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(54) Title: Benzamide Treatment Of Dementia Associated With Aids Virus (HIV-1) Infection

(57) Abstract:

Benzamide-based compositions are disclosed to have activity as therapeutic and prophylactic agents in the treatment of conditions associated with HIV-1 virus infection, referred to in advanced stages as dementia associated with HIV infection or HIV Dementia.

(56) Documents Cited: WO 95/28153 A

BENZAMIDE TREATMENT OF DEMENTIA ASSOCIATED WITH AIDS VIRUS (HIV-1) INFECTION

Field of the Invention

This invention relates to the treatment of dementia associated with AIDS virus (HIV-1) infection. More particularly it concerns compositions and methods for prophylactically or therapeutically treating this condition.

Background Information

This Background Information section is divided into two parts. The first provides information on the condition being treated by this invention, the dementia associated with AIDS virus infection. The second provides information concerning benzamides and their use as medicaments, benzamides being the active agents employed in the methods and compositions of this invention.

HIV Dementia (AIDS Dementia Complex)

Acquired Immune Deficiency syndrome (AIDS) is often accompanied by neurological complications at later states of the disease. Approximately one third of adults and one half of children with AIDS eventually have these complications. These neurological conditions involve a complex set of cognitive, motor and behavioral dysfunctions which have been grouped under the names "AIDS Dementia Complex" (ADC) or more properly "HIV-associated dementia" or "HIV dementia". As many as 50% of infected children have neurological deficits manifested as delayed developmental milestones. Neurological diseases associated with HIV infection include myelopathy, peripheral neuropathy and myopathy. The neuropathological alterations that

accompany HIV infection in the CNS include myelin pallor, increased astrogliosis, neuronal loss, and loss of dendritic arborization as well as a decrease in the presynaptic area. Resulting neurologic dysfunction can impair daily function, work productivity and in severe cases mandate expensive institutional care. Although early losses in mental capacity are not considered full-blown dementia, they nevertheless reflect neuronal damage associated with HIV-1. At present there are no effective therapies for HIV-dementia. The medicaments described herein should minimize the neuronal damage and prevent the progression of neuronal damage thus allowing extended functional capabilities of the affected individuals and hence considerable savings to society.

In the United States alone, over 1 million individuals are infected with HIV and approximately one third of this group have AIDS. Thus, the potential target population for an anti-HIV dementia therapeutic treatment is currently greater than 100,000 patients/year and the target population which would acutely benefit from a prophylactic HIV dementia treatment some ten times that. The need for treatments of HIV dementia is expected to grow as more effective therapies allow persons with AIDS to live longer.

There is no known cure for AIDS available at the present time and in the absence of an effective treatment to completely eliminate the virus from afflicted individuals it is unlikely that any completely effective treatment for HIV dementia is available. Zidovudine (AZT) has been used extensively to treat the AIDS infection. Although there is now doubt as to the long term effectiveness of this treatment because of high mutational frequency of the virus there is no doubt that AZT has been effective in treating HIV dementia on a short-term basis. The neurological symptoms associated with HIV dementia have been treated with certain drugs. For instance, the psychosis associated with HIV dementia has been treated with haloperidol and thioridazine. Molindone has been used for psychotic and delirious HIV dementia patients.

Methylphenidate has been used for treatment of depression associated with HIV dementia. Electro-convulsive therapy has been used for HIV-induced stupor. All of these treatments serve to ameliorate symptoms of HIV dementia. None treat HIV dementia, itself.

The envelope glycoprotein of HIV, gp120, has been implicated in the pathogenesis of HIV dementia. This protein which is shed abundantly by infected cells has been found to be neurotoxic to neurons in culture at extremely low concentrations, to impair learning, to induce cytokines, and to reduce cerebral glucose utilization. Hill et al. (.Hill, J.M., Mervis, R.R., Avidor, R., Moody, T.W. and Brenneman, D.E. (1993) *Brain Res.*, 603:222-233.) have shown that in neonatal rats, administration of gp120 causes morphological damage to the brain as well as retardation of the development of complex motor behaviors.

No approved treatments are available. Use of calcium channel antagonists and NMDA antagonists have been proposed as possible therapies by Lipton. Numerous calcium channel antagonists are available on the market, eg, nimodipine, but NMDA antagonists are still being studied clinically by many companies, primarily for acute use in stroke or chronic use in epilepsy and Parkinson's disease. Amantadine, which is on the market as an anti-viral, is now known to possess NMDA antagonist properties. A closer cogener of amantadine, memantidine, is on the market in Europe and has been proposed by Lipton as a possible candidate for treatment of HIV dementia. Another agent which is available for testing is nitroglycerin. Under certain circumstances, the NO generated from the nitroglycerin can protect neurons from overstimulation of the NMDA receptors with the resulting calcium and glutamate excitotoxicity. However, cardiovascular effects and the extremely erratic pharmacokinetics of nitroglycerin make this approach seem problematic.

In work related to the present invention, together with Robert Floyd, I discovered that certain nitrone compounds exhibited activity as agents against

HIV-dementia. This separate invention is covered in another patent application filed simultaneously herewith.

Benzamides as Medicaments

This invention's approach to mitigating HIV dementia employs a family of benzamide analogues as the active agent. Commonly owned United States patent number 5,472,777 describes certain benzamides and their use in treating neurological conditions. Commonly owned Patent Cooperation Treaty application PCT/US96/04538 describes the compounds employed herein and describe their use as pharmaceutical compositions for conditions not specifically including HIV-dementia.

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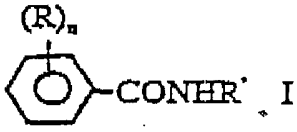
Statement of the Invention

It has now been found that certain benzamide compounds have activity in the treatment of AIDS Dementia Complex (HIV dementia).

This discovery can take the form of benzamide-based pharmaceutical compositions having activity against HIV-dementia. These compositions include one or more of the acetamidobenzamide, aminobenzamide or nitrobenzamide compounds of Formula I as active agent in a pharmaceutically acceptable carrier.



I

In Formula I R' is a saturated alkyl of from 3 to 5 carbon atoms, each R is independently -NH-CO-CH_3, -NO_2 or -NH_2, and n is 1 or 2, with the following provisos: 1) when n is 1 and R is -NO_2 at the 4 position of the ring, R' is not *tert*-butyl, *iso*-butyl, or propyl; 2) when n is 1 and R is -NO_2 at the 2 position of the ring, R' is not *iso*-butyl or propyl; and 3) when n is 2 and R' is *tert*-

butyl and both Rs are -NO_2, the R groups are not at the 3 and 5 positions of the ring. The carrier is preferably an oral carrier but can be an injectable carrier as well. These pharmaceutical compositions can be in bulk form but more typically are presented in unit dosage form.

In another aspect this invention provides a therapeutic method for treating a patient suffering from HIV-dementia. This method involves administering to the patient an effective HIV-dementia-treating amount of one or more of the pharmaceutical compositions just described.

In another aspect this invention provides a prophylactic method for protecting a patient susceptible to HIV-dementia. This method involves administering to the patient an effective HIV-dementia prophylactic amount of one or more of the pharmaceutical compositions just described.

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Brief Description of the Drawings

The invention will be further described with reference being made to the drawings in which

Fig. 1 is a bar graph showing the protective effect of a benzamide in a HIV-dementia related cell culture test.

Fig. 2 is a bar graph showing the protective effect of a benzamide in a HIV-dementia related cell culture test.

Fig. 3 is a bar graph showing apoptosis response observed in a cell aggregation test with a benzamide.

Fig. 4 is a plot of bioavailability of benzamide as a function of time.

Detailed Description of the Invention

The Benzamides

The treatment of this invention employs one or more benzamides as its active agent. This invention employs certain acetamidobenzamides, aminobenzamides and nitrobenzamides as active pharmaceutical agents. The

benzamides are described by Formula I. In this formula, R' is a saturated alkyl of from 3 to 5 carbon atoms and n is 1 or 2.

The acetamido, amino or nitro group (or groups) may be found anywhere on the ring. Preferred embodiments include when n is 1 and the acetamido group is at the 2, 3 or 4 position of the ring and when n is 2 and the acetamido groups are at the 2 and 3, 2 and 4, 2 and 5, 2 and 6, 3 and 4, or 3 and 5 positions of the ring.

With respect to the alkyl substituents, R', compounds wherein R' is an alkyl which does not have a hydrogen on the alpha carbon, that is, the carbon which bonds to the nitrogen of the ring, are preferred. Examples of these preferred R' groups are *tert*-butyl and *tert*-amyl.

Acetamidobenzamides of Formula I of particular interest are:

N-*tert*-butyl-4-acetamidobenzamide,

N-*iso*-propyl-4-acetamidobenzamide,

N-*tert*-amyl-4-acetamidobenzamide,

N-*tert*-butyl-3-acetamidobenzamide, and

N-methylcyclopropyl-4-acetamidobenzamide.

