5-(2-IMIDAZOLINYLAMINO)-BENZIMIDAZOLE DERIVATIVES, THEIR PREPARATION AND THEIR USE AS .ALPHA.-ADRENOCEPTOR AGONISTS WITH IMPROVED METABOLIC STABILITY

TECHNICAL FIELD

The subject invention is directed to certain substituted benzimidazole compounds that have improved resistance to metabolism in primates. The subject compounds are alpha adrenoceptor agonists and are useful in treating alpha agonist associated disorders.

BACKGROUND OF THE INVENTION

Alpha adrenergic receptors, agonists, antagonists, and compounds related in structure to those of this invention are disclosed in the following references: Timmermans, P.B.M.W.M., A. T. Chiu & M.J.M.C. Thoolen, "12.1 α-Adrenergic Receptors", Comprehensive Medicinal Chemistry, Vol. 3, Membranes & Receptors, P. G. Sammes & J. B. Taylor, eds., Pergamon Press (1990), pp. 133-185; Timmermans, P.B.M.W.M. & P.A. van Zwieten, "α-Adrenoceptor Agonists and Antagonists", Drugs of the Future, Vol. 9, No. 1, (January, 1984), pp. 41-55; Megens, A.A.H.P., J. E. Leysen, F.H.L. Awouters & C.J.E. Niemegeers, "Further Validation of *in vivo* and *in vitro* Pharmacological Procedures for Assessing the α_1 and α_2-Selectivity of Test Compounds: (2) α-Adrenoceptor Agonists", European Journal of Pharmacology, Vol. 129 (1986), pp. 57-64; Timmermans, P.B.M.W.M., A. de Jonge, M.J.M.C. Thoolen, B. Wilffert, H. Batink & P.A. van Zwieten, "Quantitative Relationships between α-Adrenergic Activity and Binding Affinity of α-Adrenoceptor Agonists and Antagonists", Journal of Medicinal Chemistry, Vol. 27 (1984) pp. 495-503; van Meel, J.C.A., A. de Jonge, P.B.M.W.M. Timmermans & P. A. van Zwieten, "Selectivity of Some Alpha Adrenoceptor Agonists for Peripheral Alpha-1 and Alpha-2 Adrenoceptors in the Normotensive Rat", The Journal of Pharmacology and Experimental Therapeutics, Vol. 219, No. 3 (1981), pp. 760-767; Chapleo, C.B., J.C. Doxey, P.L. Myers, M. Myers, C.F.C. Smith & M. R. Stillings, "Effect of 1,4-Dioxanyl Substitution on the Adrenergic Activity of Some Standard α-Adrenoreceptor Agents", European Journal of Medicinal Chemistry, Vol. 24 (1989), pp. 619-622; Chapleo, C.B., R.C.M. Butler, D.C. England, P.L. Myers, A.G. Roach, C.F.C. Smith, M.R. Stillings & I:F. Tulloch, "Heteroaromatic Analogues of the α_2-Adrenoreceptor Partial Agonist Clonidine", J. Med. Chem., Vol. 32 (1989), pp. 1627-1630; Clare, K.A., M.C. Scrutton & N.T. Thompson,

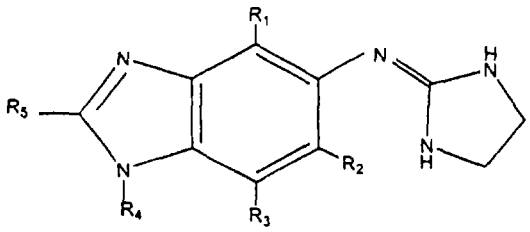
"Effects of α_2-Adrenoceptor Agonists and of Related Compounds on Aggregation of, and on Adenylate Cyclase Activity in, Human Platelets", Br. J. Pharmac., Vol. 82 (1984), pp. 467-476; U.S. Patent No. 3,890,319 issued to Danielewicz, Snarey & Thomas on June 17, 1975; and U.S. Patent No. 5,091,528 issued to Gluchowski on February 25, 1992.

Alpha-2 adrenergic agonists are useful for treating a variety of disorders including: respiratory disorders (e.g., asthma, nasal congestion, COPD, cough, cystic fibrosis), gastrointestinal disorders (e.g., diahrrea, irritable bowel syndrome), ocular disorders (e.g., glaucoma), cardiovascular disorders (e.g., myocardial ischemia, shock, arrhythmias, angina, congestive heart failure), benign prostatic hypertrophy and migraine. However, many compounds disclosed in the art and related in structure to those of this invention are not alpha-2 adrenoceptor selective (e.g., they interact with other alpha receptors such as alpha-1 adrenoceptors). Alpha-2 adrenoceptor selectivity is desirable when treating alpha-2 associated or alpha-2 mediated disorders. For example, alpha-2 adrenergic agonists that possess significant alpha-1 adrenergic effects are known to cause cardiovascular side effects such as hypertension. In addition, many compounds disclosed in the art and related in structure to those of this invention possess significant central nervous system (CNS) activity which may lead to undesirable side effects such as severe sedation.

It has also been observed that some alpha adrenergic agonists are subject to extensive metabolic transformation in primates. Such metabolic transformation results in inactivation of the parent compound or in the formation of an active metabolite with a different pharmacological profile from the parent compound. Of particular importance to the present invention is the metabolic transformation that occurs to some alpha adrenergic benzimidazoles that are peripherally acting alpha-2-adrenoceptor selective agonists. Metabolic N-methylation at the benzimidazole ring may result in compounds that (1) are inactive; (2) are alpha-2 adrenoceptor antagonists; (3) possess enhanced activity at other undesired receptors, such as at alpha-1 adrenoceptors; and/or (4) have an increased potential for CNS activity. Thus, there is a continuing need for peripherally acting selective alpha-2 adrenergic compounds that have lower CNS activity and that resist metabolic transformation into undesirable compounds.

SUMMARY OF THE INVENTION

The present invention is directed to compounds having a structure according to the following formula:



wherein:

(a) R1 is alkyl;

(b) R2 is selected from the group consisting of: hydrogen, alkyl, methoxy, cyano, and halo;

(c) R3 is selected from the group consisting of: hydrogen, methyl, hydroxy, cyano and halo;

(d) R4 is selected from the group consisting of: hydrogen, methyl, ethyl and isopropyl;

(e) R5 is selected from the group consisting of: hydrogen, methyl, amino, methoxy, hydroxy, cyano and halo;

(f) provided that at least one of R2, R3, R4 or R5 is other than hydrogen or fluorine;

(g) provided that when R1 is methyl and both R2 and R5 are hydrogen, R3 is other than methyl or halo;

(h) provided that when R3 is cyano, R1 is methyl; and

any tautomer of the above structure or a pharmaceutically acceptable salt, or biohydrolyzable ester, amide, or imide thereof.

The compounds of the present invention are useful in treating many medical disorders, including for example, respiratory disorders, ocular disorders, gastrointestinal disorders, disorders associated with sympathetic nervous system activity, migraine, peripheral pain, and disorders where vasoconstriction would provide a benefit. Accordingly, the invention further provides pharmaceutical compositions comprising these compounds. The invention still further provides methods of treatment using these compounds or the compositions containing them

DETAILED DESCRIPTION OF THE INVENTION

Terms and Definitions

"Alkyl" is an unsubstituted saturated or unsaturated hydrocarbon chain having 1 to 3 carbon atoms. Alkyl chains may be straight, branched or cyclized. Preferred alkyl groups are methyl, ethyl, and cyclopropyl.

"Biohydrolyzable amide" refers to an amide of a compound of the invention that is readily converted *in vivo* by a subject to yield an active compound of the invention.

"Biohydrolyzable ester" refers to an ester of a compound of the invention that is readily converted by a subject to yield an active compound of the invention.

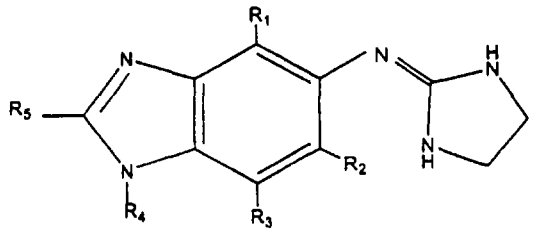
"Halo", "halogen", or "halide" is a chloro, bromo, fluoro or iodo. Preferred halo are chloro, bromo, and iodo. More preferred halo are chloro and bromo.

"Pharmaceutically-acceptable salt" is a cationic salt formed at any acidic (e.g., carboxyl) group, or an anionic salt formed at any basic (e.g., amino) group. Many such salts are known in the art, as described in World Patent Publication 87/05297, Johnston et al., published September 11, 1987, incorporated by reference herein. Preferred cationic salts include the alkali metal salts (such as sodium and potassium), alkaline earth metal salts (such as magnesium and calcium) and organic salts. Preferred anionic salts include halides, sulfonates, carboxylates, phosphates, and the like. Clearly contemplated in such salts are addition salts that may provide an optical center, where once there was none. For example, a chiral tartrate salt may be prepared from the compounds of the invention, and this definition includes such chiral salts.

"Primate" includes humans.

Compounds

The present invention involves compounds having the following structure:



In the above structure, R1 is alkyl. Preferred R1 is methyl, ethyl or cyclopropyl.

In the above structure, R2 is hydrogen, alkyl, methoxy, cyano, or halo. Preferred R2 is hydrogen, alkyl, or cyano. More preferred R2 is methyl or halo.

