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UNITED STATES DISTRICT COURT

CENTRAL DISTRICT OF CALIFORNIA – WESTERN DIVISION

ASTELLAS US LLC, ASTELLAS PHARMA US, INC., ITEM DEVELOPMENT AB, AND KING PHARMACEUTICALS RESEARCH AND DEVELOPMENT, INC.

Plaintiffs,

v.

ANAZAOHEALTH CORPORATION, NUVIEW RADIOPHARMACEUTICALS, INC., PAUL J. CROWE, KEITH RUSTVOLD, MTS HEALTH SUPPLIES, INC., NABIL SABA, AND GHASSAN SALAYMEH

Defendants,

Case No. CV08-01083 JFW (VBKx)

[Hon. John F. Walter]

SECOND AMENDED COMPLAINT FOR PATENT INFRINGEMENT, INTENTIONAL INTERFERENCE WITH PROSPECTIVE ECONOMIC ADVANTAGE, AND UNFAIR COMPETITION; DECLARATION RE JOINDER PURSUANT TO LOCAL RULE 19-2

NUVIEW RADIOPHARMACEUTICALS, INC., PAUL J. CROWE, AND KEITH RUSTVOLD

Counter Claimants,

v.

ASTELLAS US LLC, ASTELLAS PHARMA US, INC., ITEM DEVELOPMENT AB, AND KING PHARMACEUTICALS RESEARCH AND DEVELOPMENT, INC.

Counter Defendants,

NUVIEW RADIOPHARMACEUTICALS, INC., PAUL J. CROWE, AND KEITH RUSTVOLD

Cross Claimants,

v.

ANAZAOHEALTH CORPORATION

Cross Defendant.

COMPLAINT

Plaintiffs, Astellas US LLC and Astellas Pharma US, Inc. (collectively "Astellas"), Item Development AB ("Item"), and King Pharmaceuticals Research and Development, Inc. ("King"), for their complaint against Defendants NuView Radiopharmaceuticals, Inc. ("NuView"), Paul J. Crowe, and Keith Rustvold (collectively "the NuView defendants") and MTS Health Supplies, Inc. ("MTS"), Nabil Saba, and Ghassan Salaymeh (collectively "the MTS defendants"), allege as follows:

SUMMARY AND NATURE OF THE ACTION

1. This is an action under the patent laws of the United States, 35 U.S.C. §§ 1 *et seq.* for infringement by Defendants of patent rights exclusively licensed to Astellas, unfair competition in violation of the California Business and Professions Code § 17200

et seq. and § 4381, and interference with prospective economic advantage.

2. Plaintiff Astellas markets Adenoscan® (hereinafter "Adenoscan"), a branded pharmaceutical product used as a diagnostic for myocardial perfusion imaging ("MPI"). Such use is covered by patent rights Astellas licenses exclusively from plaintiffs King and Item. MPI is an imaging procedure used to diagnose and assess the severity of coronary artery disease. The NuView defendants embarked on an illegal scheme to market an unbranded copy of Adenoscan that contains the same active pharmaceutical ingredient, adenosine, but has never been approved by the U.S. Food and Drug Administration (FDA) for any purpose. In violation of the California Pharmacy Law, NuView advertised the services of AnazaoHealth Corporation ("Anazao") to fill prescriptions for its unapproved adenosine and supply it within California. Anazao and the NuView defendants purported to sell and solicit prescriptions for their unapproved product for use not as a diagnostic, but for alleged treatment of "reperfusion injury" in patients suffering from myocardial infarction (heart attack). Such use for treatment of reperfusion injury has never been approved by the FDA and, on information and belief, is not accepted or used by physicians who routinely treat myocardial infarction and is certainly not performed by the physicians routinely involved in providing imaging services to whom NuView's marketing efforts were directed.

3. On information and belief, reference to this purported therapeutic use of the unapproved product was merely a ruse intended to deflect attention from the NuView defendants' willful patent infringement resulting from the offers to sell and sales of that product as a substitute for Adenoscan in MPI. On information and belief, the NuView defendants perpetuated this ruse by providing false and misleading information on their website, in their marketing materials, and even on the label and information accompanying the unapproved product to suggest that it was for use in reperfusion injury when, in fact, it was knowingly marketed and distributed for use in MPI. The NuView defendants specifically targeted for sales and furnished their

product to clinics that perform the diagnostic MPI procedure but would never treat reperfusion injury. The NuView sales representatives actively encouraged the use of the unapproved product for MPI in a manner that constituted infringement of plaintiffs' patent rights and contradicted the product information provided by Anazao and NuView.

4. Also on information and belief, Anazao and NuView priced their product, as befits its status as an unapproved copy, at a substantial discount compared to authentic Adenoscan. However, the cost savings were not passed along to patients and patients were unaware that they were receiving an unapproved drug rather than authentic Adenoscan.

5. The NuView defendants have also marketed, sold, and distributed adenosine products supplied by the MTS defendants for use in MPI procedures.

6. The MTS defendants have also independently marketed, sold, and distributed adenosine products for use in MPI procedures.

7. The Nuview defendants' and the MTS defendants' conduct constitutes willful patent infringement, intentional interference with prospective economic advantage, and unfair competition that has caused and will continue to injure and cause damages to Astellas, Item, and King unless enjoined by this Court.

THE PARTIES

8. Plaintiff Astellas US LLC is a Delaware limited liability corporation having an office and principal place of business at Three Parkway North, Deerfield, Illinois 60015-2548.

9. Plaintiff Astellas Pharma US, Inc. is a Delaware corporation having an office and principal place of business at Three Parkway North, Deerfield, Illinois 60015-2548.

10. Plaintiff Item Development AB is a Swedish corporation having an office and principal place of business at Svanholmsvagen 2A, Stocksund, SE-18275, Sweden.

11. Plaintiff King is a Delaware corporation having an office and principal place of business at 4000 CentreGreen Way, Suite 300, Cary, North Carolina 27513.

12. On information and belief, Anazao is a Florida corporation having a principal place of business at 5710 Hoover Boulevard, Tampa, Florida 33634-5339.

13. Defendant NuView Radiopharmaceuticals is an entity having a website (www.nuviewinfo.com) that indicates a principal place of business at 1107 Snowberry Street, Park City, Utah 84098. However, according to the website for the Secretary of State of Utah, NuView Radiopharmaceuticals is not a licensed business in Utah.

14. On information and belief, 1107 Snowberrry Street, Park City, Utah 84098 is a residence within a gated community which is zoned for residential use.

15. On information and belief, defendant Paul J. Crowe is the founder, chairman, and chief executive officer of NuView Radiopharmaceuticals and a resident of California.

16. On information and belief, defendant Keith Rustvold is the National Sales Coordinator for NuView Radiopharmaceuticals and a resident of California.

17. According to records of the Secretary of State for the State of Delaware, NuView Radiopharmaceuticals is or was at one time a Delaware corporation.

18. On information and belief, Defendant MTS is a California corporation having a website (www.mtshs.com) that indicates a principal place of business at 1390 W. 6th St. Suite 122, Corona, CA 92882.

19. On information and belief, defendant Nabil Saba is chief financial officer for MTS and a resident of California.

20. On information and belief, defendant Ghassan Salaymeh is a chief executive officer of MTS and a resident of California.

JURISDICTION AND VENUE

21. This is an action for pecuniary and injunctive relief for patent infringement arising under the patent laws of the United States, Title 35, United States Code, for

unfair competition arising under the California Business and Professions Code § 17200 et seq. and § 4381, and for interference with prospective economic advantage under the common law of the State of California.

22. This Court has jurisdiction over the subject matter of the patent claim as provided in 28 U.S.C. § 1338. This Court has supplemental jurisdiction over the state law claims under 28 U.S.C. § 1367.

23. Personal jurisdiction by this Court over the NuView defendants is proper because the NuView defendants have in the past and continue to transact and/or solicit business throughout the United States, including in this district, and their infringing activities have occurred and continue to occur throughout the United States and in this district.

24. According to NuView's website, NuView maintains a sales force in California for the purpose of serving customers in the state and in this district.

25. Of the states represented on the NuView website, NuView devotes more sales representatives to California than any other state.

26. The NuView website indicates that at least three sales representatives are assigned to territories within the Central District of California.

27. On information and belief, defendants NuView and Keith Rustvold have contacted physicians by telephone and in person within the Central District of California and offered to sell and/or sold NuView's products.

28. Personal jurisdiction by this Court over the MTS defendants is proper because the MTS defendants have in the past and continue to transact and/or solicit business throughout the United States, including in this district, and their infringing activities have occurred and continue to occur throughout the United States and in this district.

29. Venue properly lies in this district under the provisions of 28 U.S.C. §§ 1391 and 1400 because NuView and MTS reside in the district for venue purposes, having purposely and repeatedly availed themselves of the privilege of doing business

within the district, because a substantial part of the events giving rise to the claim occurred in this district, and because, on information and belief, all the defendants reside in the same state.

MYOCARDIAL PERFUSION IMAGING AND ASTELLAS'S ADENOSCAN PRODUCT

30. Myocardial perfusion imaging (MPI) is a diagnostic technique for studying the blood flow in the heart. It is often used in conjunction with a radiopharmaceutical tracer, for example thallium-201 or technetium, to detect the presence or assess the severity of coronary artery disease.

31. Patients undergoing MPI typically exercise on a treadmill to increase blood flow to the heart.

32. Physicians compare images made after exercise "stress" to images made while the heart is at rest to identify areas of inadequate blood flow and diagnose whether a patient has coronary artery disease and how severe the disease may be.

33. For patients who are unable to exercise on a treadmill, physicians can perform a so-called "pharmacologic stress" procedure.

34. Adenosine is a naturally occurring compound that can be used to cause pharmacologic stress by vasodilation in conjunction with MPI.

35. When used as a vasodilator for pharmacologic stress, adenosine is typically administered by intravenous infusion at a dose of 140 micrograms/kg/min over a period of minutes.

36. Astellas markets Adenoscan, an adenosine-based product approved by the Food and Drug Administration ("FDA") for use as an adjunct to thallium-201 myocardial perfusion scintigraphy in patients unable to exercise adequately. Adenoscan contains 3 mg/ml of adenosine and is sold in 20 ml or 30 ml vials.

37. Sales of Astellas's Adenoscan amount to more than $300 million per year nationwide and in excess of $20 million annually in California.

38. While some Adenoscan customers are associated with hospitals, many purchasers of Adenoscan are free-standing out-patient clinics.

39. These outpatient clinics provide specialized services to patients in the form of diagnostic cardiac imaging. While they may be located near a hospital, outpatient clinics do not provide the range of cardiac care services found in a hospital.

40. In particular, outpatient clinics generally do not provide acute cardiac care to patients suffering from a myocardial infarction (heart attack). To the contrary, if a patient in an outpatient clinic suffers a myocardial infarction during a stress-imaging procedure, that patient is usually quickly transferred to a hospital emergency room that is better equipped to provide the full range of care required for such life-threatening situations.

41. In outpatient clinics, Adenoscan is administered under the oversight of a physician, typically a cardiologist. Nuclear medicine technologists may also be involved in the actual administration and handling of Adenoscan and the associated radiopharmaceutical tracer agents used in connection with MPI.

42. Often, nuclear medicine technologists are the point of contact for sales of Adenoscan in that they are responsible for maintaining the supply of the drug in the facility and ordering additional drug when supplies get low. The point of contact for Adenoscan sales may also be an office manager who is not a nuclear medicine technologist.

43. Adenoscan can be purchased directly from Astellas or through distributors. Astellas has a sales force that interacts directly with physicians and technologists in hospitals and outpatient clinics, providing information about Adenoscan as well as arranging for educational training programs offered by Astellas.

44. Astellas has been marketing Adenoscan throughout the United States, including California, since the mid-1990s.

45. Only Astellas has received final FDA approval to market adenosine in the form of Adenoscan for use in MPI.

THE '296 PATENT

46. On March 24, 1998, the United States Patent and Trademark Office ("PTO") duly and legally issued United States Patent No. 5,731,296 ("the '296 patent") to Item for "SELECTIVE VASODILATION BY CONTINUOUS ADENOSINE INFUSION." A copy of the '296 patent is attached hereto as Exhibit A.