N-*tert*-butyl-4-acetamidobenzamide is the most preferred acetamidobenzamide.

The aminobenzamides and nitrobenzamides employed as active agents are described by Formula I when R is an amino or nitro group. In this formula, R' is a saturated alkyl of from 3 to 5 carbon atoms and n is 1 or 2 subject to the same preferences for substituents and their positions set forth with reference to the acetamidobenzamides and further subject to the provisos that 1) when n is 1 and R is $-NO_2$ at the 4 position of the ring, R' is not *tert*-butyl, *iso*-butyl, or propyl; 2) when n is 1 and R is $-NO_2$ at the 2 position of the ring, R' is not *iso*-butyl or propyl; and 3) when n is 2 and R' is *tert*-butyl and both Rs are $-NO_2$, the R groups are not at the 3 and 5 positions of the ring.

Aminobenzamides and nitrobenzamides of Formula I of particular interest as active agents are:

N-*iso*-propyl-4-nitrobenzamide,

N-*tert*-butyl-3-nitrobenzamide,

N-*tert*-butyl-2-nitrobenzamide,

N-*n*-butyl-4-nitrobenzamide,

N-*n*-propyl-4-nitrobenzamide,

N-*tert*-butyl-3,5-dinitrobenzamide,

N-1-methylpropyl-4-nitrobenzamide,

N-*tert*-butyl-4-aminobenzamide and

N-*tert*-butyl-3-aminobenzamide.

When the benzamide compound contains an amino group, such as is the case with N-*tert*-butyl-3-aminobenzamide and N-*tert*-butyl-4-aminobenzamide, the amine functionality can be present as such or as a salt. In the salt form the amino is protonated to the cation form in combination with a pharmaceutically acceptable anion, such as chloride, bromide, iodide, hydroxyl, nitrate, sulfonate, methane sulfonate, acetate, tartrate, oxalate, succinate, or palmoate. When these aminobenzamides are referred to it is to be understood that these salts are included as well.

Commonly owned United States Patent number 5,472,983, referred to above, discloses several benzamides useful in treating neurodegenerative diseases based on their protective action in the MPTP mouse model of Parkinson's disease. The compound N-*tert*-butyl-4-acetamidobenzamide of the present invention is an *in vivo* biotransformation product of one of these benzamides (N-*tert*-butyl-4-nitrobenzamide) which has been found in the blood of rats and mice to which N-*tert*-butyl-4-nitrobenzamide has been administered orally.

Mixtures of two or more of these materials may be employed, if desired.

Pharmaceutical Compositions

The benzamide compound(s) is formulated into pharmaceutical compositions suitable for oral or parenteral, e.g. intravenous or intramuscular injection administration.

The compositions for oral administration can take the form of liquid solutions or suspensions, powders, tablets, capsules or the like. In such compositions, the nitrone or its salt is usually a minor component (0.1 to say 50% by weight) with the remainder being various vehicles or carriers and processing aids helpful for forming the desired dosing form. A liquid form may include a suitable aqueous or nonaqueous vehicle with buffers, suspending dispensing agents, colorants, flavors and the like.

A solid form may include, for example, any of the following ingredients, or compounds of a similar nature: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a disintegrating agent such as alginic acid, Primogel, or corn starch; a lubricant such as magnesium stearate; a glidant such as colloidal silicon dioxide; a sweetening agent such as sucrose or saccharin; or a flavoring agent such as peppermint, sugar, methyl salicylate, or orange flavoring.

In the case of injectable compositions, they are commonly based upon injectable sterile saline or phosphate-buffered saline or other injectable carriers known in the art. Again the active nitrone is typically a minor component, often being from about 0.05 to 10% by weight with the remainder being the injectable carrier and the like.

These components for orally administrable or injectable compositions are merely representative. Other materials as well as processing techniques and the like are set forth in Part 8 of Remington's Pharmaceutical Sciences, 17th

edition, 1985, Mack Publishing Company, Easton, Pennsylvania, which is incorporated by reference.

One can also administer the compounds of the invention in sustained release forms or from sustained release drug delivery systems. A description of representative sustained release materials can be found in the incorporated materials in Remington's Pharmaceutical Sciences.

Conditions Treated and Treatment Regimens

The conditions treated with the benzamide-containing compositions generally include HIV dementia and the various symptoms which fall within the HIV dementia definition. The benzamide-containing formulations can be administered to achieve a therapeutic effect and slow or counteract the progression of HIV dementia or they can be administered prophylactically to patients not yet exhibiting HIV dementia but exposed to the HIV-1 virus. The benzamide-containing composition is administered in manners designed to get the drug into the patient's bloodstream and across the blood-brain barrier into the patient's brain. One excellent mode for accomplishing this is intravenous administration. Intravenous dose levels for treating these conditions range from about 0.01 mg/kg/hour to about 10 mg/kg/hour, all for from about 1 to about 120 hours and especially 1 to 96 hours. A preloading bolus of from about 10 to about 500 mg may also be administered to achieve adequate steady state levels. Other forms of parenteral administration, such as intramuscular injection can be used, as well. In this case, similar dose levels are employed.

While parenteral administration is attractive from a drug delivery point of view, it should be recognized that the course of HIV infection can stretch over many months or even years so oral dosing may be preferred for patient convenience and tolerance. With oral dosing, one to three oral doses per day, each from about 0.02 to about 50 mg/kg are called for with preferred doses

being from about 0.04 to about 10 mg/kg. These same dosing levels and regimens would be used for prophylactic treatment as well.

In any treatment regimen, the health care professional should assess the patient's condition and determine whether or not the patient would benefit from benzamide treatment. Some degree of experimentation to determine an optimal doing level and pattern may be called for.

A positive dose-response relationship has been observed. As such and bearing in mind the severity of the side effects and the advantages of providing maximum possible protection or amelioration, it may be desired in some settings to administer large amounts of benzamide such as those described above.

Methods of Preparation of Compounds

The benzamide compounds employed herein can be prepared using commonly available starting materials and readily achievable reactions.

One representative preparation route, which is illustrated with *tert*-butyl amine, but which may be used with any alkyl amine, involves the following reactions:

(A)

III

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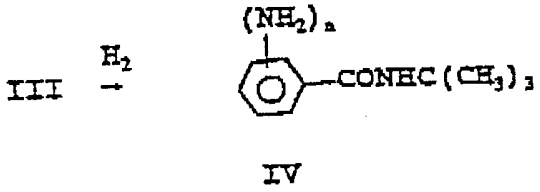
where X is halo such as I, Br, F or Cl.

(B)



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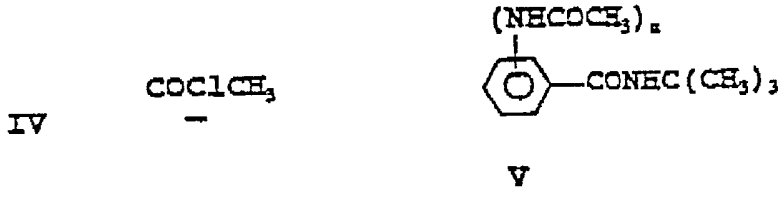

In step (A) the N-*tert*-butyl nitrobenzamides (III) are formed. This reaction should be carried out at temperatures below 10°C.

This step (A) yields as benzamides III, the compounds of the invention where R is $-NO_2$.

In step (B) the nitro groups in the mono- or di-nitro benzamide III are subjected to reduction. This is commonly carried out with a reducing agent such as hydrazine and an appropriate catalyst such as a heterogeneous platinum, iron oxide hydroxide, palladium or nickel catalyst, typically on a support, or with hydrogen gas and a catalyst.

This step (B) yields as benzamides IV, the compounds of the invention where R is NH_2.

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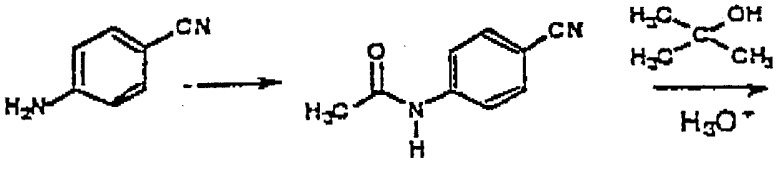
In step (C) the amino-benzamides IV are converted to acetamidobenzamides V by reaction with an acetyl halide such as acetylchloride. This reaction is carried out in the presence of a mild base and at low to ambient temperatures such as - 20°C to + 20°C. This yields the compounds of the invention where R is acetamido.