In the above structure, R3 is hydrogen, methyl, hydroxy, cyano or halo. Preferred R3 is cyano or hydroxy when R1 is methyl. Most preferred R3 is cyano when R1 is methyl. Preferred R3 is methyl or halo when R1 is other than methyl.

In the above structure, R4 is hydrogen, methyl, ethyl or isopropyl. Preferred R4 is hydrogen or methyl, more preferably hydrogen.

In the above structure, R5 is hydrogen, methyl, amino, methoxy, hydroxy, cyano or halo. Preferred R5 is hydrogen, methyl, or halo.

In the above structure, at least one of R2, R3, R4, and R5 is other than hydrogen or fluorine. In addition, when R1 is methyl and both R2 and R5 are hydrogen, then R3 is other than methyl or halo. Finally, when R3 is cyano, R1 is methyl.

The invention includes tautomers of the above structure. For example, when tautomer D of a molecule is shown (see below), it is understood to include tautomer E. Thus, the disclosure of one tautomeric form discloses each and all of the tautomers.



D E

The invention also includes pharmaceutically acceptable acid addition salts, biohydrolyzable esters, amides, and imides of the above structure.

The compounds of the invention are sufficiently basic to form acid-addition salts. The compounds are useful both in the free base form and the form of acid-addition salts, and both forms are within the purview of the invention. The acid-addition salts are in some cases a more convenient form for use. In practice, the use of the salt form inherently amounts to the use of the base form of the active. Acids used to prepare acid-addition salts include preferably those which produce, when combined with the free base, medicinally acceptable salts. These salts have anions that are relatively innocuous to the animal organism, such as a mammal, in medicinal doses of the salts so that the beneficial property inherent in the free base are not vitiated by any side effects ascribable to the acid's anions.

Examples of appropriate acid-addition salts include, but at not limited to hydrochloride, hydrobromide, hydroiodide, sulfate, hydrogensulfate, acetate,

trifluoroacetate, nitrate, maleate, citrate, fumarate, formate, stearate, succinate, mallate, malonate, adipate, glutarate, lactate, propionate, butyrate, tartrate, methanesulfonate, trifluoromethanesulfonate, p-toluenesulfonate, dodecyl sulfate, cyclohexanesulfamate, and the like. However, other appropriate medicinally acceptable salts within the scope of the invention are those derived from other mineral acids and organic acids. The acid-addition salts of the basic compounds are prepared by several methods. For example the free base can be dissolved in an aqueous alcohol solution containing the appropriate acid and the salt is isolated by evaporation of the solution. Alternatively, they may be prepared by reacting the free base with an acid in an organic solvent so that the salt separates directly. Where separation of the salt is difficult, it can be precipitated with a second organic solvent, or can be obtained by concentration of the solution.

Although medicinally acceptable salts of the basic compounds are preferred, all acid-addition salts are within the scope of the present invention. All acid-addition salts are useful as sources of the free base form, even if the particular salt per se is desired only as an intermediate product. For example, when the salt is formed only for purposes of purification or identification, or when it is used as an intermediate in preparing a medicinally acceptable salt by ion exchange procedures, these salts are clearly contemplated to be a part of this invention.

The compounds of the invention are useful for the treatment of a variety of diseases, disorders, and conditions that are modulated by alpha-2 adrenoceptors or by alpha-2 adrenoceptor activity. As used herein, the terms "disease," "disorder" and "condition" are used interchangeably. As used herein, a disorder described by the terms "modulated by alpha-2 adrenoceptors," or "modulated by alpha-2 adrenoceptor activity" refers to a disorder, condition or disease where alpha-2 adrenoceptor activity is an effective means of alleviating the disorder or one or more of the biological manifestations of the disease or disorder; or interferes with one or more points in the biological cascade either leading to the disorder or responsible for the underlying disorder; or alleviates one or more symptoms of the disorder. Thus, disorders subject to "modulation" include those for which:

- The lack of alpha-2 activity is a "cause" of the disorder or one or more of the biological manifestations, whether the activity was altered genetically, by infection, by irritation, by internal stimulus or by some other cause;
- The disease or disorder or the observable manifestation or manifestations of the disease or disorder are alleviated by alpha-2 activity. The lack of alpha-2 activity need not be causally related to the disease or disorder or the observable manifestations thereof;

- Alpha-2 activity interferes with part of the biochemical or cellular cascade that results in or relates to the disease or disorder. In this respect, the alpha-2 activity alters the cascade, and thus controls the disease, condition or disorder.

The compounds of the invention are peripherally-selective alpha-2 adrenoceptor agonists. Alpha-2 adrenoceptors are distributed both inside and outside of the central nervous system. Thus, for example, a compound which displays a higher degree of central nervous system activity is preferred, but not limited to, use in central nervous system indications such as certain cardiovascular disorders (e.g., hypertension), pain, substance abuse and/or withdrawal. By centrally acting it is meant that they have some action on the alpha-2 adrenoceptors in the central nervous system in addition to their action at peripheral alpha-2 adrenoceptors.

Peripherally-acting compounds are preferred for, but not limited to, the treatment of respiratory disorders, ocular disorders, migraine, certain cardiovascular disorders, and certain gastrointestinal disorders. By peripherally acting, it is meant is that these compounds do not readily cross the blood-brain barrier and thus act primarily on alpha-2 adrenoceptors in the periphery. In addition, further specificity of action of these compounds can be achieved by delivering the agent to the region where activity is desired (for example, topical administration to the eye, nasal mucosa or respiratory tract), thereby reducing systemic exposure. Such peripherally-selective compounds have reduced CNS side effect potentials, particularly with respect to sedation. Methods are available in the art to determine which compounds are less centrally-acting than others.

The compounds of the subject invention have no or only weak alpha-1 agonist activity and have little or no effect on the central nervous system, even when dosed systemically.

Thus, the compounds of the invention are particularly useful for the treatment of respiratory disorders including nasal congestion associated with allergies, colds, and other nasal disorders, (as well as the sequelae of congestion of the mucous membranes, for example, sinusitis and otitis media), cough, chronic obstructive pulmonary disease and asthma. At effective doses, it has been found that undesired side effects can be avoided.

The compounds of the invention are also useful for the treatment of ocular disorders such as ocular hypertension, glaucoma, hyperemia, conjunctivitis, and uveitis.

The compounds of the invention are also useful for controlling gastrointestinal disorders, such as diarrhea, irritable bowel syndrome, hyperchlorhydria and peptic ulcer.

The compounds of the invention are also useful for diseases and disorders associated with sympathetic nervous system activity, including hypertension, myocardial ischemia, cardiac reperfusion injury, angina, cardiac arrhythmia, heart failure and benign prostatic hypertrophy.

The compounds of the invention are also useful for the prophylactic or acute treatment of migraine.

The compounds of the invention are also useful for the treatment of peripheral pain states associated with various disorders (for example, peripheral neuralgia).

The compounds of the invention are also useful for other diseases and disorders where vasoconstriction, particularly of veins, would provide a benefit, including septic or cardiogenic shock, elevated intracranial pressure, hemmorhoids, venous insufficiency, varicose veins, and menopausal flushing.

The pharmacological activity and selectivity of these compounds can be determined using published test procedures. The alpha-2 selectivity of the compounds is determined by measuring receptor binding affinities and *in vitro* functional potencies in a variety of tissues known to possess alpha-2 and/or alpha-1 receptors. (See, e.g., The Alpha-2 Adrenergic Receptors, L.E. Limbird, ed., Humana Press, Clifton, NJ.) The following *in vivo* assays are typically conducted in rodents or other species. Central nervous system activity is determined by measuring locomotor activity as an index of sedation. (See, e.g., Spyraki, C. & H. Fibiger, "Clonidine-induced Sedation in Rats: Evidence for Mediation by Postsynaptic Alpha-2 Adrenoreceptors", Journal of Neural Transmission, Vol. 54 (1982), pp. 153-163). Nasal decongestant activity is measured using rhinomanometry as an estimate of nasal airway resistance. (See, e.g., Salem, S. & E. Clemente, "A New Experimental Method for Evaluating Drugs in the Nasal Cavity", Archives of Otolaryngology, Vol. 96 (1972), pp. 524-529). Antiglaucoma activity is determined by measuring intraocular pressure. (See, e.g., Potter, D., "Adrenergic Pharmacology of Aqueous Human Dynamics", Pharmacological Reviews, Vol. 13 (1981), pp. 133-153). Antidiarrheal activity is determined by measuring the ability of the compounds to inhibit prostaglandin-induced diarrhea. (See, e.g., Thollander, M., P. Hellstrom & T. Svensson, "Suppression of Castor Oil-Induced Diarrhea by Alpha-2 Adrenoceptor Agonists", Alimentary Pharmacology and Therapeutics, Vol. 5 (1991), pp. 255-262). Efficacy in treating irritable bowel