47. Item is the assignee of the '296 patent by virtue of documents duly recorded at the United States Patent and Trademark Office.

48. Astellas is the exclusive licensee of certain rights in the '296 patent and enjoys the right to bring suit under this patent.

49. The FDA-approved method of using Astellas's Adenoscan product in MPI is covered by one or more claims of the '296 patent and the '296 patent is listed in the FDA's publication, Approved Drug Products with Therapeutic Equivalence Evaluations" (known as the "Orange Book") in connection with Adenoscan.

THE '877 PATENT

50. On December 10, 1991, the United States Patent and Trademark Office duly and legally issued United States Patent No. 5,070,877 ("the '877 patent") to Medco Research, Inc. (hereinafter "Medco") for a NOVEL METHOD OF MYOCARDIAL IMAGING. A copy of the '877 patent is attached hereto as Exhibit B.

51. Subsequently, the assets of Medco were acquired by King. King is the assignee of the '877 patent by virtue of documents duly recorded at the United States Patent and Trademark Office.

52. Astellas is the exclusive licensee, with right to bring suit, of certain rights in the '877 patent.

53. The FDA-approved method of using Astellas's Adenoscan product in MPI is covered by one or more claims of the '877 patent and the '877 patent is listed in the FDA's publication, Approved Drug Products with Therapeutic Equivalence Evaluations (known as the "Orange Book") in connection with Adenoscan.

NUVIEW AND ANAZAO'S ACTIVITIES IN CALIFORNIA

54. NuView's website describes NuView as "an emerging company focusing on commercializing quality diagnostic imaging agents and targeted therapies for diagnosing cancer and heart disease."

55. Among the products offered by NuView was a product described as "Compounded Adenosine."

56. NuView's website claimed that "[a]denosine has been found to offer beneficial outcomes to patients experiencing acute myocardial infarction (heart attack)" and that "[i]n contrast to other pharmaceutical agents that have shown disappointing effects in man, adenosine, a naturally occurring compound, has consistently resulted in a marked reduction in heart damage and has also improved clinical outcomes in patients treated early with heart attacks."

57. According to the NuView website, "[a]n intravenous infusion of adenosine for 3 hours should be considered as adjunctive therapy in high-risk patients undergoing reperfusion therapy."

58. The use of adenosine as a therapeutic in patients experiencing acute myocardial infarction has not been approved by the FDA.

59. On information and belief, NuView has not sought to obtain such FDA approval for the use of adenosine as a therapeutic for patients suffering from acute myocardial infarction.

60. The NuView website also contained an "Infusion Protocol Chart" describing an "Adenosine (3 mg/ml) Infusion Protocol for Acute Myocardial Infarction" which involves intravenously pumping an undiluted preparation of NuView's compounded adenosine into a patient "to achieve a dose of 70 [micrograms]/kg/min."

61. A patient administered an adenosine infusion according to the "Infusion Protocol Chart" would receive a total volume of between 210 and 588 milliliters of adenosine. The unapproved adenosine product was not, however, offered for sale in

packaging adapted for the administration of such large amounts of adenosine. Instead, it was offered in smaller vials suited for use as an unauthorized replacement for Adenoscan in MPI procedures.

62. A document on the NuView Website titled "NuView Product Order Form" offered "Compounded Adenosine 3 mg/ml" in configurations of 10 x 50 ml vials per box or 20 x 25 ml vials per box for $1500, with a notation "Signature required, as directed for cardiac reperfusion injury."

63. The "Product Order Form" from the NuView website also included spaces to provide a "Patient Name" associated with each order and states "Prescription filled by AnazaoHealth Corporation™."

64. According to a January 8, 2008 letter from counsel for Anazao to in-house counsel for Astellas, NuView and Anazao entered into an agreement whereby NuView marketed adenosine for Anazao. On information and belief, Anazao was aware of NuView's practices in marketing adenosine.

65. Anazao had its own website, www.anazaohealth.com, on which it advertised "Custom Pharmacy" services.

66. The Anazao website described Anazao's Custom Pharmacy Services as follows: "AnazaoHealth creates customized medications and compounds pharmaceuticals, dietary supplements and hormones for individual patients under physician care. Our custom-compounded therapeutics are individually tailored to meet the special needs of your patients."

67. The Anazao website also contained an order form for adenosine at a concentration of 3 mg/ml with the notation "Sig: As directed for cardiac reperfusion injury."

68. Neither NuView nor Anazao's product order form or website mentioned the use of adenosine for myocardial perfusion imaging.

69. Although the use of adenosine for MPI was not mentioned on the NuView website or on its order form, NuView, through its sales representatives, actively

induced the use of adenosine prepared by Anazao as a pharmacologic stress agent in MPI and Anazao provided its adenosine product for that use. On information and belief, Anazao was aware of NuView's actions inducing the use of its adenosine for MPI.

70. More specifically, the NuView defendants' sales representatives contacted nuclear technologists at free-standing outpatient imaging clinics throughout the United States, in California, and in this District that perform MPI procedures and offered adenosine made by Anazao as a substitute for Astellas's Adenoscan adenosine product in MPI procedures.

71. On information and belief, NuView and its representatives were aware that these clinics do not provide acute coronary care to patients suffering from myocardial infarction and that consequently, such clinics would not use adenosine for treatment of reperfusion injury as described on the NuView website and in NuView's product literature.

72. By way of example, Anazao has sent a customer invoice to California Heart and Vascular Clinic in El Centro California for the purchase of adenosine, listing NuView as the "salesperson" and describing the product as "Adenosine (Nuclear)." MPI is considered a "nuclear" procedure. Treatment of reperfusion injury is not.

73. In addition, the NuView defendants have encouraged customers to seek Medicaid reimbursement for using NuView's adenosine product in place of Adenoscan for MPI. Specifically, on information and belief, in January 2008, NuView sales representative, Sally Torney, wrote to a physician's office concerning NuView's product as follows:

> I know you had a question about re-imbursement for the NuView adenosine. I spoke with our national sales director (Keith Rustvold) about getting an EOB for insurance purposes and he told me that all you need to do is bill it the same way that you would for the other adenosine. There should be no

questions or issues. Sometimes Medicaid will have a question about it because of the difference in the vial sizes, but adenosine is adenosine, and the price is the driving factor.

74. On information and belief the "other adenosine" referred to is Adenoscan, which is sold and used only for MPI.

75. On information and belief, the NuView defendants are aware of Astellas's patent rights with respect to Adenoscan.

76. Anazao has admitted in a letter written by its counsel to Astellas that "Anazao is aware that Astellas Pharma US, Inc. ("Astellas") is either the owner or exclusive licensee of unexpired patents regarding Adenoscan® adenosine and bearing numbers, U.S. Pat. 5,070,877 and 5,731,296, respectively."

77. On information and belief, NuView's product configuration and references to reperfusion injury on its website and in its ordering and information materials were intended to conceal its active inducement of infringement of the '296 and '877 patents.

78. On information and belief, NuView representative Sally Torney communicated to a physician's office in early 2008 that "Our company (because of patent restraints) can only make a 25 ML and 50 ML vial, but if you look at our prices, they are still more than 50% less than the other adenosine."

79. On information and belief, the adenosine product promoted and sold by NuView and shipped by Anazao was specifically configured for use in MPI and both NuView and Anazao were aware that the product would be used for MPI by the customers at nuclear imaging clinics where the product is shipped in violation of Astellas's patent rights.

80. NuView's sales representatives have made sales contacts both in person and by telephone at clinics that use Astellas's Adenoscan product in MPI procedures.

81. As part of these sales contacts, NuView's sales representatives have assured Astellas's customers that NuView's product is identical to Adenoscan, even though the NuView/Anazao product is not approved by the FDA for use in MPI and

neither the product nor the manufacturing facility has been subject to the same stringent controls and regulations as Astellas's Adenoscan.

82. In face-to-face sales calls on nuclear technologists, NuView's sales representatives have also made direct price comparisons to Adenoscan, encouraging Astellas's customers to switch to adenosine made by Anazao based on its lower price per milligram of solution.

83. On information and belief, at the time of these sales calls, NuView was aware that the customers were regularly purchasing Adenoscan from Astellas and would continue to do so absent NuView's actions.

84. As a result of NuView's activities in California and this district, physicians' offices have ceased regular purchases of Adenoscan and have instead purchased adenosine made by Anazao and used it in MPI procedures in violation of Plaintiffs' patent rights.

NUVIEW'S VIOLATIONS OF CALIFORNIA LAW

85. Chapter 9, Sec. 4000 *et seq*. of the California Business and Professions Code contains the California Pharmacy Law.

86. Adenosine is classified as a "dangerous drug" under CBPC § 4022.

87. California Business and Professions Code § 4076 requires that a "pharmacist shall not dispense any prescription except in a container that meets the requirements of state and federal law and is correctly labeled" with various types of information, including the directions for the use of the drug and the name of the patient.

88. California Business and Professions Code § 4077 states that, except under certain specific circumstances, "no person shall dispense any dangerous drug upon prescription except in a container correctly labeled with the information required by Section 4076."

89. California Business and Professions Code § 4078 states that "[n]o person shall place a false or misleading label on a prescription" and that "[n]o prescriber shall

direct that a prescription be labeled with any information that is false or misleading."

90. On information and belief, NuView and Anazao's product was distributed with a false or misleading label that indicated the drug was for use in "reperfusion injury" rather than the intended use of MPI and NuView and Anazao distributed with the product a "Medical Professional Information Sheet" that falsely and misleadingly described dosing and information for use of adenosine in inhibiting "reperfusion injury" rather than MPI, the use for which the product was sold and actually employed.

91. On information and belief, NuView is not licensed as a pharmacy or a pharmacy wholesaler in the State of California, in violation of at least California Business and Professions Code § 4160.

92. On further information and belief, the NuView defendants unlawfully advertised the pharmacy services of Anazao in violation of California Business and Professions Code § 4340.

93. NuView and Anazao's use of false and misleading labeling information, including its distribution of a false and misleading "Medical Professional Information Sheet" also violated at least California Health and Safety Code §§ 111330, 111440, 111445, and 111450 which prohibit misbranding of drugs with false or misleading labeling and further prohibit sale and delivery of such misbranded drugs.

94. Because NuView sold adenosine for use in MPI procedures and not for reperfusion injury therapy as described on their websites and in their product literature, the website statements and product literature constituted false advertising under at least California Health and Safety Code §§ 110390, 110395, 110398, 110400.

95. NuView's website statements and product literature further constituted false advertising under California Business and Professions Code § 17500.

96. NuView also violated California Health and Safety Code § 110403, which prohibits any person from advertising any drug "represented to have any effect in . . . [h]eart and vascular diseases" unless such advertisements are limited to medical professionals or if the drug is approved for the particular curative or therapeutic effect

advertised. NuView advertised the alleged effects of their adenosine product in treatment of cardiac "reperfusion injury" through product literature and statements on their respective websites that are not limited in their audience to medical professionals.

NUVIEW'S FALSE AND MISLEADING COMPARISONS TO ADENOSCAN

97. NuView, through its representatives, has repeatedly compared its adenosine product made by Anazao to Astellas's Adenoscan product and assured potential customers that the two products are interchangeable.

98. The adenosine-based drug product advertised and sold by NuView and Anazao was one that Anazao formulated itself under a practice known as "pharmacy compounding" and constituted an unapproved drug.

99. By definition, pharmacy compounding "involves making a new drug whose safety and efficacy have not been demonstrated with the kind of data that FDA ordinarily would require in reviewing a new drug application." *Federal and State Role in Pharmacy Compounding and Reconstitution: Exploring the Right Mix to Protect Patients, Hearing Before the Comm. on Health Educ., Labor and Pensions,* 108th Cong. 39 (2004) (Statement of Steven K. Galson, M.D., M.Ph., Acting Director, Center for Drug Evaluation and Research, U.S. Food and Drug Administration, Department of Health and Human Services) (hereinafter "*Galson Statement*"); *see also* FDA's Compliance Policy Guide, Section 460.200 (June 22, 2002) (http://www.fda.gov/ora/compliance_ref/cpg/cpgdrg/cpg460-200.html). Nor has the FDA determined that compounded drug products have been manufactured under rigorous good manufacturing practice requirements.