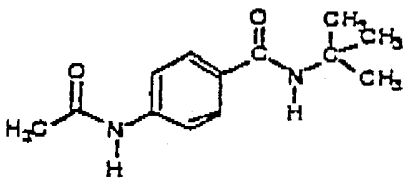
Alternate synthetic schemes may also be used to prepare the compounds. Examples of these alternate routes are set forth below using N-*tert*-butyl-4-acetamidobenzamide as the representative compound. Other compounds may be prepared using these alternate methods by starting with appropriate starting materials, such as 2- or 3- amino- or nitro-benzonitrile or 2,3-, 2,4-, 2,5-, 2,6-, 3,4- or 3,5- diamino- or dinitro-benzonitrile and the appropriate alcohol (Alternate Route 1) or similarly substituted toluene compounds and the appropriate alkyl amine (Alternate Route 3).

Alternate Route 1

This route begins with acetylation of, for example, 4-aminobenzonitrile (A) to compound (B) using standard methods. Acid hydrolysis of *tert*-butanol in the presence of 4-acetamidobenzonitrile (B), provides a feasible synthetic pathway to N-*tert*-butyl-4-acetamidobenzamide.

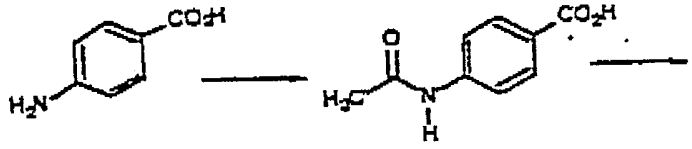


A B

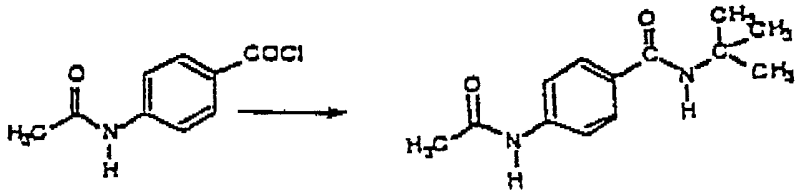


Alternate Route 2

Acetylation, using standard methods, of the inexpensive starting material PABA (C) affords a cheap method to produce 4-acetamidobenzoic acid (D). Conversion of (D) to the acid chloride (E) using standard methods (e.g., $SOCl_2$) and subsequent amidation using standard methods, such as those described previously, produces N-*tert*-butyl-4-acetamidobenzamide from inexpensive raw materials.



C D



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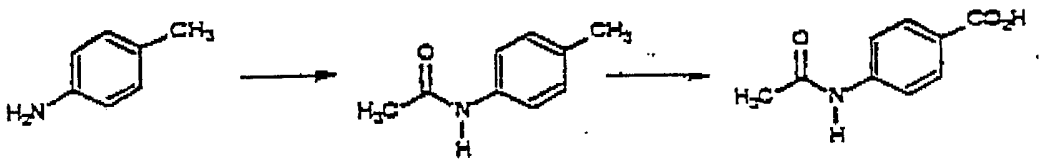
Alternate Route 3

Another method for the preparation of the compounds begins with acetylation, using standard methods, of, for example, paratoluidine (F) to 4-acetamidotoluene (G). The synthetic intermediate (G) may be converted to 4-acetamidobenzoic acid (D) with common oxidizing agents (e.g., $KMnO_4$) and

subsequently transformed to N-*tert*-butyl-4-acetamidobenzamide as outlined in Alternate Route 2.



F G D

Examples

The invention will be further described by the following Examples. These are provided to illustrate several preferred embodiments of the invention but are not to be construed as limiting its scope which is, instead, defined by the appended claims. Examples 1 to 19 demonstrate the preparation of acetamidobenzamides, as well as nitro- and aminobenzamides, which are representative of the benzamide compounds employed in the compositions and methods of this invention. Examples 20 to 24 demonstrate the preparation of pharmaceutical compositions based on the compounds. Thereafter biological test results illustrating the activity of the compositions of the invention are provided.

Example 1

Preparation of N-*tert*-butyl-4-aminobenzamide

tert-Butyl amine (14.6 g, 0.200 mole) was stirred in ethyl acetate (150 mL, purified by washing with 5% sodium carbonate solution, saturated sodium chloride solution, drying over anhydrous magnesium sulfate, and filtering through fluted filter paper) and cooled to 5° C with an ice bath. 4-nitrobenzoyl chloride (18.6 g, 0.100 mole) in purified ethyl acetate (75 mL)

was added dropwise at such a rate to maintain the temperature below 10° C. The ice bath was removed upon complete addition of benzoyl chloride solution and the reaction stirred for 4 hours. The reaction mixture was then filtered on a Büchner funnel, the filtrate washed three times with 5% HCl, once with saturated sodium chloride, dried over anhydrous magnesium sulfate, filtered through fluted filter paper, and the solvent stripped off leaving white crystalline product. The product was dried in a vacuum oven at 24 mm and 45° C for 14 hours. This procedure produced 17.13 g of crystals of N-*tert*-butyl-4-nitrobenzamide (77% yield), mp 162-163° C. Proton nuclear magnetic resonance (89.55 MHz in $CDCl_3$) showed absorptions at 8.257 ppm (d, 8.8 Hz, 2H; 3,5-aryl H); 7.878 ppm (d, 8.8 Hz, 2H; 2,6-aryl H); 6.097 ppm (bs, 1H; N-H); 1.500 ppm (s, 9H; *tert*-butyl H).

Palladium on carbon (5%, 75 mg) was added to N-*tert*-butyl-4-nitrobenzamide (5 g, 22.5 mmole) in 95% ethanol at 55°C. A solution of hydrazine (1.2 mL) in 95% ethanol (10 mL) was added dropwise over 30 min. and more Pd/C added (75 mg). The reaction was refluxed 3 hours, hydrazine (0.5 g) in 95% ethanol (5 mL) was added and the reaction was refluxed for another hour. The reaction was filtered on a buchner funnel, the volume of solvent reduced under vacuum, and extracted with dichloromethane. The combined extracts were dried over magnesium sulfate and solvent stripped, leaving 3.90 g of N-*tert*-butyl-4-aminobenzamide (90% yield), melting point 125 - 127 °C. 90 MHz proton NMR (in $CDCl_3$) showed absorbances at 7.290 ppm (2H, d, 8.8 Hz; 2,6 aryl H); 6.368 ppm (2H, d, 8.8 Hz; 3,5 aryl H); 5.45 ppm (1 H, bs; NHC=O); 3.727 ppm (2H, bs; aryl-NH_2); 1.186 ppm (9 H, s; t-butyl H).

Example 2

Preparation of N-*tert*-butyl-4-acetamidobenzamide

Acetyl chloride (0.45 g, 5.7 mmole) in ethyl acetate (25 mL) was added dropwise to N-*tert*-butyl-4-aminobenzamide (1.0 g, 5.2 mmole) and triethyl amine (0.58 g, 5.7 mmole) in ethyl acetate at 3° C at such a rate to maintain the temperature below 10° C. The reaction was allowed to warm to room temperature, stirred 1 hour, and washed with 5% HCl. Recrystallization from acetone gave 1.08 g N-*tert*-butyl-4-acetamidobenzamide (89% yield), melting point 119 - 121 °C. 90 MHz proton NMR (in DMSO-d6) showed absorbances at 9.726 ppm (1H, bs, N-H); 7.715 ppm (4H, dd, 4.4 Hz; aryl H); 7.295 ppm (1 H, bs; NH); 2.844 ppm (3H, s; CH_3CO); 1.448 ppm (9 H, s; t-butyl H).

Example 3

Preparation of N-*tert*-butyl-3-nitrobenzamide

N-*tert*-butyl-3-aminobenzamide and

N-*tert*-butyl-3-acetamidobenzamide

The amidation procedures of Example 1 were followed using 3-nitrobenzoyl chloride instead of 4-nitrobenzoyl chloride. This gave N-*tert*-butyl-3-nitrobenzamide in 92% yield, melting point 123-125 °C. Proton NMR (in $CDCl_3$) showed absorptions at 8.517 ppm (2-aryl H, s, 1H); 8.337 ppm (4-aryl H, d, 8.8 Hz, 1H); 8.121 ppm (6-aryl H, d, 6.4 Hz, 1H); 7.618 ppm (5-aryl H, m, 1H); 6.032 ppm (N-H, bs, 1H); 1.484 ppm (t-butyl H, s, 9 H).

Iron (III) oxide hydroxide catalyzed hydrazine reduction produced N-*tert*-butyl-3-aminobenzamide in 53% yield, melting point 118-120 °C. Proton NMR (in $CDCl_3$) showed absorbances at 7.088 ppm (4-6 -aryl H, m, 3 H); 6.794 ppm (2-aryl H, s, 1H); 5.902 ppm (N-H, bs, 1H); 3.145 ppm (aryl N-H, bs, 2H); 1.458 ppm (t-butyl H, s, 9 H).