syndrome is determined by measuring the ability of compounds to reduce the stress-induced increase in fecal output. (See, e.g., Barone, F., J. Deegan, W. Price, P. Fowler, J. Fondacaro & H. Ormsbee III, "Cold-restraint stress increases rat fecal pellet output and colonic transit", American Journal of Physiology, Vol. 258 (1990), pp. G329-G337). Antiulcer and reduction of hyperchlorhydria efficacy is determined by measuring the reduction in gastric acid secretion produced by these compounds (See, e.g., Tazi-Saad, K., J. Chariot, M. Del Tacca & C. Roze, "Effect of α2-adrenoceptor agonists on gastric pepsin and acid secretion in the rat", British Journal of Pharmacology, Vol. 106 (1992), pp. 790-796). Antiasthma activity is determined by measuring the effect of the compound on bronchoconstriction associated with pulmonary challenges such as inhaled antigens. (See, e.g., Chang, J. J. Musser & J. Hand, "Effects of a Novel Leukotriene D_4 Antagonist with 5-Lipoxygenase and Cyclooxygenase Inhibitory Activity, Wy-45,911, on Leukotriene-D_4- and Antigen-Induced Bronchoconstriction in Guinea Pig", International Archives of Allergy and Applied Immunology, Vol. 86 (1988), pp. 48-54; and Delehunt, J., A. Perruchound, L. Yerger, B. Marchette, J. Stevenson & W. Abraham, "The Role of Slow-Reacting Substance of Anaphylaxis in the Late Bronchial Response After Antigen Challenge in Allergic Sheep", American Reviews of Respiratory Disease, Vol. 130 (1984), pp. 748-754). Activity in cough is determined by measuring the number and latency of the cough response to respiratory challenges such as inhaled citric acid. (See, e.g., Callaway, J. & R. King, "Effects of Inhaled α2-Adrenoceptor and $GABA_B$ Receptor Agonists on Citric Acid-Induced Cough and Tidal Volume Changes in Guinea Pigs", European Journal of Pharmacology, Vol. 220 (1992), pp. 187-195). The sympatholytic activity of these compounds is determined by measuring the reduction of plasma catecholamines (See, e.g., R. Urban, B. Szabo & K. Starke "Involvement of peripheral presynaptic inhibition in the reduction of sympathetic tone by moxonidine, rilmenidine and UK 14,304", European Journal of Pharmacology, Vol. 282 (1995), pp. 29-37) or the reduction in renal sympathetic nerve activity (See, e.g., Feng, Q., S. Carlsson, P. Thoren & T. Hedner, "Effects of clonidine on renal sympathetic nerve activity, natriuresis and diuresis in chronic congestive heart failure rats", Journal of Pharmacology and Experimental Therapeutics, Vol. 261 (1992), pp. 1129-1135), providing the basis for their benefit in heart failure and benign prostatic hypertrophy. The hypotensive effect of these compounds is measured directly as a reduction in mean blood pressure (See, e.g., Timmermans, P. & P. Van Zwieten, "Central and peripheral α-adrenergic effects of some imidazolidines", European Journal of Pharmacology, Vol. 45 (1977), pp. 229-236).

Clinical studies have demonstrated the beneficial effect of alpha-2 agonists in the prevention of myocardial ischemia during surgery (See, e.g., Talke, P., J. Li, U. Jain, J. Leung, K. Drasner, M. Hollenberg & D. Mangano, "Effects of Perioperative Dexmedetomidine Infusion in Patients Undergoing Vascular Surgery", Anesthesiology, Vol. 82 (1995), pp. 620-633) and in the prevention of angina (See, e.g., Wright, R.A., P. Decroly, T. Kharkevitch & M. Oliver, "Exercise Tolerance in Angina is Improved by Mivazerol--an α2-Adrenoceptor Agonist", Cardiovascular Drugs and Therapy, Vol. 7 (1993), pp. 929-934). The efficacy of these compounds in cardiac reperfusion injury is demonstrated by measuring the reduction of cardiac necrosis and neutrophil infiltration (See, e.g., Weyrich, A., X. Ma, & A. Lefer, "The Role of L-Arginine in Ameliorating Reperfusion Injury After Myocardial Ischemia in the Cat", Circulation, Vol. 86 (1992), pp. 279-288). The cardiac antiarrhythmic effect of these compounds is demonstrated by measuring the inhibition of ouabain induced arrhythmias (See, e.g., Thomas, G. & P. Stephen, "Effects of Two Imidazolines (ST-91 and ST-93) on the Cardiac Arrhythmias and Lethality Induced by Ouabain in Guinea-Pig", Asia-Pacific Journal of Pharmacology, Vol. 8 (1993), pp.109-113; and Samson, R., J. Cai, E. Shibata, J. Martins & H. Lee, "Electrophysiological effects of α2-adrenergic stimulation in canine cardiac Purkinje fibers", American Journal of Physiology, Vol. 268 (1995), pp. H2024-H2035). The vasoconstrictor activity of these compounds is demonstrated by measuring the contractile properties on isolated arteries and veins *in vitro* (See, e.g., Flavahan, N., T. Rimele, J. Cooke & M. Vanhoutte, "Characterization of Postjunctional Alpha-1 and Alpha-2 Adrenoceptors Activated by Exogenous or Nerve-Released Norepinephrine in the Canine Saphenous Vein", Journal of Pharmacology and Experimental Therapeutics, Vol. 230 (1984), pp. 699-705). The effectiveness of these compounds at reducing intracranial pressure is demonstrated by measurement of this property in a canine model of subarachnoid hemorrhage (See, e.g., McCormick, J., P. McCormick, J. Zabramski & R. Spetzler, "Intracranial pressure reduction by a central alpha-2 adrenoreceptor agonist after subarachnoid hemorrhage", Neurosurgery, Vol. 32 (1993), pp. 974-979). The inhibition of menopausal flushing is demonstrated by measuring the reduction of facial blood flow in the rat (See, e.g., Escott, K., D. Beattie, H. Connor & S. Brain, "The modulation of the increase in rat facial skin blood flow observed after trigeminal ganglion stimulation", European Journal of Pharmacology, Vol. 284 (1995), pp. 69-76) as demonstrated for alpha-2 adrenergic agonists on cutaneous blood flow in the tail (See, e.g., Redfern, W., M. MacLean, R. Clague & J. McGrath, "The role of alpha-2 adrenoceptors in the vasculature of the rat tail", British Journal of

Pharmacology, Vol. 114 (1995), pp. 1724-1730). The antimigraine effect of these compounds is demonstrated by measuring the reduction of dural neurogenic inflammation to trigeminal ganglion stimulation in the rat (See, e.g., Matsubara, T., M. Moskowitz & Z. Huang, "UK-14,304, R(-)-alpha-methyl-histamine and SMS 201-995 block plasma protein leakage within dura mater by prejunctional mechanisms", European Journal of Pharmacology, Vol. 224 (1992), pp. 145-150).

Metabolic Stability

It has been observed that some peripherally acting, alpha-2-selective adrenergic agonist benzimidazoles which appear metabolically stable in vitro and in vivo in rodents, are subject to metabolic transformation in primates (i.e., monkeys and humans) via N-methylation at the benzimidazole ring. Such metabolic transformation has been shown to alter the profile of these benzimidazoles such that they may be metabolized into compounds that (1) are inactive; (2) are alpha-2 adrenoceptor antagonists; (3) possess enhanced activity at other undesired receptors, such as at alpha-1 adrenoceptors; and/or (4) have an increased potential for CNS activity. The compounds of the present invention are peripherally acting selective alpha-2 adrenergic compounds that have lower CNS activity and that resist metabolic transformation into such undesirable compounds.

Metabolic stability of the compounds described above is evaluated in vitro in a precision cut liver slice assay and in vivo in pharmacokinetic studies in primates. The precision cut liver slice assay is a well recognized, validated in vitro model to study xenobiotic metabolism in animal species and humans. (See Ekins, S. "Past, present, and future applications of precision-cut liver slices for in vitro xenobiotic metabolism." (Department of Medicine and Therapeutics, University of Aberdeen, UK.) Drug-Metab-Rev. (November, 1996) Vol. 28, No. 4: pp. 591-623). This assay is used to evaluate the metabolic activity of alpha-2 adrenergic agonists. The assay provides data on the biotransformations taking place within intact hepatocytes of the species of interest. Thus the full compliment of phase I and phase II metabolic enzymes are available to metabolize the drug as is the case in vivo.

For the pharmacokinetic studies, the compounds are administered orally to cynomolgus monkeys and measurements of administered benzimidazole compounds and corresponding N-methyl metabolites are made using 100 µL aliquots of urine collected over various time-periods post-dose. Typically, a chemical homolog or stable-isotope-labeled internal standard is added to each sample and then diluted 100x in water Ten µL of prepared sample is then

analyzed by gradient HPLC, with tandem mass spectrometry detection. Single ion reaction monitoring schemes are employed to selectively detect the test compound, its N-methyl metabolite (if present), and the internal standard.

The compounds of the present invention show little to no metabolic N-methylation in these assays. In contrast, N-methyl metabolites were found for other alpha-2 selective benzimidazole compounds such as 5-(2-imidazolinylamino)-4-methylbenzimidazole and 4-ethyl-5-(2-imidazolinylamino)benzimidazole. 5-(2-Imidazolinylamino)-4-methylbenzimidazole provides a very similar pharmacological profile to 7-cyano-5-(2-imidazolinylamino)-4-methylbenzimidazole (see Example 1 below). That is, both compounds are very potent and selective alpha-2 adrenergic agonists, with very low CNS activity. In the precision cut liver slice assay, there is no evidence of the methyl metabolite for 7-cyano-5-(2-imidazolinylamino)-4-methylbenzimidazole. However, 5-(2-imidazolinylamino)-4-methylbenzimidazole, is rapidly metabolized in this assay and it was found that its metabolite is an alpha-2 adrenergic agonist with a significantly higher potential for CNS activity than the parent compound. 4-ethyl-5-(2-Imidazolinylamino)benzimidazole, another selective alpha-2 adrenergic agonist, is rapidly and extensively N-methylated in primates. Its metabolite is a very potent alpha-2 antagonist, rather than an alpha-2 agonist.