100. The FDA frowns upon pharmacy compounding of commercially available FDA drug products such as Astellas's Adenoscan product. "[C]opying commercially-approved products in compounding pharmacies circumvents important public health requirements and undermines the drug approval process--the evidence based system of drug review that consumers and health professionals rely on for safe and effective

drugs." *Galson Statement* at 40.

101. NuView's misleading marketing and promotion of Anazao's adenosine drug product resulted in deception and potential harm to patients, the ultimate consumers of the product. Patients were unlikely to know that they were being administered an unapproved copy rather than an authentic FDA-approved drug and any cost savings associated with the lower priced NuView and Anazao product were unlikely to be passed along to patients.

102. While substitution of "generic" drugs for a branded alternative is common and is sanctioned under California Law, Anazao's product was not a "generic" drug in that it is not subject to the FDA approval process for generic drugs and had not been manufactured in an FDA inspected and regulated facility for the manufacture of generic drugs. To the extent that the NuView defendants referred to or sought to create the impression that their unapproved adenosine product was a "generic" equivalent of Adenoscan, such actions were false and misleading.

NUVIEW AND MTS'S ACTIVITIES IN CALIFORNIA

103. MTS's website describes MTS as "a medical-surgical healthcare product distribution company."

104. Among the products offered by MTS is a product described as "Adenosine."

105. The MTS adenosine product is approved by the FDA for the treatment of paroxysmal supraventricular tachycardia ("PSVT"), which refers to a condition where the heart experiences an abnormal conduction of electricity that causes the atrium, and secondarily the ventricles, to beat very rapidly.

106. MTS's adenosine product is not approved by the FDA as a pharmacologic stress agent for use in conjunction with MPI. Therefore, pursuant to FDA regulations, the label for MTS's adenosine product does not identify MPI as an approved use and does not provide any directions for using the product in MPI.

107. On information and belief, the Nuview defendants and the MTS defendants are actively promoting and inducing the use of adenosine as a pharmacologic stress agent in MPI, *i.e.*, for an "off-label" use not approved by the FDA.

108. More specifically, the Nuview defendants and the MTS defendants have contacted free-standing outpatient imaging clinics in this District and elsewhere that perform MPI procedures and offered adenosine as a substitute for Astellas's Adenoscan adenosine product in MPI procedures.

109. On information and belief, as a result of the Nuview defendants' and the MTS defendants' activities, physicians in California and elsewhere have used or will use adenosine sold by NuView or MTS as a substitute for Adenoscan in MPI.

110. On information and belief, the Nuview defendants and the MTS defendants are aware of Astellas's patent rights with respect to Adenoscan and the use of adenosine in MPI.

111. On information and belief, the Nuview defendants and the MTS defendants know or should have known that their actions have and will cause MTS's adenosine product to be used for MPI by the customers at nuclear imaging clinics where the product is shipped in violation of Astellas's patent rights.

COUNT I

INDUCEMENT OF INFRINGEMENT OF U.S. PATENT NOS. 5,731,296 AND 5,070,877 BY THE NUVIEW DEFENDANTS AND THE MTS DEFENDANTS

112. Paragraphs 1-111 are incorporated herein by reference.

113. The use of continuous adenosine infusion for MPI infringes one or more claims of the '296 patent and one or more claims of the '877 patent. Thus, on information and belief, Defendants' customers have directly infringed the '296 and '877 patents by using adenosine purchased from Defendants in MPI.

114. The NuView defendants and the MTS defendants have actively and

knowingly aided and abetted the direct infringement of the '296 and '877 patents.

115. Aware of Astellas's patent rights, the NuView defendants and the MTS defendants have actively and knowingly induced infringement of the '296 and '877 patents by intentionally encouraging the use of, offering for sale, and selling adenosine as a substitute for Adenoscan in MPI.

116. Even if customers were to use adenosine to treat cardiac reperfusion injury by continuous infusion at 70 mcg/kg/min according to the methods set forth on the websites of Anazao and NuView and in their product literature, such use would infringe one or more claims of the '296 patent.

117. Through at least their website solicitations and corresponding product literature, the NuView defendants thus actively and knowingly induced and encouraged direct infringement of the '296 patent in violation of 35 U.S.C. § 271(b) even to the extent that NuView contends that any of their customers actually used their adenosine product for treatment of cardiac reperfusion injury rather than MPI.

118. As a consequence of these infringing activities by MTS and NuView, Plaintiffs have been damaged in an amount not yet determined.

119. Plaintiffs will continue to be damaged as a consequence of Defendants' infringing activities unless those activities are preliminarily and permanently enjoined.

COUNT II

CONTRIBUTORY INFRINGEMENT OF U.S. PATENT NOS. 5,731,296 AND 5,070,877 BY THE NUVIEW DEFENDANTS

120. Paragraphs 1-119 are incorporated herein by reference.

121. The activities of the NuView defendants in marketing adenosine compounded by Anazao for use in MPI also constitute contributory infringement of one or more claims of the '296 patent and one or more claims of the '877 patent under 35 U.S.C. § 271(c).

122. The NuView defendants have offered for sale and sold an adenosine

product for use in practicing the patented methods claimed in the '296 and '877 patents, which use constitutes a material part of the claimed inventions, knowing that the adenosine product is especially made or adapted for use in infringing the patents, and that the adenosine product is not a staple article or commodity of commerce suitable for substantial noninfringing use. On information and belief, NuView's customers have directly infringed the '296 and '877 patents by using adenosine purchased from NuView in MPI.

123. On information and belief, there is no substantial use of NuView's unapproved adenosine product for reperfusion injury and certainly no such use by the customers to whom defendants direct their adenosine sales efforts.

124. Even if customers were to use adenosine to treat cardiac reperfusion injury by continuous infusion at 70 mcg/kg/min according to the methods set forth on the websites of Anazao and NuView and in their product literature, such use would infringe one or more claims of the '296 patent.

125. Thus, even if NuView were to prove a substantial use of adenosine to treat cardiac reperfusion injury, that use would still fall within the scope of one or more claims of the '296 patent and would not constitute a substantial noninfringing use.

126. As a consequence of these infringing activities by the NuView defendants, Plaintiffs have been damaged in an amount not yet determined.

127. Plaintiffs will continue to be damaged as a consequence of the NuView defendants' infringing activities unless those activities are preliminarily and permanently enjoined.

COUNT III

WILLFUL PATENT INFRINGEMENT OF U.S. PATENT NOS. 5,731,296 AND 5,070,877 BY THE NUVIEW DEFENDANTS AND THE MTS DEFENDANTS

128. Paragraphs 1-127 are incorporated herein by reference.

129. In proceeding to make, offer, and sell their unauthorized compounded version of Adenoscan, the NuView defendants have knowingly and recklessly disregarded the '296 and '877 patents and Astellas's exclusive marketing rights under that patent.

130. In proceeding to make, offer, and sell adenosine for use in MPI, the NuView defendants and the MTS defendants have knowingly and recklessly disregarded the '296 and '877 patents and Astellas's exclusive marketing rights under that patent.

131. Defendants' activities constitute willful patent infringement warranting imposition of treble damages under 35 U.S.C. § 284.

132. Defendants' flagrant disregard of the '296 and '877 patents makes this an exceptional case warranting imposition of attorney fees against MTS and NuView under 35 U.S.C. § 285.

COUNT IV

INTENTIONAL INTERFERENCE WITH PROSPECTIVE ECONOMIC ADVANTAGE BY THE NUVIEW DEFENDANTS AND THE MTS DEFENDANTS

133. Paragraphs 1-132 are incorporated herein by reference.

134. MTS and NuView had knowledge of the existing business relationships between Astellas and Astellas's customers that would likely have resulted in future economic benefit to Astellas. Despite this knowledge, MTS and NuView intentionally interfered with these relationships by proceeding to market, offer for sale, and sell to Astellas's customers adenosine for use in MPI according to business practices that are prohibited by California Law.

135. MTS and NuView's actions have caused actual disruption of Astellas's business relationships and resultant economic harm to Plaintiffs in an amount not yet determined.

136. Plaintiffs will continue to be damaged as a consequence of Defendants' actions unless those actions are preliminarily and permanently enjoined.

COUNT V

UNFAIR COMPETITION BY THE NUVIEW DEFENDANTS UNDER THE CALIFORNIA BUSINESS AND PROFESSIONS CODE §17200, *ET SEQ.* AND CALIFORNIA BUSINESS AND PROFESSIONS CODE § 4381

137. Paragraphs 1-136 are incorporated herein by reference.

138. In marketing, offering for sale, and selling its unauthorized compounded adenosine for continuous infusion for MPI to customers as described herein, NuView engaged in unlawful, unfair or fraudulent business practices in violation of the Business and Professions Code § 17200, et seq.

139. As set forth in detail above, the NuView defendants' unlawful conduct has included numerous violations of California law, including violations of California Pharmacy Law.

140. Violations of California Pharmacy Law constitute unfair competition pursuant to Business and Professions Code § 17200, et seq. and California Business and Professions Code § 4381.

141. The NuView defendants' conduct, even where not unlawful, offends public policy as it has been established by statutes and the common law. As set forth above, the NuView defendants' scheme to develop and perpetuate an elaborate ruse concerning the use of adenosine to treat cardiac reperfusion injury to deflect attention from the unlawful and infringing acts of marketing and providing unapproved adenosine for use in MPI is immoral, unethical, unscrupulous and substantially injurious to consumers as well as to Plaintiffs.

142. As a consequence of the NuView defendants actions, including their violation of California Business and Professions Code § 4381, Plaintiffs have suffered damages in an amount not yet determined.

143. Plaintiffs will continue to be injured as a consequence of Defendants' actions unless those actions are preliminarily and permanently enjoined.

COUNT VI

UNFAIR COMPETITION BY THE NUVIEW DEFENDANTS AND THE MTS DEFENDANTS UNDER THE CALIFORNIA BUSINESS AND PROFESSIONS CODE §17200, *ET SEQ.*

144. Paragraphs 1-143 are incorporated herein by reference.

145. In marketing, offering for sale, and selling an adenosine product labeled for treatment of PSVT for continuous infusion in MPI to customers as described herein, the NuView defendants and the MTS defendants have engaged in unlawful, unfair or fraudulent business practices in violation of the Business and Professions Code § 17200, et seq.

146. Defendants' off-label promotion and sale of adenosine labeled for treatment of PSVT as a substitute for Adenoscan in MPI constitutes a violation of California law including at least California Health and Safety Code § 111440, which prohibits the sale, offer for sale or delivery of misbranded drugs, and § 111375, which requires adequate directions for the use of any drug.

147. Defendants' off-label sale of adenosine labeled for treatment of PSVT as a substitute for Adenoscan in MPI is immoral, unethical, oppressive, unscrupulous, and/or substantially injurious to consumers, and further contravenes public policy as established by statutes and common law.

148. As a consequence of Defendants' actions, Plaintiffs have suffered damages in an amount not yet determined.

149. Defendants' actions constitute unfair competition pursuant to Business and Professions Code § 17200, et seq.

150. Plaintiffs will continue to be injured as a consequence of Defendants' actions unless those actions are preliminarily and permanently enjoined.