Acetylation of N-*tert*-butyl-3-aminobenzamide as described in Example 2 gave N-*tert*-butyl-3-acetamidobenzamide in 75% yield, melting point 194-

195°C. Proton NMR (in $CDCl_3$) showed absorptions at 7.778 ppm (4-6 -aryl H, m, 3 H); 7.392 ppm (2-aryl H, s, 1H); 6.08 ppm (N-H, bs, 1H); 2.174 ppm (acetyl CH_3, s, 9 H); 1.500 ppm (t-butyl H, s, 9 H).

Example 4

Preparation ofN-*tert*-butyl-2-nitrobenzamide and N-*tert*-butyl-2-acetamidobenzamide

The method of Example 3 is repeated using 2-nitrobenzoyl chloride in the amidation step. This yields N-*tert*-butyl-2-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-butyl-2-aminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-butyl-2-acetamidobenzamide.

Example 5

Preparation ofN-*iso*-propyl-4-nitrobenzamide and N-*iso*-propyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and *iso*-propyl amine in the amidation step. This yields N-*iso*-propyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*iso*-propyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-*iso*-propyl-4-acetamidobenzamide.

Example 6

Preparation ofN-*tert*-amyl-4-nitrobenzamide and N-*tert*-amyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and *tert*-amyl amine in the amidation step. This yields N-*tert*-amyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-amyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-amyl-4-acetamidobenzamide.

Example 7

Preparation of N-*iso*-butyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and *iso*-butyl amine in the amidation step. This yields N-*iso*-butyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*iso*-butyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-*iso*-butyl-4-acetamidobenzamide.

Example 8

Preparation of N-*n*-butyl-4-nitrobenzamide and N-*n*-butyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and *n*-butyl amine in the amidation step. This yields N-*n*-butyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*n*-butyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-*n*-butyl-4-acetamidobenzamide.

Example 9

Preparation of N-*n*-propyl-4-nitrobenzamide and N-*n*-propyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and *n*-propyl amine in the amidation step. This yields N-*n*-propyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*n*-propyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-*n*-propyl-4-acetamidobenzamide.

Example 10

Preparation of N-1,2-dimethylpropyl-4-nitrobenzamide and N-1,2-dimethylpropyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and 1,2-dimethylpropyl amine in the amidation step. This yields N-1,2-dimethylpropyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-1,2-dimethylpropyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-1,2-dimethylpropyl-4-acetamidobenzamide.

Example 11

Preparation of N-*n*-pentyl-4-nitrobenzamide and N-*n*-pentyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and *n*-pentyl amine in the amidation step. This yields N-*n*-pentyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*n*-pentyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-*n*-pentyl-4-acetamidobenzamide.

Example 12

Preparation of N-2-methylbutyl-4-nitrobenzamide and N-2-methylbutyl-4-acetamidobenzamide

The method of Example 3 is repeated using 4-nitrobenzoyl chloride and 2-methylbutyl amine in the amidation step. This yields N-2-methylbutyl-4-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-2-methylbutyl-4-aminobenzamide.

Acetylation of the aminobenzamide yields N-2-methylbutyl-4-acetamidobenzamide.

Example 13

Preparation of N-*n*-pentyl-2-nitrobenzamide and N-*n*-pentyl-2-acetamidobenzamide

The method of Example 3 is repeated using 2-nitrobenzoyl chloride and *n*-pentyl amine in the amidation step. This yields N-*n*-pentyl-2-nitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*n*-pentyl-2-aminobenzamide.

Acetylation of the aminobenzamide yields N-*n*-pentyl-2-acetamidobenzamide.

Example 14

Preparation of N-*tert*-butyl-2,3-diacetamidobenzamide

The method of Example 3 is repeated using 2,3-dinitrobenzoyl chloride in the amidation step. This yields N-*tert*-butyl-2,3-dinitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-butyl-2,3-diaminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-butyl-2,3-diacetamidobenzamide.

Example 15

Preparation of N-*tert*-amyl-2,4-diacetamidobenzamide

The method of Example 3 is repeated using 2,4-dinitrobenzoyl chloride and *tert*-amyl amine in the amidation step. This yields N-*tert*-amyl-2,4-dinitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-amyl-2,4-diaminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-amyl-2,4-diacetamidobenzamide.

Example 16

Preparation of N-*tert*-butyl-2,5-diacetamidobenzamide

The method of Example 3 is repeated using 2,5-dinitrobenzoyl chloride in the amidation step. This yields N-*tert*-butyl-2,5-dinitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-butyl-2,5-diaminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-butyl-2,5-diacetamidobenzamide.

Example 17

Preparation of N-*tert*-butyl-2,6-diacetamidobenzamide

The method of Example 3 is repeated using 2,6-dinitrobenzoyl chloride in the amidation step. This yields N-*tert*-butyl-2,6-dinitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-butyl-2,6-diaminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-butyl-2,6-diacetamidobenzamide.

Example 18

Preparation of N-*tert*-butyl-3,4-diacetamidobenzamide

The method of Example 3 is repeated using 3,4-dinitrobenzoyl chloride in the amidation step. This yields N-*tert*-butyl-3,4-dinitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-butyl-3,4-diaminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-butyl-3,4-diacetamidobenzamide.

Example 19

Preparation of N-*tert*-butyl-3,5-diacetamidobenzamide

The method of Example 3 is repeated using 3,5-dinitrobenzoyl chloride in the amidation step. This yields N-*tert*-butyl-3,5-dinitrobenzamide.

Reduction of the nitrobenzamide with hydrazine yields N-*tert*-butyl-3,5-diaminobenzamide.

Acetylation of the aminobenzamide yields N-*tert*-butyl-3,5-diacetamidobenzamide.

Preparation of Pharmaceutical Compositions

Example 20

The compound of Example 1 is admixed as a dry powder with a dry gelatin binder in an approximate 1:2 weight ratio. A minor amount of magnesium stearate is added as a lubricant. The mixture is formed into 240-270 mg tablets (80-90 mg of active benzamide) in a tablet press. If these

tablets were administered to a patient suffering from HIV dementia on a daily, twice daily or thrice daily regimen they would slow the progress of the patient's disease.

Example 21

The compound of Example 2 is admixed as a dry powder with a starch diluent in an approximate 1:1 weight ratio. The mixture is filled into 250 mg capsules (125 mg of active benzamide). If these capsules were administered to a patient susceptible to coming down with HIV dementia on a daily, twice daily or thrice daily regimen they would slow or prevent the onset of the HIV dementia.

Example 22

The compound of Example 3 is suspended in a sweetened flavored aqueous medium to a concentration of approximately 50 mg/mL. If 5 mLs of this liquid material was administered to a patient suffering from HIV dementia on a daily, twice daily or thrice daily regimen they would slow the progress of the patient's disease.

Example 23

The compound of Example 4 is admixed as a dry powder with a dry gelatin binder in an approximate 1:2 weight ratio. A minor amount of magnesium stearate is added as a lubricant. The mixture is formed into 450-900 mg tablets (150-300 mg of active benzamide) in a tablet press. If these tablets were administered to a patient suffering from HIV dementia on a daily, twice daily or thrice daily regimen they would slow the progress of the patient's disease.

Example 24

The compound of Example 14 is dissolved in a buffered sterile saline injectable aqueous medium to a concentration of approximately 5 mg/ml. If 50 mLs of this liquid material was administered to a patient suffering from HIV dementia on a daily, twice daily or thrice daily regimen this dose would slow the progress of the patient's disease.

It will be appreciated that any of the compounds of Formula I could be employed in any of these representative formulations, and that any of these formulations could be administered in any of these manners so as to treat any of the HIV dementia conditions described in this specification.

Biological Testing

These tests utilized two neural cell culture systems for determining the efficacy of N-*tert*-butyl-4-acetamidobenzamide ("Compound I") in reversing neurotoxicity which mimic that observed with HIV dementia. In both assays, human neural cell cultures were used either as a bilayer (neurons on an astrocyte layer) or a three dimensional model (brain cell aggregates). TNF-∝ (100 pg/ml) was used as the neurotoxin and the length of incubation was 72 hours. A considerable body of evidence supports the notion that TNF-∝ is one of the neurotoxins responsible for HIV dementia. Brain concentrations of TNF-∝ are elevated in deep grey matter from AIDS patients with mild HIV dementia. Achim, C, Heyes, MP, Wiley, CA (1993) Quantitation of human immunodefficiency virus, immune activation facyors, and quinolinic acid in AIDS brains, *J Clin Invest* 91: 2769-2775. The distribution of messenger RNA expressing TNF-∝ in the brain follows a similar pattern. Wesselingh, SL, Power, C, Glass, JD, Tyor, WR et al. (1993) Intracereberal cytokine messenger RNA expression in aquired immuniodeficiency syndrome dementia, *Annals of Neurology*, 33: 576-582. Gelbard et al. have shown that HIV-1 infected monocytes in culture with astroglial cells produce concentrations

(>200 pg/ml) of TNF-∝ sufficient to cause neurotoxicity. Gelbard, HA, Dzenko, KA, DiLoreto, D, delCero, C, delCerro, M, Epstein, LG (1994) Neurotoxic effects of tumor necrosis factor alpha in primary human neuronal cultures are mediated by activation of the glutamate AMPA receptor subtype: Implications for AIDS neuropathogenesis, *Dev Neurosci*, 15: 417-422. TNF-∝ is reported to cause its neurotoxicity by inducing apoptosis. Selmaj, K, Raine, CS, Farocq, M, Norton, WT, Brosnan, CF (1991) Cytokine cytotoxicity against oligodendrocytes. Apoptosis induced by Lymphotoxins, *J Immunol*, 147: 1522-1529. Recently, it was shown that gp120 exerts toxic effects through induction of IL-6 and TNF-∝. Yeung, MC, Pulliam, L., Lau, AS (1995) The HIV envelope protein gp120 is toxic to human brain-cell cultures through the induction of interleukin-6 and tumor necrosis factor-α, *AIDS*, 9:137-143.