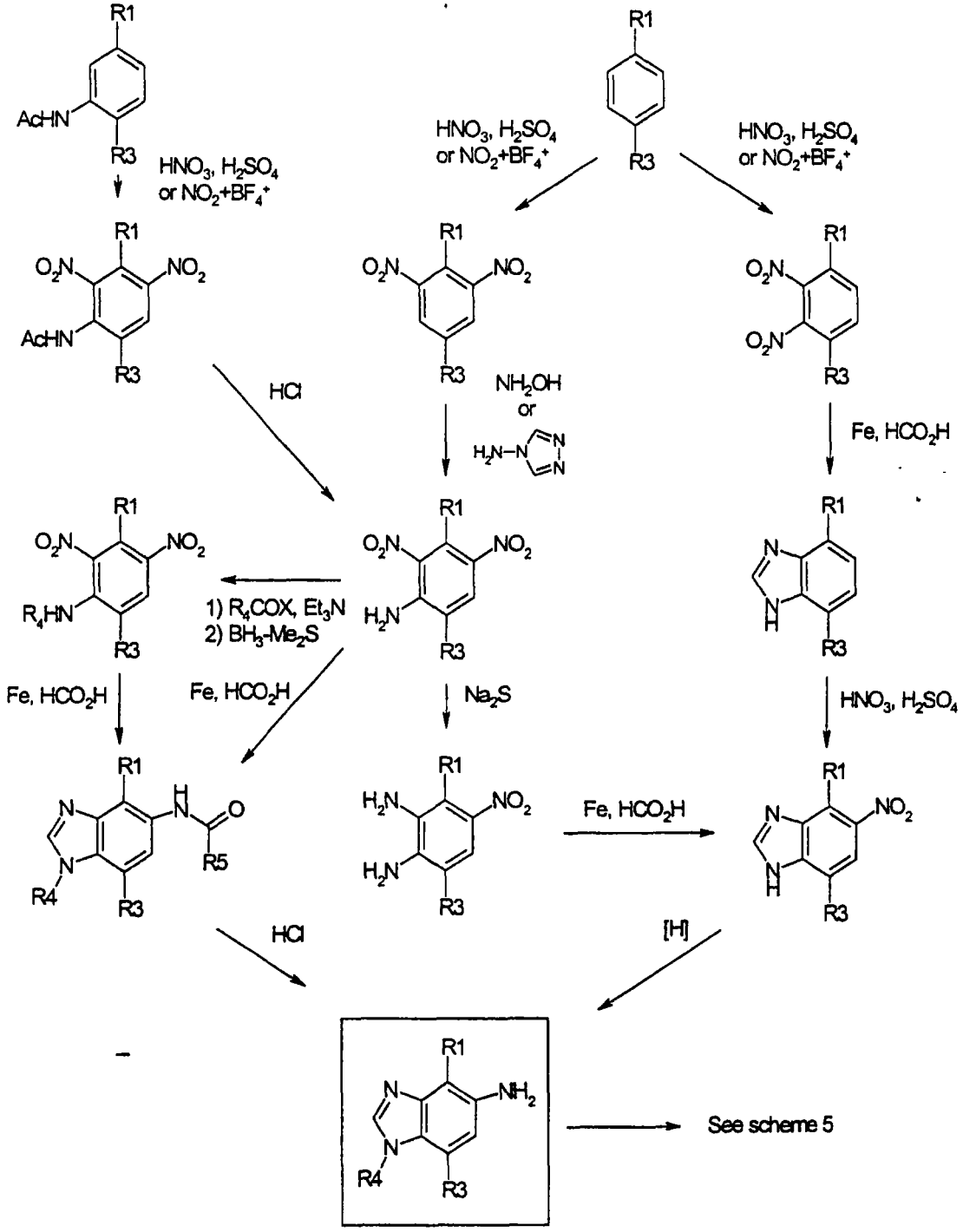
The results indicate that the metabolic transformation of benzimidazoles through N-methylation can lead to rapid formation of undesired metabolites that have different pharmacological effects relative to the parent compound and that these effects are not easily predictable. Without being bound by theory it is contemplated that the factor favorably affecting the metabolic stability of the benzimidazole compounds of this invention is the sterical hindrance provided by substituents in close proximity to the benzimidazole nitrogens.

The compounds of this invention can be made using conventional organic syntheses. Particularly preferred syntheses are carried out using the following general schemes, Schemes 1-5. In the following general reaction schemes, R1, R2, R3, R4, and R5 are as defined above. For clarity, R1, R2, R3, R4, and/or R5 do not appear on the intermediates within a specific scheme unless they are prepared or needed in that specific scheme. Preferably, R_1 is part of the starting material (see Scheme 1). R2 can be part of the starting material or introduced via amination or bromination followed by functional group manipulation (see Scheme 2). R3 can be part of the starting material (see Scheme 1) or obtained by manipulation of a carboxylic acid (see Scheme 3). R4 is introduced by alkylation of an aniline substrate prior to the benzimidazole ring formation (see Scheme 1). R5 or a direct precursor to R5 is introduced during the benzimidazole ring formation

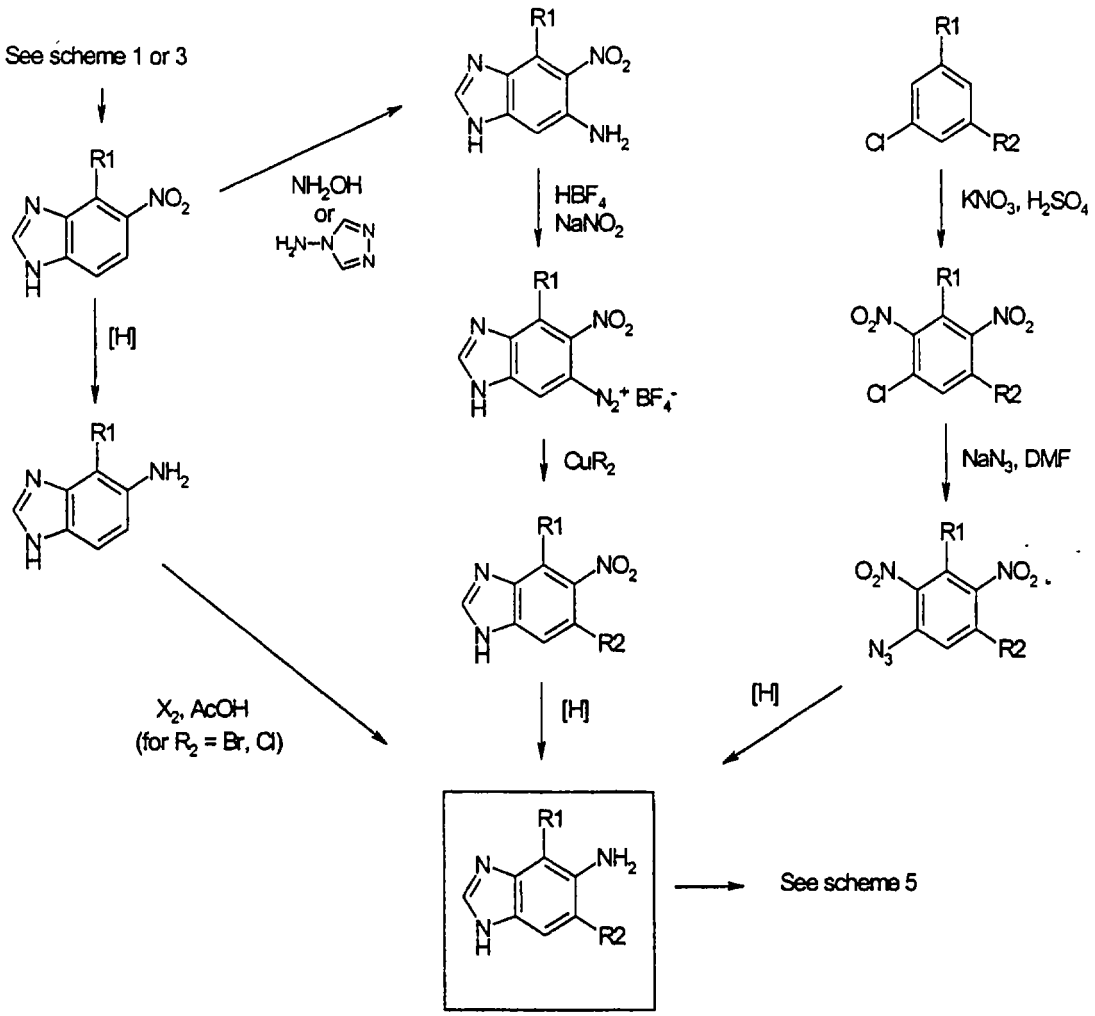
(see Scheme 4). Finally, the 5-(2-imidazolinylamino) group is conveniently obtained from the aminobenzimidazoles prepared according to Schemes 1-4 (see Scheme 5).

The starting materials depicted within the schemes are commercially available or are made from commercially available starting materials and methods known to one of ordinary skill in the art. The skilled artisan may change temperature, pressure, atmosphere, solvents, or the order of reactions as appropriate. Additionally, the skilled artisan may use protecting groups to block side reactions or increase yields as appropriate. All such modifications can be readily be carried out by the skilled artisan in the art of organic chemistry, and thus are within the scope of the invention.