REQUEST FOR RELIEF

WHEREFORE, Plaintiffs pray for judgment against Defendants as follows:

A. that Defendants have actively induced infringement and/or committed acts of contributory infringement with respect to one or more claims of the '296 and '877 patents;

B. that Defendants, their officers, agents, attorneys, and employees, and those acting in privity or concert with them or any of them, be preliminarily and permanently enjoined from further acts of infringement;

C. that Plaintiffs have been irreparably harmed by Defendants' infringement of the '296 and '877 patents;

D. that Defendants' infringement of the '296 and '877 patents was willful and deliberate;

E. that Defendants be ordered to account for and pay to Astellas, Item, and King all damages caused to Astellas, Item and King by reason of infringement of the '296 and '877 patents, including Astellas's lost profits, and that such damages be trebled by reason of the deliberate and willful infringement of the '296 and '877 patents pursuant to 35 U.S.C. § 284;

F. that Astellas, Item, and King be granted reasonable attorney fees pursuant to 35 U.S.C. § 285;

G. that Defendants have unfairly competed with Plaintiffs in violation of the California Business and Professions Code § 17200;

H. that Defendants, their officers, agents, attorneys, and employees, and those acting in privity or concert with them or any of them, be preliminarily and permanently enjoined pursuant to California Business and Professions Code § 17203 from further violation of the California Business and Professions Code § 17200 and that Defendants be ordered to disgorge moneys received by virtue of their unfair practices;

I. that the NuView defendants have violated California Business and Professions Code § 4381;

J. that Plaintiffs should be awarded actual and treble damages, reasonable attorneys' fees and costs of suit pursuant to California Business and Professions Code § 4381(b) and (c);

K. that Defendants be preliminarily and permanently enjoined from further violation of the California Pharmacy Law pursuant to California Business and Professions Code § 4381(b) and (c);

L. that compensatory and punitive damages be awarded against Defendants in favor of Astellas, Item, and King for Defendant's intentional interference with prospective economic advantage; and

M. that Plaintiffs be granted such other and further relief as the case may require and the Court may deem just and proper, together with costs in this action.

Dated: November 20, 2008

MORRIS POLICH & PURDY LLP

By _____
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UNITED STATES DISTRICT COURT

CENTRAL DISTRICT OF CALIFORNIA – WESTERN DIVISION

ASTELLAS US LLC, ASTELLAS PHARMA US, INC., ITEM DEVELOPMENT AB, AND KING PHARMACEUTICALS RESEARCH AND DEVELOPMENT, INC.

Plaintiffs,

v.

ANAZAOHEALTH CORPORATION, NUVIEW RADIOPHARMACEUTICALS, INC., PAUL J. CROWE, KEITH RUSTVOLD, MTS HEALTH SUPPLIES, INC., NABIL SABA, AND GHASSAN SALAYMEH

Defendants,

Case No. CV08-01083 JFW (VBKx)

DECLARATION RE JOINDER PURSUANT TO LOCAL RULE 19-2

NUVIEW RADIOPHARMACEUTICALS, INC., PAUL J. CROWE, AND KEITH RUSTVOLD

Counter Claimants,

v.

ASTELLAS US LLC, ASTELLAS PHARMA US, INC., ITEM DEVELOPMENT AB, AND KING PHARMACEUTICALS RESEARCH AND DEVELOPMENT, INC.

Counter Defendants,

NUVIEW RADIOPHARMACEUTICALS, INC., PAUL J. CROWE, AND KEITH RUSTVOLD

Cross Claimants,

v.

ANAZAOHEALTH CORPORATION

Cross Defendant.

DECLARATION OF COUNSEL UNDER LOCAL RULE 19-2

This action involves causes of action for infringement of U.S. Patent Nos. 5,070,877 (the '877 Patent) and 5,731,296 (the '296 Patent). Although plaintiff King Pharmaceuticals Research and Development, Inc. (King) owns the '877 patent and plaintiff Item Development AB (Item) owns the '296 patent, both patents are exclusively licensed to plaintiffs Astellas US LLC and Astellas Pharma US, Inc. (collectively Astellas) who control the enforcement of the two patents and have the right to bring suit under the patents. King and Item are joined as co-plaintiffs to avoid a dispute over necessary parties.

The interests of justice will be advanced, and a multiplicity of actions avoided, by allowing these causes of action to proceed together in a single lawsuit because the infringement claims under both patents arise from a common nucleus of facts. Both

patents cover different aspects of the method of using Astellas's branded pharmaceutical product, Adenoscan®, as a pharmacologic stress agent to diagnose cardiac disease.

As set forth in detail in the complaint, Plaintiffs' causes of action relate, in part, to the NuView defendants' unauthorized sales of an unapproved copy of Adenoscan® whose use infringes both of the patents licensed by Astellas. Moreover, the NuView defendants' efforts to hide their infringement of both these patents through an elaborate ruse gives rise to Plaintiffs' actions for unfair competition and intentional interference with prospective economic advantage. Separating the two patent actions and requiring them to be heard individually would result in duplication of effort by the Court because the infringing activities arise out of the same transactions for both patents. Moreover, much of the conduct at issue in the unfair competition and interference with prospective economic advantage counts is the same conduct at issue in the patent infringement counts.

Finally, the technological background concerning adenosine, its physiological effects, and its use in humans is relevant to both patents.

I declare under penalty of perjury under the laws of the United States of America that the foregoing is true and correct to the best of my knowledge, information, and belief.

Executed on November 20, 2008, in Los Angeles, California.

Donald L. Ridge

EXHIBIT A

US005731296A

United States Patent [19]

Sollevi

[11] Patent Number: 5,731,296

[45] Date of Patent: Mar. 24, 1998

[54] **SELECTIVE VASODILATION BY CONTINUOUS ADENOSINE INFUSION**

[75] Inventor: **Alf Sollevi**, Bromma, Sweden

[73] Assignee: **Item Development AB**, Stocksund, Sweden

[21] Appl. No.: **31,666**

[22] Filed: **Mar. 15, 1993**

Related U.S. Application Data

[62] Division of Ser. No. 821,395, Jan. 14, 1992, Pat. No. 5,231,086, which is a continuation of Ser. No. 630,413, Dec. 19, 1990, Pat. No. 5,104,859, which is a continuation of Ser. No. 138,306, Dec. 28, 1987, abandoned, which is a continuation-in-part of Ser. No. 30,245, Mar. 24, 1987, abandoned, which is a continuation-in-part of Ser. No. 779,516, Sep. 24, 1985, abandoned.

[51] **Int. Cl.**[6] **A61K 31/70**

[52] **U.S. Cl.** **536/46**; 514/47; 536/27.6

[58] **Field of Search** 514/46

[56] **References Cited**

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Primary Examiner—John Kight
Assistant Examiner—L. Eric Crane
Attorney, Agent, or Firm—White & Case

[57] **ABSTRACT**

This invention is concerned with the use of adenosine as an agent for the treatment of human beings. More particularly, this invention is concerned with the administration of adenosine to human patients by continuous intravenous infusion for, inter alia, control of blood pressure, use as a selective vasodilator, decreasing pulmonary vascular resistance, treating acute pulmonary hypertension in conjunction with idiopathic respiratory distress syndrome, in diagnosing pulmonary hypertension in conjunction with cardiac septum defects, in percutaneous transluminal angioplasty (PTCA), in coronary thrombolysis (CTL) and in radionucleide scintography.

9 Claims, No Drawings

SELECTIVE VASODILATION BY CONTINUOUS ADENOSINE INFUSION

This application is a divisional of Ser. No. 07/821,395, filed Jan. 14, 1992, now U.S. Pat. No. 5,231,086, which is a continuation of Ser. No. 07/630,413, filed Dec. 19, 1990, now U.S. Pat. No. 5,104,859, which is a continuation of Ser. No. 138,306, filed Dec. 28, 1987, now abandoned, which is a continuation-in-part of Ser. No. 030,245, filed Mar. 24, 1987, now abandoned, which is a continuation-in-part of Ser. No. 779,516, filed Sep. 24, 1985, now abandoned.

This invention is concerned with the use of adenosine as an agent for the treatment of human beings. More particularly, this invention is concerned with the administration of adenosine to human patients by continuous intravenous infusion for, inter alia., control of blood pressure, use as a selective vasodilator, decreasing pulmonary vascular resistance, treating acute pulmonary hypertension, treating pulmonary hypertension in conjunction with idiopathic respiratory distress syndrome, and in diagnosing pulmonary hypertension in conjunction with cardiac septum defects.

Adenosine is a naturally occurring nucleoside composed of the purine, adenine, and the sugar, D-ribose. Normal basal plasma levels of adenosine are from about 0.1 to about 0.2 μmol per liter. In addition, it is commonly present in the body in the form of adenosine monophosphate (AMP), adenosine diphosphate (ADP) and adenosine triphosphate (ATP). Adenosine has been reported to have a variety of biological effects, depending on whether the adenosine is endogenous or exogenously administered, including sedative and anti-epileptic effects on the central nervous system and inhibitory effects on respiration, cardio-vascular effects, including prolongation of atrio-ventricular conduction time and impulse formation in the sinus node, vasodilation, antiaggregatory effect, decreased release of free fatty acids, anti-secretory effect in the stomach, and anti-diuretic effect.

As a general rule, however, adenosine and its biological effects have been largely of physiological interest. To the extent adenosine was of interest as a pharmaceutical product, that interest has centered primarily on its phosphate derivative, which now is known to be rapidly metabolized to yield adenosine and phosphate in the circulation. See Sollevi et al., Acta. Physiol. Scand. 120:171–6 (1984). However, phosphate may cause undesired side effects. For example, high levels of phosphate may cause arrhythmias secondary to chelation of magnesium and calcium. (See Dedrick, et al., Anesthesiology, 57:3A, 66 (1982)).

Furthermore, adenosine is known to produce heart block through blockage of the atrioventricular (A-V) node. As a consequence, methylxanthines such as theophylline have been proposed by Berne, et al., in U.S. Pat. No. 4,364,922 for use in preventing heart block caused by adenosine, in particular adenosine released as a consequence of cardiac ischemia or hypoxia.

In addition, it has been proposed to take advantage of adenosine's ability to block atrioventricular conductance by using it to treat tachyarrhythmias. For such use, adenosine is administered as an injectable intravenous bolus containing from about 37.5 micrograms/kg up to about 45.0 micrograms/kg of adenosine. In such a use, the adenosine has little detectable vasodilating action. Adenosine has a very short plasma half-life, of the order of 10–20 seconds (see, Fredholm and Sollevi, J. Physiol., 313:351–62 (1981)), and thus the concentration of injected adenosine is rapidly reduced to normal serum levels (about 0.15μ mol per liter). The transitory presence of the injected adenosine precludes all but the most transitory vasodilation.

Accordingly, for adenosine to be of practical value for use as a vasodilator, it must be administered continuously to maintain plasma levels sufficiently high to achieve vasodilation. The problem, however, is that such continuous administration could lead to undesired side effects, such as the above-noted heart blockage.

It also should be noted that compounds commonly used as vasodilators, such as sodium nitroprusside, nitroglycerine, isoflurane, hydralazine, prazosin and the like, have various side effects. For example, sodium nitroprusside has the drawbacks of tachyphylaxis and rebound hypertension, apparently caused by autogenous generation of angiotensin to counteract the hypotensive effect of the nitroprusside. As a consequence, the dosage of nitroprusside must be progressively increased with continued use to overcome the hypertensive effect of angiotensin, and there is a risk of rebound due to the presence of residual excess angiotensin. Nitroglycerine and prazosin suffer from the drawbacks of slow onset and unpredictable action. Isoflurane and sodium nitroprusside both have a tendency to reduce cardiac blood flow, while nitroprusside, hydralazine and prazosin increase heart rate.

Accordingly, there remains a need for a vasodilator suitable for administration by continuous intravenous infusion.

The present invention is based upon the discovery that adenosine can be administered to human patients under conditions such that significant vasodilation is achieved without the occurrence of significant heart blockage. It is based on the further discovery that adenosine has a unique, and heretofore unappreciated, activity profile in humans which differs significantly from the profiles of heretofore commonly used vasodilators. As a consequence of this discovery, it has been discovered that adenosine may be employed for the treatment of a variety of conditions by continuous intravenous infusion techniques. In particular, and as will be illustrated in greater detail below, adenosine has been found to have the following characteristics:

1. It has selective vasodilation activity, in that its effect is limited to a cardiac after-load effect. That is, its activity is limited to dilation of arteries and it has little or no effect on cardiac pre-load, i.e. as a dilator of veins.

2. Although adenosine has significant action in blocking atrio-ventricular (A-V) conductance by bolus injection, it can be administered by continuous infusion and have significant useful vasodilating action at dosages below those at which it has significant A-V activity.

3. Adenosine has significant hypotensive activity without the occurrence of significant tachyphylaxis, apparently because adenosine blocks the renin-angiotensin system of the kidney, thus preventing hypertension due to the formation of angiotensin in response to hypotension.

4. Adenosine's effect is readily controlled because it is active at relatively small doses and because of its short plasma half-life (10–20 seconds). In addition, its activity quickly ceases when adenosine administration is terminated.