Brain Aggregate Procedure

Brain cell aggregates were prepared from second trimester abortion tissue as previously described. Pulliam L., Berens, ME, Rosenblum, ML 1988. A normal human brain cell aggregate model for neurobiological studies, *J Neurosci Res* 21 :521-530. Briefly, human brain tissue between 16 and 18 weeks gestation are gently dissociated through nylon screens to obtain single cells. Approximately 4 X 10^7 cells within 4 ml DME supplemented with 0.6% dextrose, 50 mg/ml gentamicin and 10% FCS are distributed into 25 ml DeLong flasks. Aggregates are constantly rotated and incubated at 37°C in an atmosphere of 10% CO2. After 2-3 days, aggregates are transferred to 50 ml flasks and 5 ml of DME supplemented with 15% FCS (exchange medium) added. Each flask contains several thousand aggregates that can be sampled over time. Five ml of medium is exchanged every other day in culture. After 10-12 days in culture samples are taken for histology and trypan blue exclusion is performed to determine viability. Samples are screened for HIV, Hepatitis A, B, C and mycoplasma. Aggregates remain viable for approximately 40 days

in culture. Brain cell aggregates are differentiated at the time of sampling in that they express neural cell markers for identification. Brain cell aggregates contain all the cells of the CNS- approximately 40% neurons, 40% astrocytes, 10% oligodendrocytes with myelin and 10% microglia. Neural cell apoptosis/death was measured by DNA fragmentation Elisa technique according to manufactures directions (Boehringher Mannheim).

Neural Cell Bilayer Procedure

Brain aggregates were prepared as described above. Several aggregates are placed in each well of a multi-well chamber slide (Nunc) coated with Cell TAK (Collaborative Research) at a concentration of 20 ug/ml. Cells migrate from the brain aggregates within 3 days. Astrocytes form a monolayer with neurons on top and rare microglia (<1%)/oligodendrocytes (<1%). These cultures are confluent within 1 week. Monolayers can be maintained for up to three weeks. Characterization of cell types is determined by using immunohistochemistry and the antibodies neuron specific enolase (NSE, Dako) for neurons and glial fibrillary acidic protein (GFAP, Dako) for the identification of astrocytes. Confocal microscopy was used to visualize and identify neurons and astrocytes by size and shape. Neuronal viability was determined by exposing chambers with and without different treatments to AO and ethidium bromide (EtBr). Neurons and total cell counts were determined by AO staining with visual confirmation by phase microscopy. Enumeration of cell viability by computerized software was performed at the time of microscopy; in addition, a visual printout of the fields observed always accompanied the data.

Experimental Design

Experiment#	System	TNF-∝ (pg/ml)	Compound[1] (μM)
1	Neural Cell Bilayers	0	0
		0	100
		100	0
		100	100
2	Neural Cell Bilayers	0	0
		100	0
		100	100
3	Brain Aggregate	0	0
		0	100
		100	0
		100	100

[1] Test compound is N-*tert*-butyl-4-acetamidobenzamide.

Results

Experiment 1 (Figure 1): This was a human neural cell bilayer experiment. N-*tert*-butyl-4-acetamidobenzamide ("Compound I") showed some toxicity relative to the control. The TNF-∝ treatment produced a high degree of cell death, over 61%. N-*tert*-butyl-4-acetamidobenzamide treatment produced substantial protection.

Experiment 2 (Figure 2): This experiment was a repeat of experiment 1 using a different brain preparation. Results essentially duplicated those from the first experiment, except the TNF-∝ treatment gave less neuronal toxicity.

Experiment 3 (Figure 3): This experiment utilized human brain aggregates. In this experiment, apoptosis/cell death was measured by an immunoassay for quantitation of cytoplasmic histone-associated DNA fragments. In this experiment, N-*tert*-butyl-4-acetamidobenzamide treatments gave substantial protection both with and without the TNF-∝ treatments. The bars in Figure 4 represent the mean of duplicate experiments. The error bars in this figure express the individual values.

Physical/Chemical Parameters

N-*tert*-butyl-4-acetamidobenzamide was studied to determine physical/chemical properties which suggest its suitability for this application. The following results were obtained:

	N-*tert*-butyl-4-acetamidobenzamide
$t_{1/2}$(min) in Aqueous HCl Solution (pH1)	3000
Octanol-Water Partition	31

This shows that N-*tert*-butyl-4-acetamidobenzamide is lipophilic and slowly cleared from the body. N-*tert*-butyl-4-acetamidobenzamide is a compound of particular interest for HIV dementia because, at least in the rat, it shows excellent brain distribution, bioavailability and pharmacokinetic profile. N-*tert*-butyl-4-acetamidobenzamide is also significantly stable at a pH commonly observed in the stomach.

Brain penetration of N-*tert*-butyl-4-acetamidobenzamide

Following a 30 mg/kg oral dose, blood and brain samples from the same animals were analyzed for N-*tert*-butyl-4-acetamidobenzamide at 4 and 8 hours post-dose with the following results:

Time Post-Dose (hours)	Mean Brain Concentration (μg/g) +/- SEM	Mean Blood Concentration (μg/ml) +/- SEM
4	8.9 +/- 3.2	43 +/- 7.9
8	9.1 +/- 1.7	39 +/- 4.2

Absolute Bioavailability of N-*tert*-butyl-4-acetamidobenzamide Oral Suspension

The absolute bioavailability of N-*tert*-butyl-4-acetamidobenzamide in rats was determined by comparing the area under the curve following a 20 mg/kg dose of the benzamide dissolved in 1% methyl cellulose. Blood concentrations were determined at either 0, 0.083, 0.15, 0.5, 1, 2, 4, 8 and 24 hours post-dose (IV) or 0, 0.5, 1, 2, 4 and 8 hour post-dose (oral), and the AUCs determined. Four animals were dosed orally and 4 animals were dosed IV.

Route	Mean AUC +/- SEM ($\mu g\ hr\ ml^{-1}$)	Absolute Bioavailability
IV	252 +/- 73	—
Oral	130 +/- 33	52%

The pharmacokinetic profile of a 30 mg/kg dose to Sprague Dawley rats can be found in Figure 4. The apparent $t_{1/2}$ for N-*tert*-butyl-4-acetamidobenzamide in this experiment was 8 hours, a very long $t_{1/2}$ for a drug in rat- a good predictor of once-a-day dosing if N-*tert*-butyl-4-acetamidobenzamide would ever be dosed in man. Such a dosing regimen would be a significant therapeutic advantage in the clinic.

Further Brain Aggregation Studies

Further studies were conducted as follows:

Experiment #	gp120(ng/ml)	TNF-α(ng/ml)	Compound[1] (μM)
4	-	0	0
	-	1	0.1
	-	1	0.3
	-	1	3.0
5	0	-	0
	1	-	3

[1] Test compound is N-*tert*-butyl-4-acetamidobenzamide.

As shown in the following Table, Experiment 4 showed that at unexpectedly low concentrations, N-*tert*-butyl-4-acetamidobenzamide provided complete protection in human brain aggregates from DNA fragmentation, a measure of apoptosis, induced by 1 ng TNF-α. Some degree of dose proportionality was found. The results at all test compound concentrations are statistically significant at $p < 0.05$ by Student t-test from the TNF only group, but, of the compound treated groups, only the TNF ± 0.3 μM test compound group is statistically significant from the other two treatment groups.

Table

Experiment 4 Results

Experiment	DNA Fragmentation (Absorbance ± SF.n=3)	% Protection
Control	0.663 ± 0.048	–
TNF Only	1.592 ± 0.156	–
TNF + 0.1 μM Compound[1]	0.955 ± 0.101	78
TNF + 0.3 μM Compound[1]	0.835 ± 0.051	87
TNF + 3.0 μM Compound[1]	0.801 ± 0.123	90

1 Test compound is N-*tert*-butyl-4-acetamidobenzamide.

The data above suggests that protection from apoptosis can be achieved at concentrations of approximately 1 μM and below.