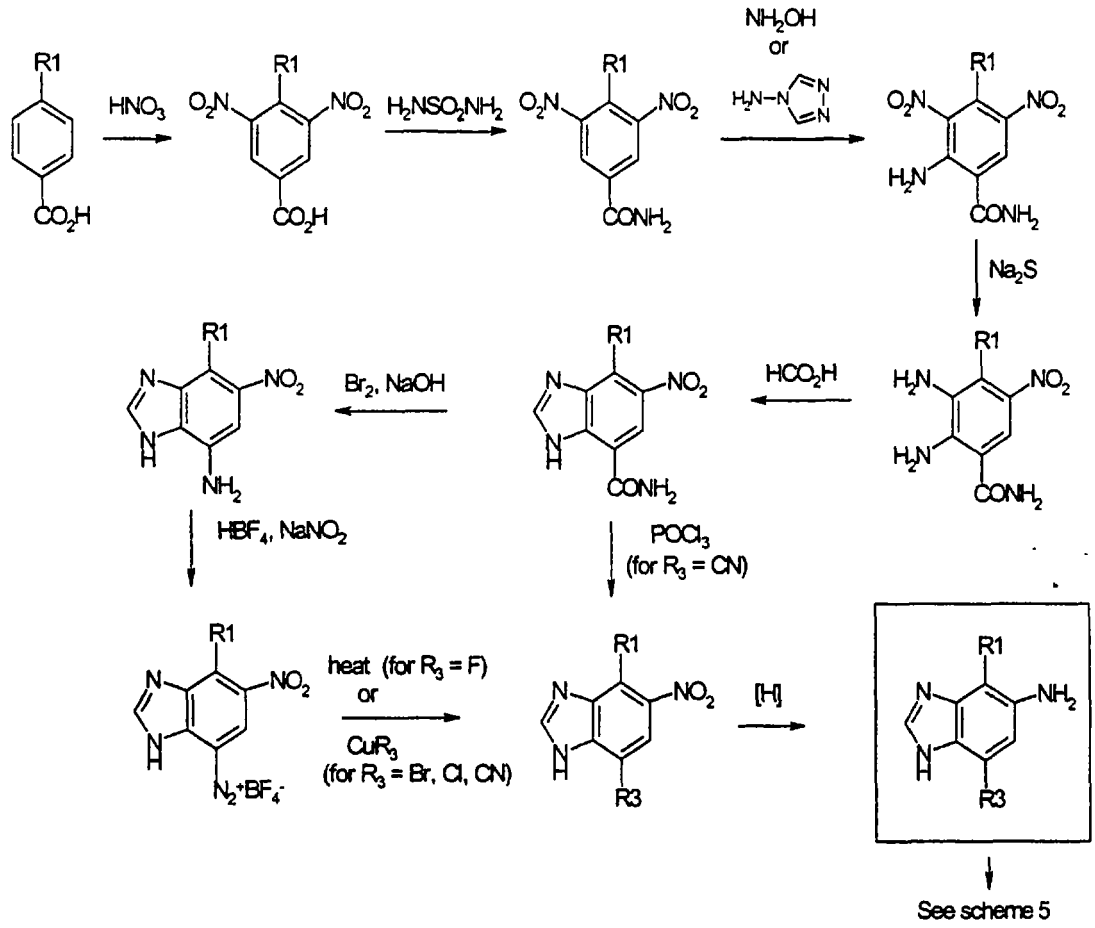
Scheme 1


R1
AcHN
R3
HNO3, H2SO4
or NO2+BF4+
O2N
NO2
HCl
NH2OH
or
H2N–N
N
Fe, HCO2H
R4HN
1) R4COX, Et3N
2) BH3-Me2S
H2N
Na2S
HNO3, H2SO4
N
H
R5
O
[H]
NH2
See scheme 5

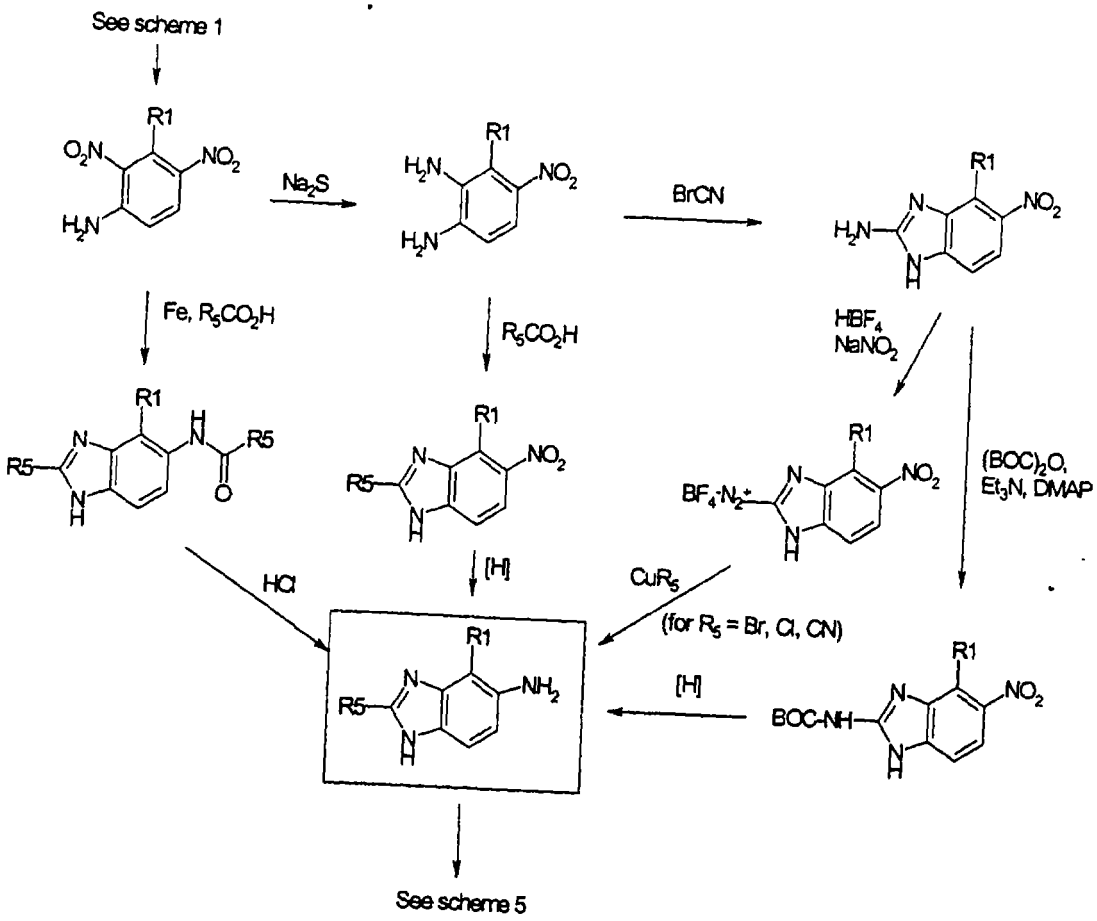
Scheme 2


See scheme 1 or 3
[H]
NH_2OH or
HBF_4 $NaNO_2$
CuR_2
X_2, AcOH (for R_2 = Br, Cl)
KNO_3, H_2SO_4
NaN_3, DMF
[H]
[H]
See scheme 5

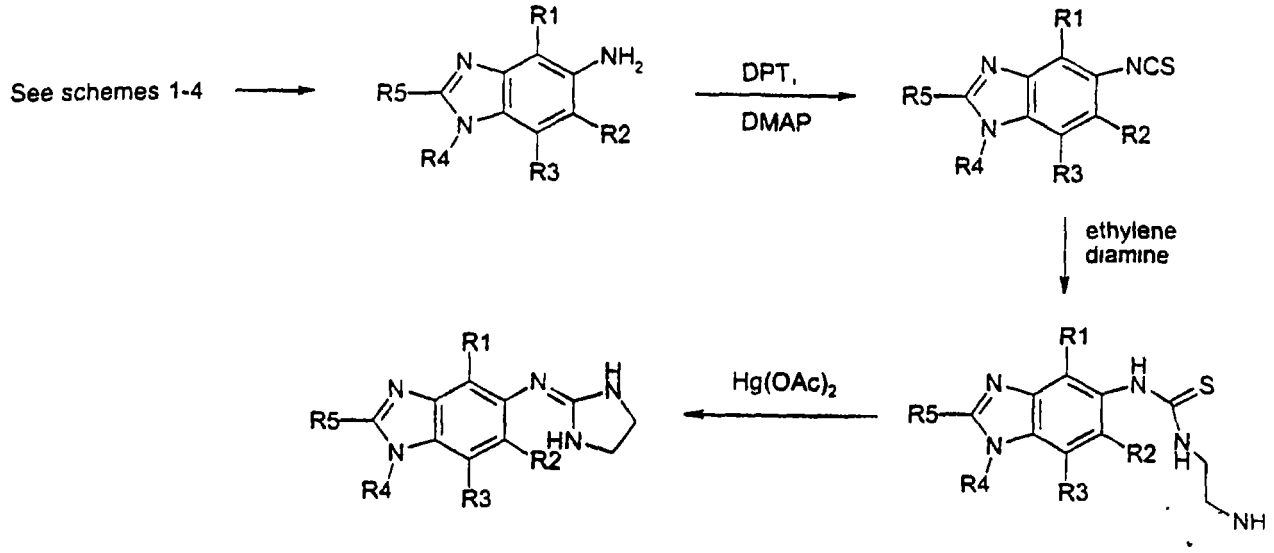
Scheme 3


R1
HNO3
O2N
NO2
CO2H
H2NSO2NH2
CONH2
NH2OH
or
H2N-N
N
N
H2N
Na2S
HCO2H
POCl3
(for R3 = CN)
Br2, NaOH
HBF4, NaNO2
N2+BF4-
heat (for R3 = F)
or
CuR3
(for R3 = Br, Cl, CN)
[H]
R3
See scheme 5

Scheme 4


See scheme 1
R1
O_2N
NO_2
H_2N
Na_2S
H_2N
NO_2
BrCN
H_2N
N
H
NO_2
Fe, R_5CO_2H
R_5CO_2H
HBF_4
$NaNO_2$
R5
N
H
O
$BF_4^-N_2^+$
$(BOC)_2O$,
Et_3N, DMAP
HCl
[H]
CuR_5
(for R_5 = Br, Cl, CN)
NH_2
[H]
BOC-NH
See scheme 5

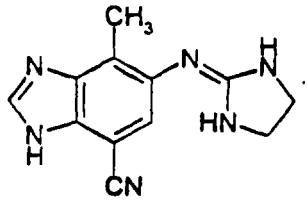
Scheme 5



Examples

The following non-limiting examples illustrate the compounds of the present invention.

