DMSO concentration is 1%). All experiments were conducted at room temperature using the standard reaction conditions described above. To determine the dissociation constants ( $K_i$ ), reaction rates were fit by non-linear regression to the Michaelis-Menten equation for competitive inhibition. The

# Explore Litigation Insights

Docket Alarm provides insights to develop a more informed litigation strategy and the peace of mind of knowing you're on top of things.

## Real-Time Litigation Alerts



Keep your litigation team up-to-date with **real-time alerts** and advanced team management tools built for the enterprise, all while greatly reducing PACER spend.

Our comprehensive service means we can handle Federal, State, and Administrative courts across the country.

## Advanced Docket Research



With over 230 million records, Docket Alarm's cloud-native docket research platform finds what other services can't. Coverage includes Federal, State, plus PTAB, TTAB, ITC and NLRB decisions, all in one place.

Identify arguments that have been successful in the past with full text, pinpoint searching. Link to case law cited within any court document via Fastcase.

## Analytics At Your Fingertips



Learn what happened the last time a particular judge, opposing counsel or company faced cases similar to yours.

Advanced out-of-the-box PTAB and TTAB analytics are always at your fingertips.

## API

Docket Alarm offers a powerful API (application programming interface) to developers that want to integrate case filings into their apps.

## LAW FIRMS

Build custom dashboards for your attorneys and clients with live data direct from the court.

Automate many repetitive legal tasks like conflict checks, document management, and marketing.

## FINANCIAL INSTITUTIONS

Litigation and bankruptcy checks for companies and debtors.

## E-DISCOVERY AND LEGAL VENDORS

Sync your system to PACER to automate legal marketing.