A 1μM concentration of N-*tert*-butyl-4-acetamidobenzamide is in the order of 0.2 μg/ml. To achieve this concentration in rat brain would require a blood concentration of only 1 μg/ml based on the brain/blood ratio data presented previously. If some degree of dose proportionality is found with lower doses of N-*tert*-butyl-4-acetamidobenzamide, a 6 mg/kg dose to rats should achieve this concentration even at 24 hours post-dose (trough value). Using liver blood flow differences to scale the clearance of drug in rats to that in man as described in Pulliam, L, Herndier, B, McGrath, MS (1991) Purified trichosanthin (GLQ223®)) exacerbation of indirect HIV-associated neurotoxicity in vitro, *AIDS*, 5: 1237-1242, a dose of 1.5 mg/kg to man would be predicted to achieve at 24 hours post-dose the 1 μM target concentration in the brain for protection from apoptosis.

Consistent with the results above, N-*tert*-butyl-4-acetamidobenzamide also provided complete protection in human brain aggregates from toxicity induced by 1 ng TNF-α, although the concentration of the benzamide needed was considerably higher than that found to prevent DNA fragmentation. These data are as follows:

Experiment	LDH Release Absorbance ± SD (n=)	% Protection
Control	0.875 ± 0.022	–
TNF	1.071 ± 0.036	–
TNF + 0.1 μM Compound[1]	1.114 ± 0.023	0
TNF + 0.3 μM Compound[1]	1.103 ± 0.034	0
TNF + 3.0 μM Compound[1]	0.864 ± 0.028	100

[1] Test compound is N-*tert*-butyl-4-acetamidobenzamide.

Experiment 5:

In this experiment, N-*tert*-butyl-4-acetamidobenzamide provided significant protection in human brain aggregates from cell toxicity induced by 1 ng gp120. The difference in absorbance was statistically significant for all groups at $p < 0.003$.

Experiment	LDH Release Absorbance ± SD(n=)	% Protection
Control	0.328 ± 0.011	–
gp120	0.575 ± 0.008	–
gp120 + 3.0 μM Compound[1]	0.427 ± 0.034	60%

1 Test compound is N-*tert*-butyl-4-acetamidobenzamide.

There was no evidence in this experiment for DNA fragmentation induced at this concentration of gp120.

Experiment 6:

Using procedures essentially the same as those described above for determining LDH release induced by TNF, programmed cell death (PCD) analysis was performed by ELISA using standardized kits (Boehriger Mannheim). The results were as follows:

Experiment 6A	PCD
Control	0 ± 0.359
TNF-α	1.18 ± 0.759
TNF-α + 10.0 μM Compound[1]	1.15 ± 0.125
TNF-α + 10.0 μM Compound[2]	1.021 ± 0.099
TNF-α + 10.0 μM Compound[3]	0.34 ± 0.029

1 Test compound is N-*tert*-butyl-4-acetamidobenzamide.

2 Test compound is N-*tert*-butyl-4-aminobenzamide.

3 Test compound is N-*tert*-amyl-4-acetamidobenzamide.

Experiment 6B	PCD
Control	0 ± 0.69
TNF-α	1.16 ± 0.088
TNF-α + 10.0 μM Compound[1]	1.05 ± 0.043
TNF-α + 10.0 μM Compound[2]	0.567 ± 0.026
TNF-α + 10.0 μM Compound[3]	0.671 ± 0.043

1 Test compound is N-*tert*-butyl-4-acetamidobenzamide.

2 Test compound is N-*tert*-butyl-4-aminobenzamide.

3 Test compound is N-*tert*-amyl-4-acetamidobenzamide.

Experiment 6C	PCD
Control	0 ± 0.032
TNF-α	0.674 ± 0.058
TNF-α + 10.0 μM Compound[4]	0.565 ± 0.042

4 Test compound is N-isopropyl-4-acetamidobenzamide.

Experiment 6D	PCD
Control	0 ± 0.018
TNF-α	0.531 ± 0.034
TNF-α + 10.0 μM Compound[4]	0.016 ± 0.03

4 Test compound is N-isopropyl-4-acetamidobenzamide.

The data from Experiments 6A-D demonstrate that various benzamides of this invention provided protection in human brain aggregates from toxicity induced by 1 ng TNF-α as measured by PCD analysis.

In vivo Tests

In order to determine the effectiveness of this approach for treating ADC, a series of in vivo biological tests were carried out.

In vivo Test A

Material and Methods Used

Sodium N-methyl D-glucamine dithiocarbamate (MGD) and the nitrone, PBN, were obtained from OMRF Spin Trap Source, Oklahoma City, Oklahoma. gp120 was obtained from Intracel Corporation, Cambridge, Massachusetts. These materials were used in the following preliminary test:

Treatment of Animals: Sprague-Dawley neonatal rats (sixteen siblings) were divided into four groups. Starting at day one after birth until day six, the neonates received 60 μl subcutaneous injections of the following treatments. Group 1: phosphate buffer-saline (PBS), Group 2: 5 ng gp120 in PBS, Group 3: 5 ng gp120 plus PBN (50 mg/kg) in PBS, and Group 4: PBN (50 mg/kg) in PBS. Rats were weighed daily and the amount of PBN injected was adjusted accordingly.

Behavioral Assessments: Time required to perform two developmental milestones were measured to determine the adverse effects of gp120 administration on behavioral development as reported by Hill et al. and to determine the possible protective action of PBN on these parameters. Behavioral parameters studied were surface righting (animal placed head down on 45° inclined screen will turn around and climb up.) These two tests have been shown to be the most sensitive tests for assessment of the neurological disorder caused by gp120 treatment. Furthermore, they can be examined early enough in the life of the animal (day 3 for surface righting and day 6 for negative geotaxis) that their determination will not interfere with NO trapping in the brain which we performed at the end of the first week of the life of the animal. Animals were tested for the time required for surface righting on day 3

and day 4 after birth, immediately prior to receiving the injections on those days, and on day 6 (2 hrs after the last injection that the animals received) as well as day 7 (20 hrs following the last injections) for the time required to perform negative geotaxis. The angle chosen for the setting used for negative geotaxis was decreased from 45° (the angle used by Hill et al) to 35° since under the experimental setting employed, animals were not able to stay on the screen set at 45° and would slide down before being able to make an attempt to turn upward.

In vivo Test B

Protection by N-*tert*-butyl-4-acetamidobenzamide from gp120-induced behavioral changes

The striking results obtained with PBN prompted preliminary experiments with N-*tert*-butyl-4-acetamidobenzamide in the same model. The results are suggestive that N-*tert*-butyl-4-acetamidobenzamide is effective as demonstrated by the data shown below obtained on neonates that had been administered gp120 at 10 ng per dose starting on 3 day old animals. N-*tert*-butyl-4-acetamidobenzamide was given at an oral dose of 35 mg/kg 2 hours prior to administering the gp120. Treatment with N-*tert*-butyl-4-acetamidobenzamide and gp120 continued daily. The negative geotaxis test was conducted on day 6.

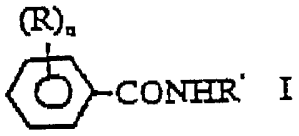
	Negative Geotaxis (sec)
Treatment	3 h Post-Last Dose gp120 (Day 6)
Vehicle	8.89 ± 3.74
gp120	18.0 ± 13.8
gp120 + Compound[1]	8.39 ± 3.94
Compound[1]	8.56 ± 5.11

[1] Test compound is N-*tert*-butyl-4-acetamidobenzamide.

The data suggests N-*tert*-butyl-4-acetamidobenzamide had a protective effect.

WHAT IS CLAIMED IS:

1. A pharmaceutical composition for treating HIV dementia comprising a benzamide compound of the formula:



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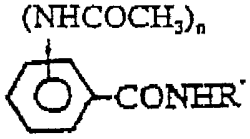
wherein R' is a saturated alkyl of from 3 to 5 carbon atoms, each R is independently $-NO_2$ or $-NH_2$ or $NHCOCH_3$, and n is 1 or 2, with the following provisos: 1) when n is 1 and R is $-NO_2$ at the 4 position of the ring, R' is not *tert*-butyl, *iso*-butyl, or propyl; 2) when n is 1 and R is $-NO_2$ at the 2 position of the ring, R' is not *iso*-butyl or propyl; and 3) when n is 2 and R' is *tert*-butyl and both Rs are $-NO_2$, the R groups are not at the 3 and 5 positions of the ring,

in a pharmaceutically acceptable carrier.

2. The pharmaceutical composition of Claim 1 wherein the benzamide compound is an acetamidobenzamide of the formula:



where R' is a saturated alkyl of from 3 to 5 carbon atoms and n is 1 or 2.