5. Adenosine is capable of significantly increasing cardiac output without significantly increasing cardiac work.

6. Adenosine, in the amounts used in accordance with the invention, is essentially non-toxic. It is rapidly taken up by the body to form ATP, and upon degradation its metabolites are present at or below levels normally resulting from physical exercise.

The foregoing activity profile permits continuous infusion of adenosine for controlled hypotension during surgery, for control of various forms of hypertensive crisis, to improve coronary circulation during surgery in patients with

ischemic heart disease, for reducing the incidence of coronary graft occlusion by increasing graft flow following coronary bypass surgery, and for reducing platelet loss during cardiac bypass surgery. It has also been found that adenosine may be used for decreasing pulmonary vascular resistance, for treating acute pulmonary hypertension, for treating acute pulmonary hypertension in conjunction with idiopathic respiratory distress syndrome (IRDS) and for diagnosing the operability of the pulmonary vasculature in patients with pulmonary hypertension in conjunction with cardiac septum defects. Adenosine also is useful in inhibiting clot formation during percutaneous transluminal coronary angioplasty (PTCA) and coronary thrombolysis (CTL), as well as an aid in visualizing miocardial irrigation for radionucleotide scintigraphy.

In accordance with this invention, adenosine may be administered to human patients by continuous intravenous infusion to provide significant vasodilation and without significant heart blockage under two conditions. First, the heart blocking action of adenosine is not detected during anesthesia when the rate of administration is 0.35 milligrams of adenosine per kilogram of body weight per minute or less. Second, the heart blocking action of adenosine is not detected, even in conscious patients, at rates of administration of about 0.10 milligram of adenosine per kilogram of body weight per minute or less.

For purposes of this invention, adenosine can be administered to the patient in any pharmaceutically acceptable form suitable for use in continuous, intravenous infusion. A preferred form is an aqueous solution of adenosine, and more preferably adenosine in isotonic saline. The concentration of adenosine in the solution is not narrowly critical, although concentrations of at least about 5 millimol (or about 1.5 milligrams) per milliliter of solution are desired to avoid the need for excessive infusion rates to achieve desired serum levels. When administering adenosine to small children, however it is possible to use concentrations as low as about 0.1 mg/ml. The concentration may be as high as the solubility limit of adenosine (about 20 millimols per liter or 5.5 to 6 milligrams per milliliter), if desired.

When used for continuous infusion in accordance with this invention, the unit dosage form typically has a volume of at least 250 milliliters, and preferably in the range of 250 to 500 milliliters, to provide an adequate supply of adenosine. Consequently, the unit dosage form generally will contain from about 0.4 to about 3 grams of adenosine. In small children, the unit dose will, of course, be correspondingly smaller than it is for adults.

The adenosine solution should be sterile and free from fungi and bacteria. Such solutions have been found to be stable at room temperature for at least two years.

Such solutions are prepared by mixing adenosine with the aqueous carrier, e.g. water or an isotonic solution, and other desired ingredients, to achieve a solution having the desired concentration, and thereafter sterilizing the solution.

Continuous infusion can be performed using any technique known to the art. Because adenosine has such a short plasma half-life and it is active at relatively low concentrations, it is desired that the method be one which minimizes or avoids fluctuations of serum adenosine levels. Accordingly use of high precision roller pumps is preferred.

As is noted above, the present invention has numerous specific applications, depending upon adenosine dosage levels and whether or not adenosine is administered to anesthesized or conscious patients. The first general category of applications is that in which adenosine is continuously administered to a patient undergoing surgery under general anesthesia at doses that do not induce heart block. Specific applications include controlled hypotension during surgery, in particular dissection and clipping of cerebral artecial aneurysms; control of hypertension crisis during surgery, for example due to release of catecholamines in the course of pheochomocytoma surgery; and improved coronary circulation and after-load reduction during abdominal aortic aneurysm surgery, especially in patients with ischemic heart disease. For such uses, dosage rates of the order of 0.05 to about 0.3 milligrams per kilogram of body weight per minute are effective amounts.

The second general category of continuous adenosine infusion applications is that in which adenosine is administered to conscious patients, also at levels below which adenosine exhibits significant heart blocking action. These levels are typically achieved at administration rates of 0.05 milligram of adenosine per kilogram per minute or less. Specific examples of conditions which may be treated with adenosine in conscious patients include prevention of occlusion of cardiac bypass grafts following bypass surgery, increased cardiac output in patients with low cardiac output, and use of adenosine as an adjunct to dopamine treatment for shock.

Blood levels of adenosine which result from an administration rate of about 10–30 micrograms per kilogram of body weight per minute (0.010–0.030 mg. per kg. per minute) minute can be used in a number of additional ways, e.g., to decrease pulmonary vascular resistance, to treat acute pulmonary hypertension and acute pulmonary hypertension in conjunction with idiopathic respiratory distress syndrome (IRDS), and to diagnose pulmonary hypertension in conjunction with cardiac septum defects.

The following examples illustrate in greater detail specific applications of continuous intravenous infusion of adenosine in accordance with this invention.

EXAMPLE I

CONTROLLED HYPOTENSION DURING ANESTHESIA

It is frequently desired to reduce the blood pressure of patients during surgery. For example, in the case of dissection and clipping of cerebral arterial aneurysms, controlled hypotension is desired to reduce the aneurysm wall tension in order to minimize the risk of rupture and bleeding. Controlled hypotension is also used to reduce bleeding during other forms of surgery.

Prior to this invention, vasodilators, such as sodium nitroprusside and nitroglycerine, were used for this purpose, but both have drawbacks. For example, sodium nitroprusside suffers from tachyphylaxis, or the need to increase the dose of nitroprusside with time due to the release of angiotensin. In addition, rebound hypertension also has been observed following use of nitroprusside. Nitroglycerine is characterized by Slow onset of action and unpredictable action.

Adenosine has been found to be a remarkably effective agent for inducing controlled hypotension during surgery. Adenosine, when administered in effective amounts, has a very rapid hypotensive effect which can be rapidly terminated due to its short half-life. Moreover, adenosine does not cause tachyphylaxis, apparently because it blocks the renin-angiotensin system of the kidney, thereby preventing formation of angiotensin which tends to counteract the hypotension. For the same reason, rebound hypertension is avoided after discontinuation of infusion.

For this indication, adenosine typically is administered intravenously via the left basilic vein or via a central vein in

an amount (or at a rate) sufficient to achieve the desired hypotensive effect. It has been found that lowering of mean arterial blood pressure to as low as 40 millimeters of mercury, as measured by a cannula in the left radial artery, is readily achieved without significant side effects. In particular, so long as the patient is under anesthesia, no blockage of atrio-ventricular conductance is observed.

The actual plasma levels of adenosine employed for controlled hypotension will vary, depending upon such factors as the particular patient, the age of the patient and the desired degree of hypotension. As a general rule, however, a reduction of mean arterial blood pressure to 40 to 50 mm Hg is achieved by adminstration of adenosine at a rate of from about 0.2 to about 0.35 milligrams of adenosine per kilogram of body weight per minute. The amount of adenosine required to achieve a given degree of hypotension can be reduced if adenosine uptake inhibitors, such as dipyridamole, are also administered to the patient. The possibility that adenosine might be useful for inducing contolled hypotension in humans was suggested by Kassell et al., J. Neurosurg., 58: 69–76 (1983), based upon tests in dogs. However, this study was performed during the administration of dipyridamole, another vasodilator that potentiates the effect of adenosine by inhibiting cellular uptake of adenosine. The dose of dipyridamole (1 mg/kg) was high, and it in fact induced a 20% reduction of the mean arterial blood pressure. The hypotensive effect of adenosine was then studied upon this hypotensive dose of dipyridamole. It was reported that hypotension to a mean arterial pressure of 40 mm/Hg could be induced and maintained with an infusion of 0.4 gram of adenosine per 100 milliliters of normal saline, at a dose of 50 μg/kg/minute. When dipyridamole was excluded in a pilot study, as much as 5–10 mg/kg/minute was required for the induction of hypotension, thereby creating an excessive fluid load. Kassel et al. noted that induction of hypotension in dogs is difficult, and speculated that "adenosine alone, without the potentiating effects of dipyrimadole, may be sufficient to produce hypotension in man without excessive volumes of fluid". As noted above, effective induction of hypotension in man is achieved at adenosine dose levels of only 0.2 to 0.35 mg/kg/minute, or 30 to 50 times lower levels than effective levels in the dog. Such low rates are hardly predictable from the information of Kassel et al.

In a study intended to demonstrate use of continuous infusion of adenosine to effect controlled hypotension in man, ten patients with no known history of cardiopulmonary diseases (seven men and three women, ages 35–58 years), scheduled for intracerebral aneurysm surgery, were selected.

One hour before the operation the patients were premedicated orally with diazepam (10–20 mg.) Atropine (0.5 mg) and droperidol (0.1 mg. per kg.) were given intravenously before induction of anesthesia. Induction was started with thiopental (5 mg. per kg) followed by phenoperidine (1–2 mg), a synthetic opiate with pharmacodymanics resembling fentanyl but with a longer duration of action and 1/10 of its analgesic potency.

Pancuronium bromide (0.1 mg per kg) was given to facilitate endotracheal intubation. Anesthesia was maintained by supplementary doses of phenoperidine and droperidol, as required. The total dose of droperidol did not exceed 0.2 mg. per kg. and was administered within the first 2 hours of anesthesia. Phenoperidine was supplemented regularly to prevent the blood pressure from exceeding the preanesthetic level (approx. 1 mg/30–60 min). Controlled hyperventilation was employed with a humidified gas mixture of 60% N_2O in oxygen to maintain $PaCO_2$ values at approximately 30 mmHg (±1.5/SSM). Mannitol (1–1.5 g. per kg) was given routinely at the start of the operation (e.g., 1–2 hours prior to the controlled hypotension). The patients were operated on in the horizontal supine position.

A 1.2-mm plastic cannula was introduced into the left radial artery to monitor systemic arterial blood pressure (MABP) and collect arterial blood. A balloon-tipped, flow-directed, quadruple lumen Swan-Ganz catheter (Model 93A-831-7.5 F, VIP) was inserted percutaneously via the left basilic vein, and its correct position in the pulmonary artery was determined by pressure tracings. The catheter was used for the monitoring of mean right atrial pressure (RAP), mean pulmonary artery pressure (PAP), and mean pulmonary capillary wedge pressure (PCWP) for the determination of cardiac output and collection of mixed venous blood and for the infusion of adenosine. Another plastic cannula was introduced percutaneously, in a retrograde direction, into the right jugular bulb for the collection of blood. The correct position was verified by x-ray.

The ECG was monitored with a standard chest (V5) lead. Heart rate was determined from the R-R interval. Blood pressures were measured by transducers placed at the midthoracic level. Cardiac output (QT) was determined in triplicate according to the thermodilution technique with a cardiac output computer (Edwards Lab, model 9510). Isotonic glucose, 10 ml at 1° C., was used as a thermal indicator. The ECG, heart rate, blood pressures, and thermodilution curves were recorded on a Grass® polygraph.

Blood gases were measured with appropriate electrodes for pH, PCO_2, and PO_2 (Radiometer, Copenhagen). The hemoglobin concentration was determined spectrophotometrically. Samples for the determination of adenosine and its metabolites were collected as described by Sollevi et al., Acta Physiol. Scand., 120: 171–76 (1984). Adenosine and inosine were purified and analyzed by HPLC as described by Fredholm and Sollevi., J. Physiol. (London), 313: 351–67 (1981). Hypoxanthine, xanthine, and uric acid were analyzed by HPLC according to the method of Schweinsberg and Loo., J. Chromatogr., 181: 103–7 (1980). Arterial levels of dipyridamole were determined by HPLC. J. Chromatogr., 162: 98–103 (1979). Blood lactate was measured according to Tfelt-Hansen and Siggard-Andersen. Scand. Clin. Lab. Invest., 27:15–19 (1971).

Measurements and blood samplings were performed immediately before hypotension, as late as possible during hypotension (1–5 min before terminating the infusion) and approximately 30 min after the hypotensive period.