Example 1



7-Cyano-5-(2-imidazolinylamino)-4-methylbenzimidazole

2,6-Dinitro-p-toluic acid. To a 500 mL roundbottom flask is added 120 mL of concentrated sulfuric acid. This is cooled to 0°C and to this is added p-toluic acid (30 g, 0.22 mole). To this mixture is slowly added a mixture of fuming nitric acid (25 mL) and concentrated sulfuric acid (100 mL) *via* an addition funnel. The resulting mixture is then stirred at 0°C for 10 minutes, slowly warmed first to room temperature, then to 90°C for 1.5 hours. The mixture is cooled to room temperature and poured into ice/water. The resulting solid is then filtered and dried to afford 2,6-dinitro-p-toluic acid as an off-white solid.

2,6-Dinitro-p-toluic carboxamide. A mixture of 2,6-dinitro-p-toluic acid (15.14 g, 66.9 mmol) and sulfamide (14.79 g, 153.8 mmol) in anhydrous pyridine

(80 mL) is stirred under an argon atmosphere at 100°C for 3 hours. The mixture is poured into ice/water and the resulting precipitate is filtered and washed with water to afford 2,6-dinitro-p-toluic carboxamide as an off-white solid.

3-Amino-2,6-dinitro-p-toluic carboxamide. To a 1 L 3-neck round bottom flask equipped with a mechanical stirrer, are placed 2,6-dinitro-p-toluic carboxamide (4.0 g, 18 mmol) and hydroxylamine hydrochloride (3.3 g, 48 mmol) in ethanol (550 mL) and water (24 mL). The mixture is cooled to 0°C and treated dropwise with a saturated solution of potassium hydroxide in methanol (80 mL) over a period of 1.5 hours. The resulting mixture is poured into a 2 L round bottom flask and diluted with 400 mL of water. The methanol and ethanol are then removed *via* rotary evaporation. A yellow precipitate formed which is filtered to give rise to 3-amino-2,6-dinitro-p-toluic carboxamide as fine yellow needles.

2,3-Diamino-6-nitro-p-toluic carboxamide. To a mixture of 3-amino-2,6-dinitro-p-toluic carboxamide (2.2 g, 9.2 mmol) in ethanol (200 mL) at 80°C is added dropwise a solution of sodium sulfide (2.2 g, 28 mmol) in water (80 mL) over a period of one hour. The mixture is stirred at 80°C for another 2 hours, then allowed to cool to room temperature and poured into ice. The mixture is extracted with ethyl acetate (5x300 mL). The combined extracts are dried over magnesium sulfate and rotary evaporated to give rise to 2,3-diamino-6-nitro-p-toluic carboxamide as a red/brown solid. The compound is used in the next step without further purification.

7-(4-Methyl-5-nitrobenzimidazolyl)carboxamide. A solution of 2,3-diamino-6-nitro-p-toluic carboxamide (1.49 g, 7.1 mmol) in formic acid (10 mL) is stirred at 100°C for two hours. The solution is cooled to room temperature and poured into ice and basified to pH=10 with concentrated ammonium hydroxide. A brown precipitate forms which is filtered to afford 7-(4-methyl-5-nitrobenzimidazolyl)carboxamide as a tan solid.

7-Cyano-4-methyl-5-nitrobenzimidazole. A mixture of 7-(4-methyl-5-nitrobenzimidazolyl)carboxamide (1.5 g, 7.0 mmol) in phosphorous oxychloride (20 mL) and toluene (20 mL) is heated to reflux under an argon atmosphere for two hours. The mixture is cooled to room temperature, poured into ice and basified to pH=10 with concentrated ammonium hydroxide. The resulting mixture is extracted with 3:1 methylene chloride/isopropyl alcohol (6x100 mL), and the combined extracts dried over magnesium sulfate and rotary evaporated. The residue is purified by flash chromatography on silica gel, eluting with 9:1:0.1 chloroform:methanol:ammonium hydroxide to afford 7-cyano-4-methyl-5-nitrobenzimidazole as a yellow solid.

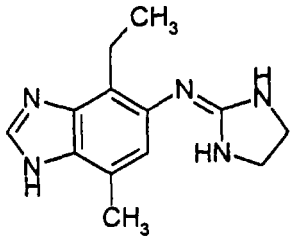
5-Amino-7-cyano-4-methylbenzimidazole. A mixture of 7-cyano-4-methyl-5-nitrobenzimidazole (0.91 g, 4.5 mmol) and 10% palladium-on-carbon (100 mg) in methanol (200 mL) is treated with an atmosphere of hydrogen (1 atm, balloon) for 14 hours. The resulting mixture is filtered through Celite and rotary evaporated. The residue is purified by flash chromatography (silica gel, 95:5 ethyl acetate:methanol) to afford 5-amino-7-cyano-4-methylbenzimidazole.

7-Cyano-5-isothiocyanato-4-methylbenzimidazole. To a solution of di-2-pyridylthionocarbonate (1.02 g, 3.1 mmol) and 4-dimethylaminopyridine (25 mg, 0.21 mmol) in tetrahydrofuran (350 mL) is added dropwise a solution of 5-amino-7-cyano-4-methylbenzimidazole (0.36 g, 2.1 mmol) in tetrahydrofuran (50 mL). The solution is stirred for one hour at room temperature. The reaction mixture is rotary evaporated and the residue is purified by flash chromatography (silica gel, 100% ethyl acetate) to give 7-cyano-5-isothiocyanato-4-methylbenzimidazole as an off-white solid.

N-5-(7-Cyano-4-methylbenzimidazolyl)-N'-2-aminoethylthiourea. A solution of 7-cyano-5-isothiocyanato-4-methylbenzimidazole (0.29 g, 1.35 mmol) in tetrahydrofuran (30 mL) is added dropwise to a solution of ethylenediamine (0.41 g, 6.8 mmol) in tetrahydrofuran (30 mL). A white precipitate forms after the solution has stirred at room temperature for 15 minutes. The reaction mixture is rotary evaporated to afford N-5-(7-cyano-4-methylbenzimidazolyl)-N'-2-aminoethylthiourea as an off-white solid.

7-Cyano-5-(2-imidazolinylamino)-4-methylbenzimidazole. To a 500 mL round bottom flask are added methanol (150 mL) and N-5-(7-cyano-4-methylbenzimidazolyl)-N'-2-aminoethylthiourea (0.31 g, 1.1 mmol). This mixture is heated slightly with a heat gun to provide a homogeneous mixture. To this mixture is added mercuric acetate (0.39 g, 1.2 mmol). The resulting mixture is stirred for 4 hours at room temperature then filtered through Celite and concentrated to afford 7-cyano-5-(2-imidazolinylamino)-4-methylbenzimidazole as a white foam.

Example 2



4-Ethyl-5-(2-imidazolinylamino)-7-methylbenzimidazole

3-(1-Hydroxyethyl)-6-methylaniline. To an ice-cold solution of 4-methyl-3-nitroacetophenone (25 g, 139 mmol) in methanol (200 mL) is added sodium borohydride (6.2 g, 163 mmol) over 15 minutes. The mixture is stirred at room temperature for 1 hour, then is quenched with water. The mixture is rotary evaporated and the residue is partitioned between water and ethyl acetate. The organic layer is dried (magnesium sulfate) and rotary evaporated to afford a light brown viscous oil. The oil is diluted with ethyl acetate (200 mL), 5% palladium-on-carbon (5 g) is added and the mixture is treated with hydrogen at 40 psi for 18 hours. The mixture is then filtered on Celite and the filtrate is rotary evaporated to afford 3-(1-hydroxyethyl)-6-methylaniline as a light yellow, pasty solid.

3-Ethyl-6-methylacetanilide. A mixture of 3-(1-hydroxyethyl)-6-methylaniline (21.3 g, 139 mmol), acetic anhydride (28 mL, 296 mmol), triethylamine (41 mL, 296 mmol) and 4-dimethylaminopyridine (0.5 g, 4 mmol) in methylene chloride (200 mL) is stirred at room temperature for 3 hours. Methanol (50 mL) is added and the mixture is rotary evaporated. The residue is partitioned between water and ethyl acetate. The organic layer is washed with water, 1N hydrochloric acid, water and brine, then dried (magnesium sulfate) and rotary evaporated. The residue is diluted with trifluoroacetic acid (100 mL) and cooled in an ice bath. Diethylsilane (35 mL, 270 mmol) is added and the resulting mixture is stirred at room temperature for 2 hours. The mixture is rotary evaporated and the residue is purified by flash chromatography on silica gel (hexane:ethyl acetate 3:1) to afford 3-ethyl-6-methylacetanilide as a foamy white solid.