3. The pharmaceutical composition of Claim 2 wherein n is 1.

4. The pharmaceutical composition of Claim 3 wherein R' is *tert*-butyl.

5. The pharmaceutical composition of Claim 3 wherein R' is *tert*-amyl.

6. The pharmaceutical composition of Claim 3 wherein the benzamide compound is N-*tert*-butyl-4-acetamidobenzamide.

7. The pharmaceutical composition of Claim 1 wherein the carrier is an oral carrier.

8. The pharmaceutical composition of Claim 1 wherein the carrier is an injectable carrier.

9. A method for treating HIV dementia comprising administering to a patient in need of such treating an effective HIV dementia complex-treating amount of a composition of Claims 1-8.

10. The method of Claim 9 wherein the administering is oral.

11. The method of Claim 9 wherein the administering is parenteral.

12. The method of Claim 11 wherein the administering is by injection.

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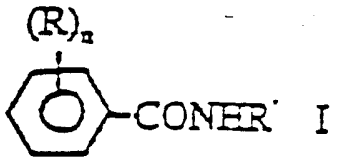
13. The method of Claim 9 wherein the treating is therapeutic.

14. The method of Claim 9 wherein the treating is prophylactic.

15. The use of a benzamide compound of the formula:



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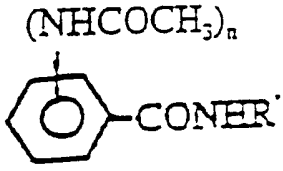
wherein R' is a saturated alkyl of from 3 to 5 carbon atoms, each R is independently $-NO_2$ or $-NH_2$ or $NHCOCH_3$, and n is 1 or 2, with the following provisos: 1) when n is 1 and R is $-NO_2$ at the 4 position of the ring, R' is not *tert*-butyl, *iso*-butyl, or propyl; 2) when n is 1 and R is $-NO_2$ at the 2 position of the ring, R' is not *iso*-butyl or propyl; and 3) when n is 2 and R' is *tert*-butyl and both Rs are $-NO_2$, the R groups are not at the 3 and 5 positions of the ring, in the manufacture of a pharmaceutical composition for the treatment of HIV dementia.

16. The use of Claim 15 wherein the benzamide compound is an acetamidobenzamide of the formula:



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where R' is a saturated alkyl of from 3 to 5 carbon atoms and n is 1 or 2.

17. The use of Claim 16 wherein n is 1.

18. The use of Claim 17 wherein R' is *tert*-butyl.

19. The use of Claim 17 wherein R' is *tert*-amyl.

20. The use of Claim 17 wherein the benzamide compound is N-*tert*-butyl-4-acetamidobenzamide.

FISHER, CORMACK & BOTHA
Patent Agents for the Applicant
Dated this 21st day of October 1998

Sgd: D. Gilson

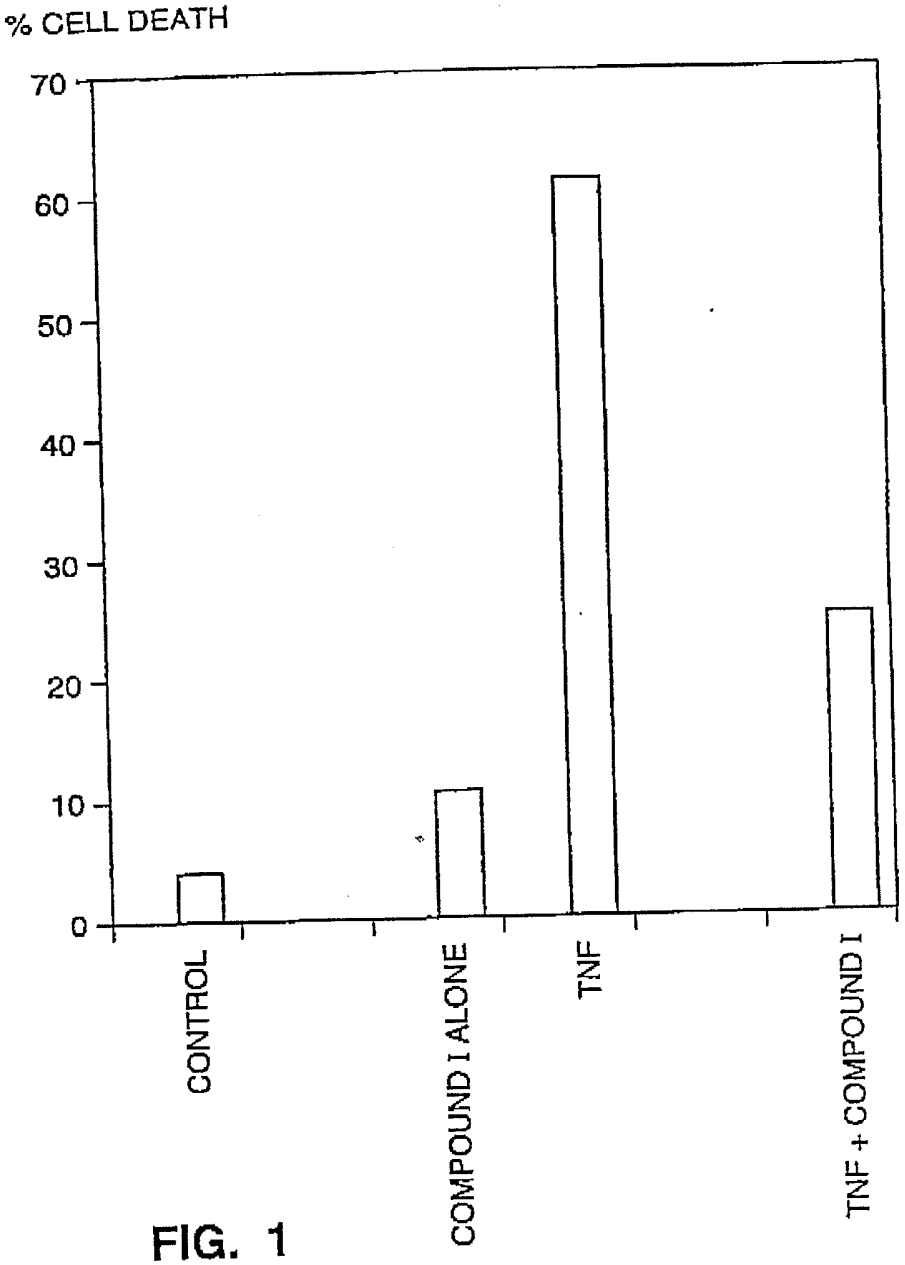

FIG. 1

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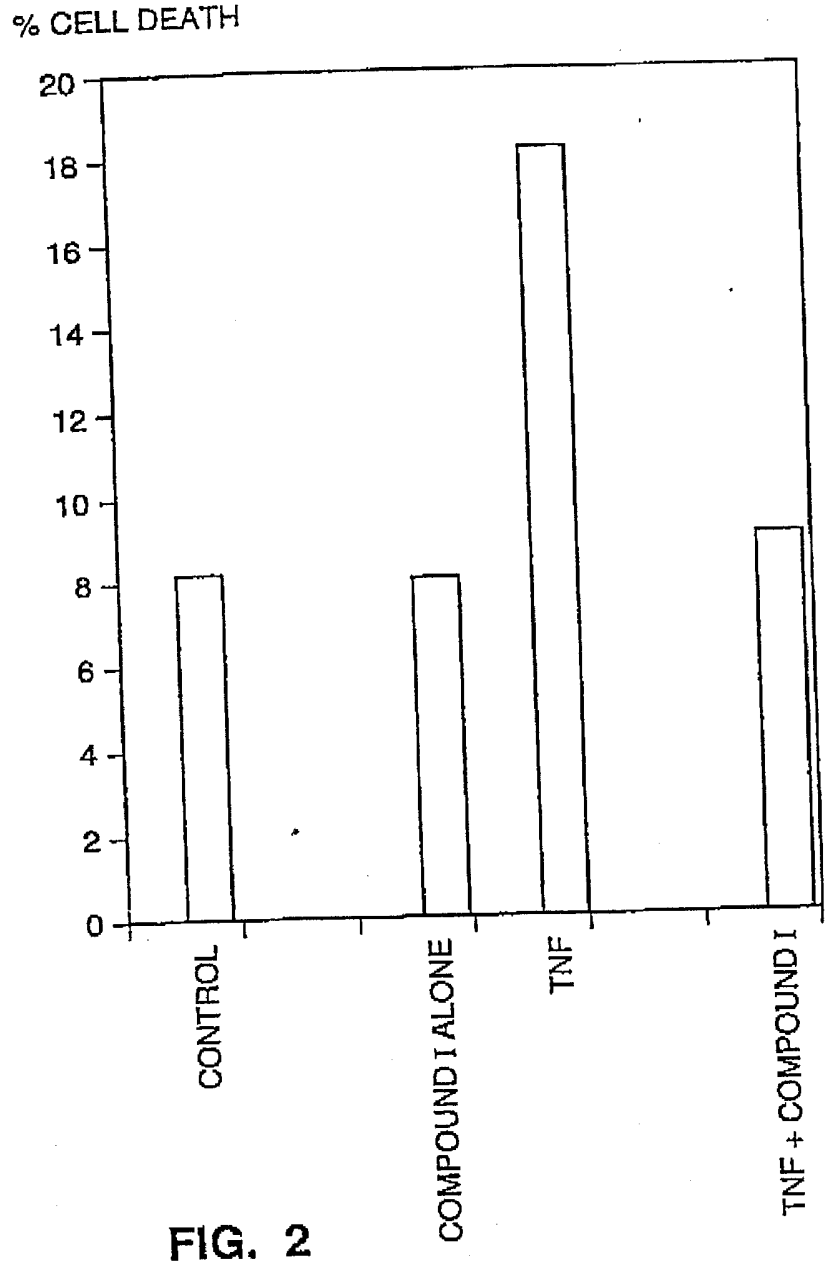