Dipyridamole (5 mg. per ml) was infused iv (0.3–0.4 mg. per kg. over a period of 5–10 min) approximately 20 minutes prior to the induction of controlled hypotension. This dose of dipyridamole produced clinically relevant drug levels in the plasma (1.2±0.3 μM, SEM) during the hypotensive periods. (See Pedersen, J. Chromatogr., 162: 98–103 (1979).

Adenosine (5 mM, 1.34 mg. per ml in isotonic saline) was administered by continuous infusion (Critikon roller pump, 2102A, superior vena cava) for 12–71 minutes ($\bar{x}$=33±3 SEM) at a rate of 0.01–0.32 mg. per kg per min ($\bar{x}$=0.14±0.04 SEM, corresponding to 8.0±2.7 mg. per min). The infusion was started at a rate of 0.01 mg. per kg. per minute, which was doubled at 15 second intervals until the desired MABP level of 40–50 mmHg was reached. The corresponding volume of infused adenosine solution ranged from 0.5 to 17 ml. per min ($\bar{x}$=6±2 SEM). The mean hypotensive period was 32±8 min. The total adenosine dose did not exceed 1.5 grams. Serum creatinine was determined before and on two consecutive days after operation. The standard ECG was recorded the day before and the day after operation.

Systemic vascular resistance (SVR) was derived from the formula

$$SVR(\text{mmHg. per liter min}) = \frac{MABP - RAP}{QT}$$

and pulmonary vascular resistance (PVR) from the formula

$$PVR = \frac{PAP - PCWP}{QT}.$$

Oxygen content was derived from the formula SO_2*× 1.34×Hb+PO_2×0.03. Foex et al., Br. J. Anesth., 42: 803–4 (1970). The arteriovenous oxygen content difference ($AVDO_2$) was determined and used to calculate total oxygen consumption (VO_2) as the product of $AVDO_2$ and QT.

*SO_2=Oxygen saturation.

The results of this work are summarized in Tables I–IV, in which data are presented as means ±SEM. The statistical significance (control 1 vs. adenosine and control 1 vs. control 2) was determined by Student's t test for paired data. A P value of <0.05 was regarded as significant.

The purine levels of nine of the patients were determined prior to, during and after adenosine-induced controlled hypotension. The results are summarized in Table I.

TABLE I

Purine Levels (μM) in Arterial Plasma Before, During, and After Adenosine-induced Controlled Hypotension in Nine Patients.

			After Hypotension	
	Control 1	Hypotension	3–9 Min	20–40 Min
Adenosine	0.15 ± 0.02 (9)	2.45 ± 0.65 (9)	0.24 ± 0.06 (6)	0.19 ± 0.03 (7)
Inosine	0.04 ± 0.01 (8)	3.01 ± 1.48 (8)	0.69 ± 0.26 (5)	0.29 ± 0.15 (7)
Hypoxanthine	1.94 ± 0.53 (9)	6.28 ± 2.33 (9)	—	3.25 ± 1.00 (8)
Xanthine	5.93 ± 2.04 (7)	6.10 ± 1.67 (7)	—	3.49 ± 1.56 (7)
Uric Acid	185.2 ± 14.2 (9)	198.1 ± 22.7 (9)	—	199.5 ± 28.8 (8)

*P < 0.01
**P < 0.05, denotes significantly different from the control 1 levels.
Number of observation in parentheses.

As is evident from Table I, adenosine is present in the 10^{-7}M range during basal conditions. Continuous infusion of adenosine increased the arterial adenosine level to 2.45±0.65 μM. The adenosine metabolites inosine and hypoxanthine were increased during the infusion, whereas xanthine and uric acid levels were unaffected. Once the desired blood pressure level was reached, the infusion rate could be kept constant throughout the hypotensive period. After termination of the infusion, the arterial adenosine levels returned to control values within 3–9 min. Inosine was eliminated more slowly from the circulation and remained slightly above basal levels 20–40 minutes after the infusion.

The central hemodynamic variables were measured in all 10 patients before, during and 30 minutes after controlled hypotension and are summarized in Table II.

TABLE II

Central Hemodynamic Variables Before, During and 30 Min After Adenosine-induced Controlled Hypotension in Ten Patients.

	Control 1	Adenosine	Control 2
Systolic blood pressure (mmHg)	115 ± 6	78 ± 6*	130 ± 7**
Diastolic blood pressure (mmHg)	61 ± 3	34 ± 2*	71 ± 3**
Mean arterial blood pressure (mmHg)	82 ± 3	46 ± 2*	91 ± 4**
Right atrial pressure (mmHg)	6.8 ± 1.1	7.2 ± 0.7	5.6 ± 0.8
Pulmonary artery pressure (mmHg)	14.4 ± 1.0	16.5 ± 0.9	14.7 ± 1.4
Pulmonary capillary wedge pressure (mmHg)	9.6 ± 1.1	11.0 ± 1.0	9.9 ± 1.4
Heart rate (beats per min)	54 ± 2	69 ± 3*	58 ± 3
Cardiac output (liters per min)	4.93 ± 0.51	6.86 ± 0.71*	5.17 ± 0.54
Systemic vascular resistance (mmHg per min)	16.65 ± 1.95	6.22 ± 0.60*	17.70 ± 1.72
Pulmonary vascular resistance (mmHg per liter min)	0.97 ± 0.15	0.82 ± 0.08	0.97 ± 0.15

*P < 0.01
**P < 0.05, denotes significantly different from control 1.

The infusion of dipyridamole decreased MABP by approximately 10 mmHg in five of the patients. At the start of the adenosine infusion, MABP was not significantly different from the predipyridamole level (82±3 vs. 86±3 mmHg) as shown in Table II. Adenosine induced a decrease in MABP to 46 mmHg (43±3%) within 1–2 minutes.

The decrease in MABP was caused by a parallel decrease in both systolic and diastolic pressure. The MABP was stable throughout the hypotensive period. Cardiac output increased from 4.9 to 6.9 l per minute (44±9%) in parallel with a small increase in heart rate of 9±2 beats per minute. The SVR decreased from 16.7 to 6.2 mmHg per liter a minute, corresponding to a decrease of 61±3%, whereas PVR was unchanged. RAP, PAP, and PCWP were not influenced by adenosine.

After discontinuation of the infusion, MABP was restored within 1–5 minutes. Rebound hypertension did not occur, although the MABP was persistently approximately 10 mmHg higher after hypotension than during the control period. However, the posthypotensive MABP was not significantly higher than the MABP before administration of dipyridamole. Heart rate, QT, and SVR returned rapidly to control levels concurrently with the restoration of MABP.

Oxygen contents, consumptions and lactate concentrations in nine patients before, during and 30 minutes after controlled hypotension are summarized in Table III.

TABLE III

Total Arteriovenous Oxygen Content Difference (AVDTO2), Jugular Arteriovenous Oxygen Content Difference (AVDjO2), Total Oxygen Consumption (V_TO_2), Arterial Oxygen Tension (PaO_2), Arterial Lactate Concentration (La) and Jugular Arteriovenous Lactate Content Differences (AVDjLa) before, during, and after Adenosine-induced Hypotension in Nine Patients.

	Control 1	Adenosine	Control 2
AVD_TO_2 (ml per liter)	46.3 ± 2.3	29.3 ± 2.5*	46.8 ± 3.2
AVD_jO_2 (ml. per liter)	85.2 ± 10.5	58.1 ± 14.1**	74.9 ± 7.7
V_TO_2 (ml per min.)	220 ± 15	193 ± 16**	235 ± 16
Pa_{O2} (mmHg)	111.0 ± 9.8	97.5 ± 9.0	93.0 ± 5.2
La (mmol per liter)	1.46 ± 0.17	1.73 ± 0.20	1.82 ± 0.20**
AVD_jLa (mmol per liter)	0.02 ± 0.04	−0.05 ± 0.05	−0.06 ± 0.04

*P < 0.01
**P < 0.05, denotes significantly different from control 1.

From Table III, it can be seen that arterial oxygen tension remained unchanged during adenosine-induced hypotension. VO_2 was decreased by 13±4%, with a decrease in $AVDO_2$ of 37±5%. The arterial lactate concentration was not affected by hypotension. The cerebral $AVDO_2$ decreased similarly by 37±13%, while the arterio-jugular lactate content difference was unaltered.

After the hypotensive period, the metabolic variables returned to the control levels, except for a minor increase in the arterial lactate concentration.

The ECG the day after the operation was unchanged. The mean serum creatinine level was 83±4 μM before operation and 70±3 and 71±4 μM on the first 2 postoperative days.

Adenosine infusion rate was constant during the hypotension, which suggests the absence of tachyphylaxis.

Subsequent to the tests described above, a 20 mM solution of adenosine in isotonic saline was administered to 50 surgical patients employing techniques similar to those described, except that pretreatment with dipyridamole was omitted. Hemodynamic effects similar to those described above were observed at adenosine dosages of 0.2–0.35 mg per kg per minute. The enhanced cardiac output obtained with adenosine, in combination with the maintained right and left heart filling pressures, is in contrast with the hemodynamic effects of controlled hypotension with sodium nitroprusside or nitroglycerine, as is shown in Table IV.

TABLE IV

Hemodynamic Effects of Adenosine and Nitroglycerine-(TNG), and Nitroprusside-(SNP) Induced Controlled Hypotension during Cerebral Aneurysm Surgery with Similar Anesthetic Techniques (Data Presented as Percent from Control)

	ADENOSINE n = 10	SNP** n = 17	TNG*** n = 20
MABP	−43*	−36*	−36*
RAP	+6	−39*	−45*
PAP	+15	−27*	−34*
PCWP	+14	−44*	−45*
HR	+9*	+22*	+19*
QT	+44*	−15*	−24*

TABLE IV-continued

Hemodynamic Effects of Adenosine and Nitroglycerine-(TNG), and Nitroprusside-(SNP) Induced Controlled Hypotension during Cerebral Aneurysm Surgery with Similar Anesthetic Techniques (Data Presented as Percent from Control)

	ADENOSINE n = 10	SNP** n = 17	TNG*** n = 20
SVR	−61*	−29*	−16*
PVR	−15	+11	+12

*P < 0.01
**Data from Lagerkranser et al., Acta. Anesth. Scand., 24: 426-32 (1980).
***Data from Lagerkranser, Acta. Anesth. Scand., 26: 453-7 (1982).

From the foregoing it is evident that continuous infusion of adenosine during anesthesia is capable of significantly reducing blood pressure without evidence of tachyphylaxis while, at the same time, causing a decrease in peripheral vascular resistance, an increase in cardiac output and a moderate increase in heart rate. This suggests that adenosine acts as a hypotensive agent through dilation of the arterial resistance vasculature. In contrast, sodium nitroprusside and nitroglycerine induce hypotension by both pre- and post-capillary dilatation. In addition, controlled hypotension with adenosine better preserves oxygen tissue pressures than hypotension induced by nitroprusside.

It should be noted that, at the levels of adenosine used, blockage of atrio-ventricular conductance does not occur in these persons during anesthesia. Indeed, adenosine is used in bolus form specifically to block A-V conductance. Apparently the peak plasma concentration of adenosine is considerably higher when giving bolus injections than the steady state levels obtained during these continuous infusions.

EXAMPLE II

CONTROL OF HYPERTENSIVE CRISIS

Continuous infusion of adenosine can also be employed to control hypertensive crises occurring during surgery. Such crises can occur, for example, as a result of release of catecholamines during surgery on pheochromocytoma—a tumor characterized by the presence of catecholamine—which can cause pulmonary edema and death. At present, this situation is treated prophylactically by pre-administration of alpha- and beta-adrenoceptor blockers or vasodilators, but the effect is often insufficient. It has now been found that, in the case of a catecholamine-induced hypertensive crises, prompt infusion of adenosine will rapidly restore blood pressure to normal, and will easily maintain normal pressure until the crisis has passed. Amounts of adenosine which are effective in controlling such hypertensive crises will depend on the degree of hypertension. However, as a general rule they are about half of the amounts found useful for controlled hypotension, i.e., in the range of from about 0.1 to about 0.2 mg adenosine per kilogram of body weight per minute.