2,4-Dinitro-3-ethyl-6-methylacetanilide. To an ice-cold mixture of 3-ethyl-6-methylacetanilide (11.5 g, 64.8 mmol) in conc. sulfuric acid (90 mL) is slowly added fuming nitric acid (7 mL). The mixture is stirred for 30 minutes in the ice bath, then for 1 hour at room temperature. The mixture is poured into ice and the solid that forms is collected by filtration, washed with water and dried under vacuum. The mixture of 2,4-dinitro-3-ethyl-6-methylacetanilide and 4,5-dinitro-3-ethyl-6-methylacetanilide is separated by flash chromatography on silica gel (hexane:ethyl acetate gradient 4:1 to 2:3).

2,4-Dinitro-3-ethyl-6-methylaniline. A mixture of 2,4-dinitro-3-ethyl-6-methylacetanilide (4.0 g, 14.9 mmol), potassium carbonate (2.6 g, 19 mmol) and 6N hydrochloric acid (40 mL) in methanol (100 mL) is heated to reflux for 2 hours. The mixture is cooled to room temperature, brought to pH 9 with ammonium hydroxide and rotary evaporated. The residue is purified by flash chromatography

on silica gel (chloroform:methanol 9:1) to afford 2,4-dinitro-3-ethyl-6-methylaniline as a yellow solid.

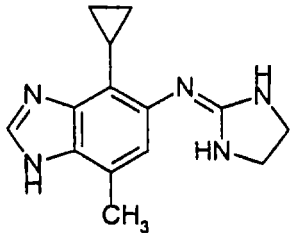
4-Ethyl-5-formamido-7-methylbenzimidazole. A mixture of 2,4-dinitro-3-ethyl-6-methylaniline (2.0 g, 8.9 mmol) and iron powder (5.0 g, 90 mmol) in 90% formic acid (36 mL) is heated to reflux for 18 hours. The mixture is cooled to room temperature, diluted with methanol (75 mL) and filtered through Celite. The filtrate is rotary evaporated and the residue is purified by flash chromatography on silica gel (chloroform:methanol 9:1) to afford 4-ethyl-5-formamido-7-methylbenzimidazole as a tan solid.

5-Amino-4-ethyl-7-methylbenzimidazole. A mixture of 4-ethyl-5-formamido-7-methylbenzimidazole (1.7 g, 8.36 mmol), potassium carbonate (2.0 g, 14.4 mmol) and 6N hydrochloric acid (34 mL) in methanol (34 mL) is heated to reflux for 1 hour. The mixture is cooled to room temperature, brought to pH 9 with ammonium hydroxide and rotary evaporated. The residue is purified by flash chromatography on silica gel (chloroform:methanol 9:1) to afford 5-amino-4-ethyl-7-methylbenzimidazole as a tan solid.

4-Ethyl-5-isothiocyanato-7-methylbenzimidazole. To a mixture of di-2-pyridyl thionocarbonate (0.72 g, 3.11 mmol) and 4-dimethylaminopyridine (0.02 g) in ethyl acetate (50 mL) is added dropwise a solution of 5-amino-4-ethyl-7-methylbenzimidazole (0.42 g, 2.39 mmol) in ethyl acetate (20 mL) and methanol (5 mL). The mixture is stirred at room temperature for 3 hours, then rotary evaporated. The residue is purified by filtration on a short pad of silica gel, eluting with ethyl acetate, to afford 4-ethyl-5-isothiocyanato-7-methylbenzimidazole as a tan solid.

4-Ethyl-5-(2-imidazolinylamino)-7-methylbenzimidazole, trifluoroacetic acid salt. To a mixture of ethylenediamine (0.65 mL, 9.66 mmol) in methylene chloride (50 mL) is added a suspension of 4-ethyl-5-isothiocyanato-7-methylbenzimidazole (0.42 g, 1.93 mmol) in methylene chloride (50 mL). The mixture is stirred for 1 hour at room temperature, then rotary evaporated. The residue is diluted with methanol (100 mL) and mercuric acetate (0.74 g, 2.32 mmol) is added. The mixture is stirred at room temperature for 2 hours. The mixture is filtered on Celite with a methanol wash of the solids. The filtrate is rotary evaporated and the reside is purified by preparative High Pressure Liquid Chromatography (HPLC) (C18 column; flow rate 45 mL/min; solvent gradient: 0.1% trifluoroacetic acid (in water)/acetonitrile starting at 95/5 and going to 0/100 over 45 minutes) to afford 4-ethyl-5-(2-imidazolinylamino)-7-methylbenzimidazole as a trifluoroacetic acid salt.

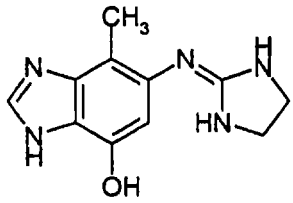
Example 3



4-Cyclopropyl-5-(2-imidazolinylamino)-7-methylbenzimidazole

Commercially available 1-(4-methylphenyl)-1-cyclopropane carboxylic acid is treated with nitronium tetrafluoroborate in sulfolane to afford 1-(4-methyl-3-nitrophenyl)-1-cyclopropane carboxylic acid. This is converted to 1-(4-methyl-3-nitrophenyl)-1-bromocyclopropane by treatment with mercuric oxide and bromine in methylene chloride. Reduction with zinc dust in the presence of calcium chloride in aqueous ethanol affords 5-cyclopropyl-2-methylaniline. Conversion to 4-cyclopropyl-5-(2-imidazolinylamino)-7-methylbenzimidazole is completed in the same manner as 4-ethyl-5-(2-imidazolinylamino)-7-methylbenzimidazole (see Example 2).

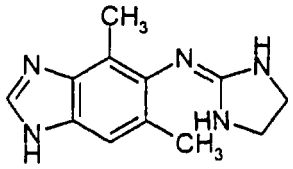
Example 4



–7-Hydroxy-5-(2-imidazolinylamino)-4-methylbenzimidazole

(2-Imidazolinylamino)-7-methoxy-4-methylbenzimidazole is made in the same manner as 4-ethyl-5-(2-imidazolinylamino)-7-methylbenzimidazole except that 2-methoxy-5-methylacetanilide is used instead of 3-ethyl-6-methylacetanilide (see Example 2). Cleavage of the methyl ether is achieved with pyridinium hydrochloride to afford 7-hydroxy-5-(2-imidazolinylamino)-4-methylbenzimidazole.

Example 5



4,6-Dimethyl-5-(2-imidazolinylamino)benzimidazole

5-Chloro-2,4-dinitro-*m*-xylene. To ice cold concentrated sulfuric acid is added 5-chloro-*m*-xylene (10.0 g, 71 mmol). With vigorous stirring, solid potassium nitrate (14.35 g, 0.14 mol) is added slowly over 30 minutes. Upon completion of addition, the reaction mixture is warmed to room temperature and stirred for 2 hours. The solid that has formed is filtered and recrystallized from ethanol/water. This material is further purified by flash chromatography on silica gel (95:5 hexane:ethyl acetate) to afford 5-chloro-2,4-dinitro-*m*-xylene as a white crystalline solid.

5-Azido-2,4-dinitro-*m*-xylene. A mixture of 5-chloro-2,4-dinitro-*m*-xylene (707 mg, 3.1 mmol), sodium azide (219 mg, 3.37 mmol) and N,N-dimethylformamide (10 mL) is heated at 80°C for 45 minutes then cooled to room temperature, poured into ice/water and extracted with ethyl acetate (3 x 50 mL). The combined organic layers are dried (magnesium sulfate), filtered, and concentrated *via* rotary evaporation to provide 5-azido-2, 4-dinitro-*m*-xylene as a yellow/brown solid.

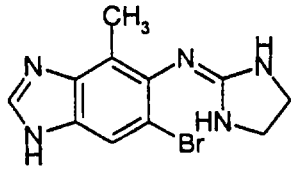
4,6-Dimethyl-5-nitrobenzimidazole. A mixture of 5-azido-2,4-dinitro-*m*-xylene (650 mg, 2.7 mmol), 10% palladium-on-carbon (100 mg) and 80% formic acid (20 mL) is heated to 80°C for 30 minutes, cooled to room temperature, and filtered through a plug of silica gel (eluting with water). The filtrate is basified (~ pH 10) with 28% ammonium hydroxide and extracted with ethyl acetate (3 x 100 mL). The combined organic layers are dried (magnesium sulfate), filtered, and concentrated to afford 4,6-dimethyl-5-nitrobenzimidazole as a yellow oil.

5-Amino-4,6-dimethylbenzimidazole. A heterogeneous mixture of 4,6-dimethyl-5-nitrobenzimidazole (410 mg, 2.14 mmol) and 10% palladium-on-carbon (50 mg) in methanol (25 mL) is treated with an atmosphere of hydrogen (1 atm, balloon) for 16 hours. The resulting mixture is filtered through Celite and rotary evaporated. The residue is purified by chromatography on silica gel (95:5 methylene chloride:methanol) to afford 5-amino-4,6-dimethylbenzimidazole as a white solid.

4,6-Dimethyl-5-isothiocyanatobenzimidazole. A mixture of 5-amino-4,6-dimethylbenzimidazole (265 mg, 1.64 mmol), tetrahydrofuran (20 mL), di-2-pyridylthionocarbonate (584 mg, 1.81 mmol), and 4-dimethylaminopyridine (20 mg, 0.016 mmol is stirred at room temperature for 2 hours. The mixture is rotary evaporated and the residue is purified by chromatography on silica gel (50:50 hexane:ethyl acetate) to provide 4,6-dimethyl-5-isothiocyanatobenzimidazole as an off-white solid.