FIG. 2

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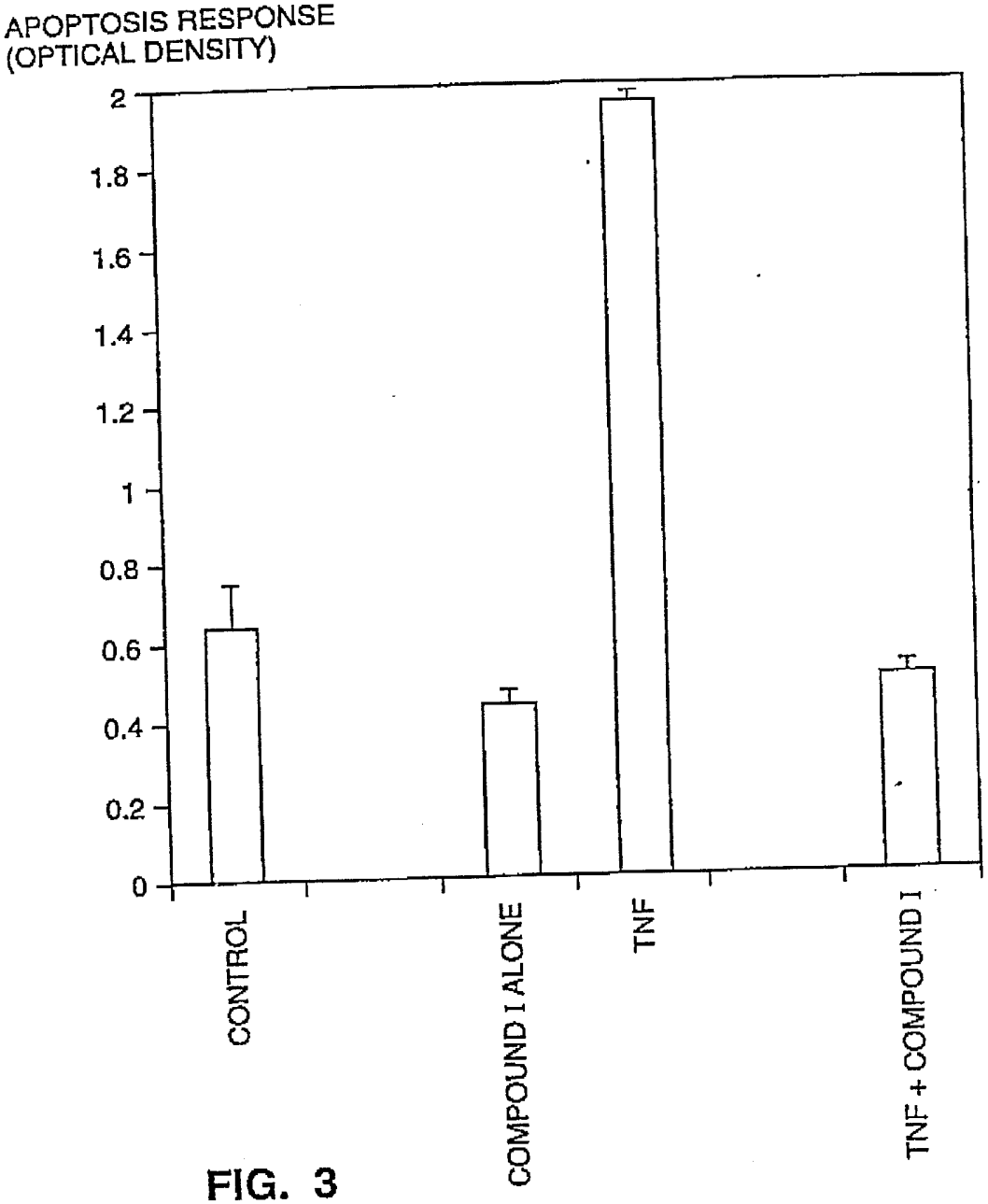

FIG. 3

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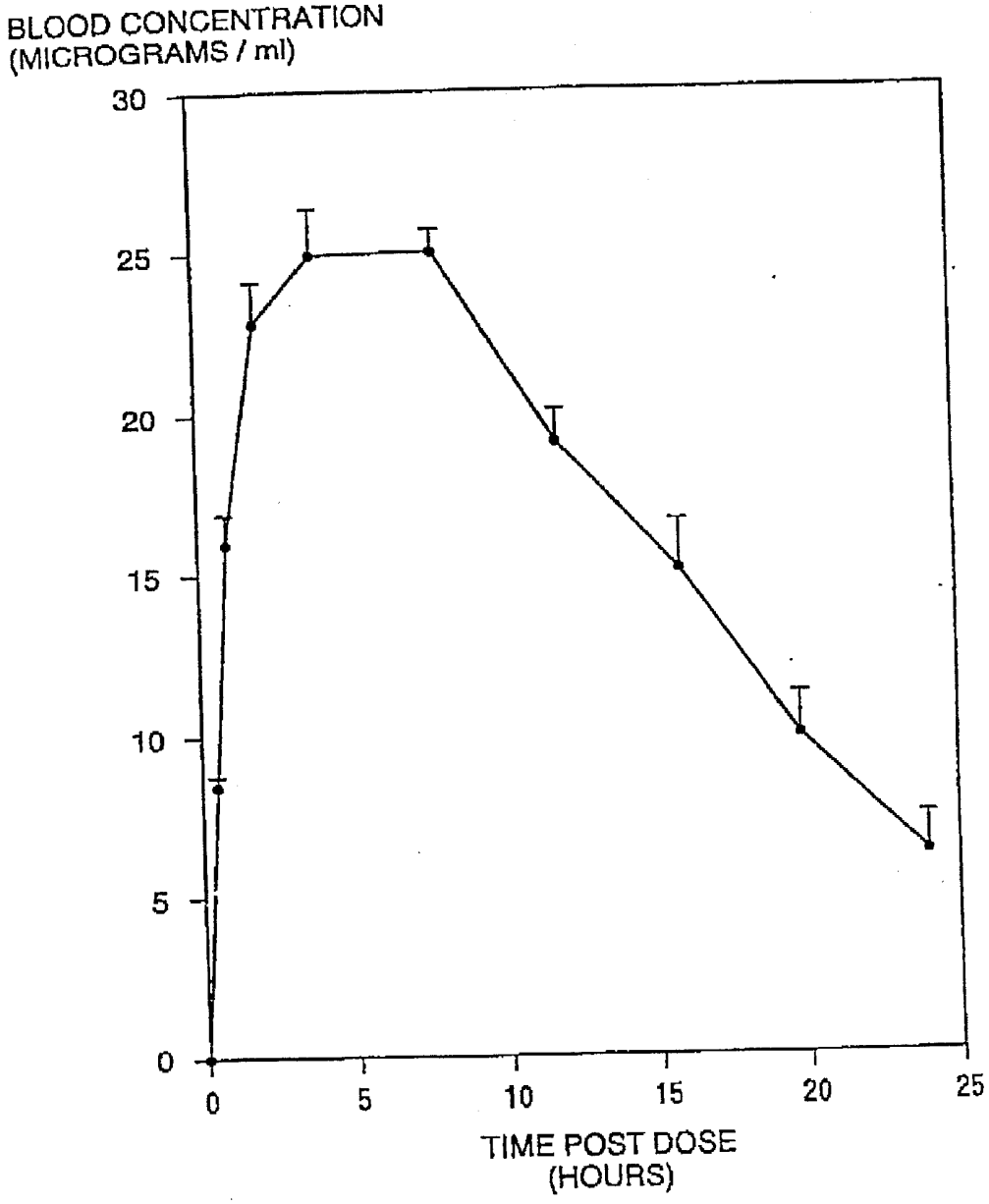

FIG. 4

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