EXAMPLE III

IMPROVED CORONARY CIRCULATION IN ISCHEMIC HEART DISEASE

Patients requiring abdominal vascular surgery, such as surgery for an aortic aneurysm, frequently also suffer from ischemic heart disease, or insufficient blood flow to the heart tissue, which may present undesirable complications in such

surgery. Accordingly, drugs with vasodilator properties, such as isoflurane and nitroprusside, have been investigated for possible use to increase myocardial blood flow and to reduce peripheral vascular resistance (after-load reduction) during such surgery; however, they have been found to have no beneficial effect with respect to coronary flow and, indeed, may reduce coronary blood flow. In contrast, adenosine administered by continuous infusion has been found very effective in increasing myocardial blood flow and, in such use, is accompanied by an increase in cardiac output.

For such application, the rate of adenosine administration should be such that there is no more than a 10–20 percent reduction in blood pressure. As a general rule, this is achieved by use of rates of administration of the order of 0.05 to about 0.1 mg. adenosine per kilogram per minute. In such a case myocardial blood flow has been found to be doubled, cardiac output has been increased by 10 to 20 percent, and blood pressure has been reduced by 10 to 20 percent, all without change in oxygen consumption and without ECG signs of ischemia.

EXAMPLE IV

CORONARY VASODILATION

It has been further found that when adenosine is administered by infusion at rates which do not induce significant hypotension, it has clinically useful regional effects in unanesthetized and anesthetized patients.

For example, adenosine at dosages of the order of 10 to 15 percent of hypotensive levels (e.g. 0.02 to 0.05 mg. per kg. per minute) can be a useful adjunct to coronary by-pass surgery, apparently due to a preferential coronary vasodilation. It has been reported that coronary artery grafts occlude more frequently during the postoperative period when low graft-flow values are obtained during surgery. See Groudin et al, Circulation, 42: Suppl 3; 106–111 (1970). It has been found that low doses of adenosine administered postoperatively increase graft blood flow without significant effect on atrio-ventricular conductance. The administration of low doses of adenosine for this purpose can be carried out for as long as is necessary to afford appropriate graft flows and reduce risk of occlusion, but ordinarily the period need not exceed 48 hours following surgery.

In a study designed to investigate the use of adenosine to inhibit occlusion of coronary grafts, nine patients (age 45–65, all taking beta-blockers) were studied during coronary artery surgery. After premedication, morphine (10–15 mg) and scopolamine (0.4–0.6 mg), anesthesia was induced by fentanyl (30 mcg/kg.b.w.). Pancuronium (0.1 mg/kg b.w.) was given to facilitate endotracheal intubation. Anesthesia was maintained with fentanyl 0.5 mg/hour, N_2O (50%) in oxygen and droperidol (0.1 mg/kg b.w.). During bypass thiomebumal (5 mg/kg b.w.) was given. Nitrous oxide was not used after bypass. Extracorporeal circulation (ECC) was performed with a roller pump and a Shiley bubble oxygenator primed with crystalloid solution. ECG (modified V_5), an arterial line and a Swan-Ganz Catheter were used for monitoring and for hemodynamic measurements. Blood flow in bypass grafts (n=15, internal mammary and venous grafts), was measured with appropriate sized square wave electromagnetic flow probes (Nycotron 732). The study was performed 20–30 minutes after the termination of ECC. After a control period (5 min), adenosine (5.3 mg/ml. clinical solution) was continuously infused in a central vein in order to induce approximately 10% reduction of mean arterial blood pressure (about 30 to 50 μg per kg. per min.). Graft flow was continuously measured before and during a 10 or 30 minute infusion of adenosine and finally during the following 5 minute control period. Data are expressed as mean ±SEM and differences were tested with Student's paired t-test against the preceding period.

The results of this study are summarized in Table V.

TABLE V

	CONTROL BEFORE	ADENOSINE	CONTROL AFTER
Mean Arterial Pressure (mmHg)	84 ± 3	74 ± 3 P < 0.01	85 ± 3 P < 0.01
Heart Rate (beats/min)	82 ± 5	82 ± 15	81 ± 6.
Cardiac Output (l/min)	4.8 ± 0.4	5.6 ± 0.3 P < 0.05	5.3 ± 0.3 n.s.
Pulmonary Artery Pressure (mean) (mmHg)	16.7 ± 1.2	18.8 ± 1.2	19.9 ± 1.0
RAP (mean) (mmHg)	4.7 ± 0.5	5.3 ± 0.4	5.8 ± 0.7
Stroke Index (ml/m^2)	35.6 ± 2.6	38.8 ± 2.0	39.6 ± 2.5
Left Ventricular Stroke Index (Joule/m^2)	0.44 ± 0.03	0.41 ± 0.03	0.49 ± 0.04
Graft flow ml/min (n = 15)	40 ± 5	77 ± 7 P < 0.001	39 ± 5 P < 0.001

As is evident from Table V, adenosine in a dose of 49±4 μg/kg/minute, a level which reduced mean arterial pressure 12%, increased cardiac output 12%, and doubled graft flow. At the same time, heart rate, mean pulmonary artery pressure, central venous pressure, stroke index and left ventricular stroke work index remained essentially unchanged. Graft flow rate was restored to its original value on termination of adenosine. No arrhythmias were observed.

This demonstrates that i.v. adenosine at low rates (30–50 μg per kg per min) induces a marked and reproducible increase in graft flow without increased myocardial work, apparently due to preferential vasodilatory effect of adenosine in the coronary vasculature.

EXAMPLE V

INCREASED CARDIAC OUTPUT

As is evident from the foregoing data, intravenously infused adenosine has the ability to increase cardiac output without increasing heart work. This is in contrast to other vasodilators, such as sodium nitroprusside, which may reduce cardiac output, depending on the hemodynamic status of the patient. As a consequence, adenosine can be used to stimulate cardiac output in patients with low cardiac output states, due, for example, to heart surgery, infarct and the like. This apparently is due to adenosine's ability to reduce after-load, without having significant effect on pre-load. In contrast, nitroprusside reduces both after-load and pre-load, and nitroglycerine is effective principally (90%) on reducing pre-load, and has only a marginal effect on after-load.

For this application, effective dosages are intermediate those used for increased graft flow and controlled hypotension. Typically the effective dose is of the order of 40–80 μg/kg/per minute. The duration of treatment can be as long as required to support the heart. It also has been found that, on termination of the adenosine, cardiac output, although less than that during treatment with adenosine, frequently remains above the cardiac output prior to treatment.

In this respect, adenosine is of value as an adjunct to dopamine treatment for cardiogenic shock. Dopamine is

frequently given to patients in shock to stimulate heart action and thereby increase blood pressure. Adenosine can be administered with dopamine to modulate peripheral resistance without compromising systemic blood pressure, and thus increase cardiac output.

Adenosine is unique in its activity in this respect, because it is able to reduce after-load without significantly increasing heart rate. In contrast, agents previously used to reduce cardiac after-load, for example, hydralazine and prazosin, increase heart rate.

EXAMPLE VI

PLATELET PROTECTION DURING CARDIOPULMONARY BYPASS

Continuous infusion of adenosine also has been found of use in protecting platelets during cardiopulmonary bypass. For such use, it is desired to maintain the adenosine dosage below that affording significant vasodilation, and a rate of about 100 μg/kg/min has been found effective. In contrast, prostacyclin, a prostaglandin used to inhibit platelet aggregation, is associated with severe systemic vasodilation and hypotension during coronary bypass surgery.

Twenty-five patients scheduled for coronary artery bypass surgery were randomly assigned to two groups—one with adenosine infusion (n=13) and the other with placebo infusion (n=12).

Routine tests of coagulation status were normal in all patients, and none was taking drugs known to affect platelet function. Intravenous anesthesia was used, either high-dose fentanyl (100–150 μg/kg) or balanced anesthesia (thiopental, fentanyl, diazepam and N_2O/O_2).

During cardiopulmonary bypass (CPB), mean arterial blood pressure (MABP) above 70 mmHg was treated with the vasodilator sodium nitroprusside (SNP), except in the final phase or rewarming. CPB was performed with SARN roller pump and a Shiley oxygenator (100A) primed with 2000 ml crystalloid solution (75 mg heparin). The perfusion rate was kept at approximately 1.8 ml/m^2 body surface. Moderate hypothermia (25° C.) was induced. Cardioplegia was obtained with Ringer's solution (with added potassium up to 20 mM/l). Heparin (3 mg/kg) was administered as a bolus injection before cannulation. The heparin effect was controlled by measurements of activated clotting time. (Hemocron® Int. Technidyne Corp, USA). This time (ACT) was >400 seconds in all patients during CPB. At the termination of CPB, the heparin effect was antagonized with protamine (c. 1.3 mg/mg heparin). ACT was checked 10–20 min after the protamine injection. ACT values 120 sec were considered satisfactory.

Platelet count (Linson 431 A cell counter) and hematocrit were determined in arterial samples before anesthesia, after thoracotomy, during CPB at 10, 20, 40, 60, 80 and 100 min, 30 min after CPB and on the postoperative day. Platelet counts were expressed in percentage of preanesthesia levels and were corrected for hemodilution MABP was monitored continuously via a catheter introduced in the radial artery. All patients received hypertonic mannitol (1–1.5 g/kg) during CPB and urine production was calculated as ml/.min of CPB (Table VI).

Peroperative blood loss and blood transfusions could not be compared between the groups, due to the smallness of the series and the involvement of many surgeons in the study. Postoperative bleeding was measured as the blood loss from the tube drainage from the end of operation until the postoperative morning. One patient in the adenosine group was excluded because of reoperation for surgical reasons (for massive bleeding due to suture insufficiency in a graft anastomosis) within 6 hours after CPB.

Adenosine (5.3 mg/ml, clinical solution) was infused at a rate of 100 μg/kg/min into the superior vena cava throughout CPB. The adenosine dose was based on five pilot cases in which a vasodilation dose-response was observed. The highest infusion rate that did not induce systemic vasodilation was chosen for this study. In six cases the plasma adenosine levels were determined by high performance liquid chromatography (HPLC) (Fredholm and Sollevi, J. Physiol. (London), 313: 351–67 (1981) in arterial and in venous (venous lines to the oxygenator) blood. The adenosine metabolites inosine, hypoxanthine and uric acid were also determined by HPLC. The samples were collected as previously described (Sollevi, Acta. Physiol. Scand., 121: 165–72 (1984) at the intervals of 10, 20, 40 and 80 minutes during CPB and 20 minutes after CPB.

Results are summarized in Tables VI and VII, below, in which data are presented as means ±SEM. Statistical significance (controls v. adenosine group) was determinted with Student's t-test for unpaired data. For significance within the groups the Wilcoxon Rank Sum test was used. $p<0.05$ was regarded as significant.

TABLE VI

	PATIENT DATA	
	Adenosine (mg per kg per min)	
	0	0.1
Age, years	57 (range 47–66)	57 (range 42–74)
Males/females	11/1	12/1
CPB-time (min)	95 ± 10	120 ± 10
Preoperative platelet counts (x 10^9 cells/l)	162 (range 107–235)	158 (range 106–251)
Peroperative urine production (ml/min CPB)	9.7 (2.1–20)	4.1 (1.1–9.5)
Postoperative blood loss (ml)	630 ± 60	640 80

TABLE VII

Arterial and Venous Concentrations (μM) of Adenosine and its Metabolites (n = 6), Before, During and After Adenosine Infusion (0.1 mg × kg−1 × min−1

		CPB				CPB 20'
	pre-CBP	10'	20'	40'	80'	post
Vein						
Adenosine	0.3 ± 0.2	3.7 ± 1.3*	5.7 ± 2.1*	4.5 ± 1.1*	3.0 ± 1.0*	0.4 ± 0.1
Inosine	0.2 ± 0.1	0.9 ± 0.3*	2.4 ± 1.2*	1.6 ± 0.5*	1.6 ± 0.4*	0.4 ± 0.1
Artery						
Adenosine	0.3 ± 0.1	0.7 ± 0.3	0.8 ± 0.4*	0.6 ± 0.3	0.4 ± 0.2	0.3 ± 0.1
Inosine	0.2 ± 0.1	1.3 ± 0.7*	2.5 ± 1.0*	1.2 ± 0.3*	1.4 ± 0.4*	0.4 ± 0.1
Hypoxanthine	3.2 ± 0.8	5.3 ± 1.2	7.7 ± 1.4*	5.3 ± 1.0	5.0 ± 0.9	4.2 ± 0.9
Uric Acid	250 ± 30	260 ± 32	269 ± 35	250 ± 32	249 ± 30	260 ± 34

*significantly different from pre-CPB value.