4,6-Dimethyl-5-(2-imidazolinylamino)benzimidazole. A solution of 4,6-dimethyl-5-isothiocyanatobenzimidazole (250 mg, 1.23 mmol) in methylene chloride (5 mL) is added dropwise to a solution of ethylenediamine (370 mg, 6.2 mmol) in methylene chloride (5 mL). The resulting solution is stirred at room temperature for 15 minutes then rotary evaporated. The residue is dissolved in methanol (10 mL) and to this solution is added mercuric acetate (390 mg, 1.23 mmol). The resulting reaction mixture is stirred at room temperature for 1 hour, filtered through a pad of silica gel and rotary evaporated. The residue is purified by chromatography on silica gel (70:30:0.5 methylene chloride:methanol:ammonium hydroxide) to afford 4,6-dimethyl-5-(2-imidazolinylamino)benzimidazole as a white solid.

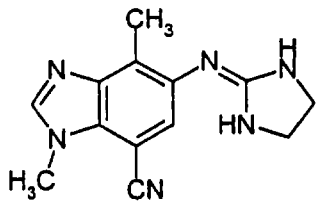
Example 6



6-Bromo-5-(2-imidazolinylamino)-4-methylbenzimidazole

Commercially available 2,6-dinitrotoluene is converted to 5-amino-4-methylbenzimidazole according to scheme 1. Bromination is achieved by treatment with bromine in acetic acid. The synthesis is then completed according to Scheme 5.

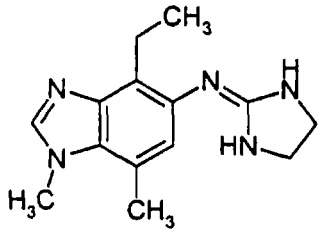
Example 7



7-Cyano-1,4-dimethyl-5-(2-imidazolinylamino)benzimidazole

This compound is made according to a combination of Schemes 1 and 3 using 3-amino-2,6-dinitro-p-toluic carboxamide as prepared in Example 1.

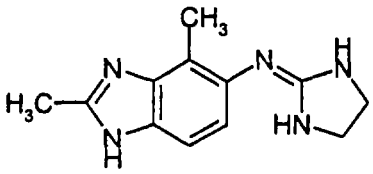
Example 8



1,7-Dimethyl-4-ethyl-5-(2-imidazolinylamino)benzimidazole

This compound is made according to Scheme 1. 2,4-Dinitro-3-ethyl-6-methylaniline is treated with paraformaldehyde in concentrated sulfuric acid to afford N-methyl-2,4-dinitro-3-ethyl-6-methylaniline. The synthesis is completed in the same manner as 4-ethyl-5-(2-imidazolinylamino)benzimidazole (see Example 2).

Example 9



2,4-Dimethyl-5-(Imidazolinylamino)benzimidazole

2,3-diamino-6-nitrotoluene. To a solution 3-methyl-2,4-dinitroaniline (30 g) in boiling ethanol (750 mL) is added dropwise over 90 minutes a solution of sodium sulfide nonahydrate (109.6 g) in water (750 mL). At the end of the addition, the mixture is heated to reflux for 30 minutes then poured into ice (2000 g) and allowed to stand until all the ice has melted. The mixture is then extracted with methylene chloride and the organic layer is dried over magnesium sulfate and rotary evaporated. The residue is purified by flash chromatography on silica gel, eluting with methylene chloride to afford 2,3-diamino-6-nitrotoluene as an orange solid.

2,4-Dimethyl-5-nitrobenzimidazole. A mixture of 2,3-diamino-6-nitrotoluene (0.945 g, 5.65 mmol), conc. hydrochloric acid (5 mL) and glacial acetic acid (30 mL)

is heated to reflux for 2 hours. The mixture is cooled to room temperature, then poured in a mixture of crushed ice (100 mL) and ammonium hydroxide (100 mL) and extracted with 20% methanol in chloroform (2 x 400 mL). The combined extracts are dried over potassium carbonate and rotary evaporated to afford 2,4-dimethyl-5-nitrobenzimidazole as a brown solid. The product is used in the following step without further purification.

1-*t*-Butoxycarbonyl-2,4-dimethyl-5-nitrobenzimidazole. A mixture of 2,4-methyl-5-nitrobenzimidazole (0.63 g, 4.3 mmol), di-*t*-butyl-dicarbonate (0.24 g, 10.8 mmol), triethylamine (0.725 mL, 5.2 mmol) and 4-dimethylaminopyridine (0.05 g) in ethyl acetate (45 mL) is stirred at room temperature overnight. The mixture is rotary evaporated and the residue purified by flash chromatography on silica gel, eluting with 10% ethyl acetate in hexane to afford 1-*t*-butoxycarbonyl-2,4-dimethyl-5-nitrobenzimidazole as a white solid.

5-Amino-1-*t*-butoxycarbonyl-2,4-dimethylbenzimidazole. To a solution of 1-*t*-butoxycarbonyl-2,4-dimethyl-5-nitrobenzimidazole (1.26 g, 4.32 mmol) in methanol (15 mL)/ethyl acetate (100 mL) are added 10% palladium-on-carbon (0.1 g) and ammonium formate (1.09 g, 17.3 mmol). The mixture is stirred at room temperature for 3 hours, then filtered on Celite with a methanol wash of the solids. The filtrate is rotary evaporated and the residue is purified by flash chromatography on silica gel, eluting with 20% ethyl acetate in hexane to afford 5-amino-1-*t*-butoxycarbonyl-2,4-dimethylbenzimidazole as a white solid.

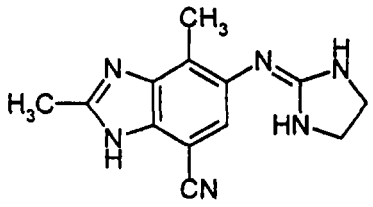
1-*t*-Butoxycarbonyl-2,4-dimethyl-5-isothiocyanatobenzimidazole. A solution of 5-amino-1-*t*-butoxycarbonyl-2,4-dimethylbenzimidazole (1.1 g, 4.2 mmol) in methylene chloride (60 mL) is added dropwise over 30 minutes to a solution of di-2-pyridyl thionocarbonate (1.9 g, 8.2 mmol) and 4-dimethylaminopyridine (0.1 g) in methylene chloride (150 mL). The mixture is stirred for 2 hours at room temperature then rotary evaporated. The residue is purified by flash chromatography on silica gel, eluting with 10% ethyl acetate/hexane to afford 1-*t*-butoxycarbonyl-2,4-dimethyl-5-isothiocyanatobenzimidazole as a white solid.

N-(1-*t*-Butoxycarbonyl-2,4-dimethyl-5-benzimidazolyl)-N'-2-aminoethylthiourea. A solution of 1-*t*-butoxycarbonyl-2,4-dimethyl-5-isothiocyanatobenzimidazoline (1.15 g, 3.8 mmol) in methylene chloride (100 mL) is added dropwise over 15 minutes to 1,2-ethylenediamine (1.26 mL, 18.9 mmol) in solution in methylene chloride (200 mL). The mixture is stirred for 2 hours at room temperature. The mixture is rotary evaporated and the residue is triturated with ether (150 mL) for 1 hour at room temperature. The solid is filtered and dried *in*

vacuo to afford N-(1-*t*-butoxycarbonyl-2,4-dimethyl-5-benzimidazolyl)-N'-2-aminoethylthiourea as a white solid.

2,4-Dimethyl-5-(2-imidazolinylamino)benzimidazole. A mixture of N-(1-*t*-butoxycarbonyl-2,4-dimethyl-5-benzimidazolyl)-N'-2-aminoethylthiourea (1.33 g, 3.66 mmol) and mercuric acetate (1.45 g, 4.54 mmol) in methanol (150 mL) is stirred at room temperature for 1 hour. The resulting black mixture is filtered on Celite with a methanol wash of the solids. The filtrate is rotary evaporated and the residue is purified by flash chromatography on a short pad of silica gel, eluting with 10% methanol/chloroform containing 1% of ammonium hydroxide. The product-containing fractions are collected and rotary evaporated to afford 2,4-dimethyl-5-(2-imidazolinylamino)benzimidazole as a white solid.

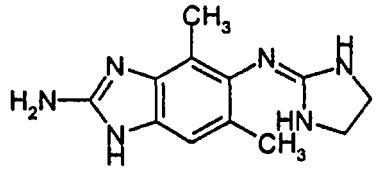
Example 10



7-Cyano-2, 4-dimethyl-5-(2-imidazolinylamino)benzimidazole

This compound is made according to Scheme 4 from 3-amino-2,6-dinitro-p-toluic carboxamide prepared in Example 1.

Example 11



2-Amino-4,6-dimethyl-5-(2-imidazolinylamino)benzimidazole

N-Acetyl-3,5-dimethylaniline. A mixture of 3,5-dimethylaniline (20 g, 165 mmol), acetic anhydride (24 mL, 247 mmol) and triethylamine (70 mL, 495 mmol) in methylene chloride (300 mL is stirred at room temperature for 16 hours. The mixture is washed with water, dried (magnesium sulfate) and rotary evaporated. The residue is triturated with hexane and filtered to afford N-acetyl-3,5-dimethylaniline (25 g).