TABLE VIII

Time of count	Platelet Count; % of Awake Count: Control	Adenosine
Awake	100	100
Pre CPB	95	97
During CPB (Duration 100 minutes)		
10 minutes	80	96
20 minutes	75	87
40 minutes	65	85
60 minutes	80	91
80 minutes	78	100
100 minutes	77	97
Post CPB, time after infusion of CPB		
30 minutes	70	88
24 hours	60	60

As shown in Table VIII, platelet count was similar in the two groups before anesthesia and was unaltered by anesthesia and thoracotomy. In the control group the platelet count fell rapidly and markedly during the first 40 minutes of CPB and remained significantly reduced during and after CPB. During adenosine infusion the initial platelet reduction was small, and was significant only at 20 and 40 minutes on CPB. From 60 minutes to the end of CPB and at 30 minutes after CPB the platelet counts were not significantly different from those before anesthesia. Throughout CPB and 30 minutes after CPB there was significant intergroup difference in platelet counts. On the day after operation the platelet counts were markedly reduced in both groups, with no significant intergroup difference.

As shown in Table VII, the arterial and venous adenosine levels were in the normal range of 0.3 μM prior to CPB. The adenosine infusion raised the venous plasma concentration to 2–10 μM, while the arterial levels were approximately doubled during the initial CPB period. Only the first adenosine metabolite, inosine, was consistently elevated during the infusion.

All parameters had returned to control levels within 20 minutes after CPB.

The mean arterial blood pressure (MABP) did not differ significantly between the two groups during CPB. In the placebo group, however, seven patients required sodium nitroprusside infusion (<5 μg/kg/min) to keep MABP below 70 mmHg. No patient in the adenosine group required vasodilator treatment. After CPB the patients in the adenosine group had slightly lower MABP, but at the end of operation there was no intergroup difference. The urine production during CPB was 250 ml/h in the adenosine group and 500 ml/h in the controls ($p<0.01$, Table VI). Transient elevation of serum creatinine levels (10–20% above normal range) was found in two patients in the adenosine group and one control patient during two postoperative days. The postoperative blood loss did not differ between the groups.

All patients were extubated within 24 hours after the operation and all recovered normally. There were no clinical signs of neurologic complications and all the patients were discharged from the hospital.

EXAMPLE VII

ADDITION OF ADENOSINE TO CARDIOPLEGIA SOLUTION

Cardioplegia is induced during open heart surgery in order to arrest the heart and to reduce myocardial oxygen consumption during cardiopulmonary bypass. This is at present generally obtained by ice-cooled solution containing high concentration of potassium (20 mmol/liter, four times the normal serum level) that is infused into the coronary vessels. It is well known that high concentrations of potassium effectively induce asystole, but also cause damage on vascular endothelium. The latter may lead to permanent stenosis of coronary vessels.

The foregoing data has clearly demonstrated in human patients that adenosine is effective as coronary vasodilator and preserves circulating platelets. Adenosine is also known to be incorporated into high energy phosphates (ATP) in various tissues. In addition, it is well known since the early work of Drury and SZent Gyorgyi (J. Physiol (London) 68:213 (1929)) that high concentration of adenosine can produce heart block.

These four effects of adenosine are all useful during the induction of cardioplegia in human patients. First, the vasodilatory effect can counteract the vasoconstrictor effect of potassium and thereby reduce the time required for administration of cardioplegia solution. This will give a more rapid cooling and thereby more rapid asystoli. Secondly, the inhibitory effect of adenosine on platelet activation can prevent platelet aggregation in the coronary circulation during this cooling phase. Third, adenosine can

serve as a substrate and be incorporated into myocardial ATP during this condition, when the heart is beating without obtaining adequate oxygen supply. Finally, the well known A-V blocking effect of high concentrations of adenosine can be used for the induction of asystoli. Then, the potassium concentration of the cardioplegia solution can be reduced to a level that do not damage the vascular endothelium.

These four effects of adenosine can all be achieved with a cardioplegia solution containing 0.5–1.5 mg/ml of adenosine, administered into the coronary circulation during the induction of cardioplegia. Ordinarily, when adenosine is administered for this purpose through the aortic root, the cardioplegia solution will be administered at the rate of 50 to 150 ml./min. over a period of about 10 to about 20 minutes.

EXAMPLE VIII

DECREASING ACUTE PULMONARY VASCULAR RESISTANCE

Increased pulmonary resistance, e.g., pulmonary hypertension may clinically manifest as a severe disturbance of myocardial function of the right ventricle, a myocardial insufficency, and an impaired whole body oxygenation. Increased pulmonary resistance may be chronic or acute, ie., occur as a consequence of surgery, or pulmonary disease. The present invention is also directed to decreasing or normalizing acute pulmonary vascular resistance and pulmonary hypertension.

Pharmacological treatment of increased pulmonary vascular resistance or acute pulmonary hypertension has focused on using traditional vasodilating agents. These agents have been shown to have limited effects on pulmonary hypertension. The vasodilatory and hypotensive effects of these agents are primarily systemic, not localized in the pulmonary vasculature.

The endogenous vasodilator adenosine, in contrast to these traditional agents, has been shown to be most effective as a dilator of blood vessels in the organs in animal models as well as in humans. In animal and clinical studies, systemic vasodilator doses of adenosine have had negligible or at most, minor effect on normal pulmonary vascular resistance. Surprisingly, however, in animal models (pig) where pulmonary hypertension has been induced by hypoxic ventilation during anesthesia, intravenous adenosine administration unexpectedly produces a marked reduction in this higher-than-normal pulmonary vascular resistance. Even more surprising is the fact that this decrease in pulmonary vascular resistance occurs at a dosage of adenosine lower than that which exhibits systemic vasodilator effects. This effect is obtained at doses of adenosine that commonly do not result in detectable vasodilator effects or effects on cardiac output or arterial oxygen function.

It is believed that adenosine can exert its vasodilatory hypotensive effects in the pulmonary circulation without inducing systemic vasodilation because of its extremely rapid elimination in the blood stream (T1/2 less than 10 seconds). Thus, a dose titration of adenosine can be performed, producing effects in the pulmonary vasculature without producing systemic effects at least in part because the adenosine is eliminated before it reaches the resistance vessels of the systemic vasculature.

The expected suitable infusion rate of adenosine in humans to normalize pulmonary vascular resistance is typically 10–30 micrograms per kilogram of body weight (0.010–0.030 mg. per kg. per minute). As a general rule, the concentration of the adenosine in the infusion solution is at least about 5 millimol (1.5 mg/ml) and may be as high as the solubility limit of adenosine (about 20 millimol or 5.5–6 mg/ml). However, in small children, the concentration of adenosine in solution may be as low as 0.1 mg/ml.

Preferably, to effectuate the maximum pulmonary hypotensive effect, the adenosine solution is administered by infusion through a catheter to maximize the exposure of the pulmonary vasculature to adenosine. This is done by infusing the adenosive solution into a central vein such as the superior vena cava, or alternatively, into the right atrium. As the adenosine solution is infused, the pulmonary vascular resistance is monitored to determine the effect of the adenosine. Infusion of adenosine is maintained until pulmonary vascular resistance returns to normal levels or until there is no further evidence of decreasing pulmonary vascular resistance. Adenosine infusion may, of course, be continued for periods of time within practical limits in particularly difficult cases of higher-than-normal pulmonary vascular resistance. Using this selective pulmonary effect of adenosine, low infusion rates may be used for the treatment of acute postoperative pulmonary hypertension that occurs in patients after heart transplant surgery or in other surgeries.

EXAMPLE IX

DECREASING PULMONARY VASCULAR RESISTANCE IN CONJUNCTION WITH IDIOPATHIC RESPIRATORY DISTRESS SYNDROME

Adenosine infusion as described in example VIII may also be used to normalize pulmonary vascular resistance pulmonary hypertension) in children with idiopathic respiratory distress syndrome (IRDS).

Such treatment may be performed by infusing the low doses of adenosine as described in example VIII into the patient's pulmonary vasculature, e.g. via catheter introduced in the pulmonary artery for a period sufficient to lower pulmonary vascular resistance. The catheter's position should be distal to the ductus arteriosus. This way, adenosine does not enter the systemic circulation. Preferably, pulmonary vascular resistance should be lowered to within normal ranges. Thus, adenosine infusion should be maintained for a period sufficient to normalize pulmonary vascular resistance. This can be done by monitoring pulmonary vascular resistance during the infusion process, and maintaining infusion until pulmonary vascular resistance levels fall to normal. In difficult cases, adenosine infusion may be maintained for periods of time within practical limits.

EXAMPLE X

A METHOD OF DIAGNOSING THE OPERABILITY OF THE PULMONARY VASCULATURE IN PATIENTS EXHIBITING PULMONARY HYPERTENSION IN CONJUNCTION WITH CARDIAC SEPTUM DEFECTS

In children with myocardial septum defects (Vitium Organicum Cordis, VOC), who also exhibit pulmonary hypertension, adenosine infusion provides a means by which the value of surgically repairing septum defects can be determined. Adenosine induced reduction of pulmonary vasculature resistance may be a rapid and simplified technique for the evaluation of the usefulness of surgical repair of septum defects. This technique is a welcome diagnostic

tool since patients with septum defects having severe morphological damage of the pulmonary vasculature caused by pulmonary hypertension would not normally be responsive to adenosine vasodilation. This condition is not improved by surgical repair of the VOC.

The methodology comprises infusing an adenosine solution (infusion rate of 0.010–0.030 mg. per kg. per minute with a concentration of about 1.5 mg./ml. to about 5.5–6.0 mg./ml.) into the blood stream of a patient to maximize the exposure of the patient's pulmonary vasculature to adenosine. This can be done by infusing a solution of adenosine into a central vein, for example, the superior vena cava, or alternatively, into the right atrium. The blood pressure in the lung artery is then measured before and after administration of the adenosine as per the previously described technique in examples I and VI (cannula introduced in the artery). A decrease of arterial blood pressure is indicative of a reduction of the pulmonary vascular resistance and this, in turn, is an indication that the pulmonary vasculature will respond to surgical repair of the VOC. On the other hand, if there is no decrease in arterial pressure, then this would be indicative of morphological damage of the type that would not be improved by surgery.

An alternative to measuring a decrease in mean arterial pressure is to measure heart minute volume. In this technique, the heart minute volume is measured (same technique as described above in Example I) during administration of adenosine for a time period long enough to observe the pressure or flow change in heart volume. An increase in heart minute volume would indicate that the operability of the pulmonary vasculature had not been impaired, and that the cardiac septum defects could be corrected with surgery. No change would indicate the septum defects could not be corrected with surgery.

EXAMPLE XI

ADENOSINE IN PERCUTANEOUS TRANSLUMINAL CORONARY ANGIOPLASTY

A new method of treating coronary artery disease in human beings is effected by inserting a special catheter, equipped with an inflatable balloon, into a coronary artery which has an angiographically demonstrable stenosis. The procedure, known as percutaneous transluminal coronary angioplasty (PTCA), is executed as follows: under radiological control, the balloon of the catheter is placed in the stenosed part of the vessel. The balloon is inflated several times, each time with increasing pressures, and for a duration of 1 minute. Thereafter, the catheter is withdrawn from coronary circulation and the flow through the so treated vessel is checked by means of coronary agniography. The resultant widening of the diseased part of the coronary vessel leads to cracks in the intimal cell layer of the vessel. This trauma leads to activation of biochemical processes leading to local production of substances able to constrict the vessel, as well as activating platelets, which are circulating in the blood, in such a way that they are more easily deposited on the site of the previous stenosis. Such a platelet deposition is the first initiation of the coagulation process, which ultimately can lead to the formation of a blood clot. All these events act in conjunction to counter the intended effect of the PTCA treatment, and may ultimately lead to a re-occlusion of the vessel. During the period the PTCA procedure has been in widespread use, re-occlusions have been found to occur within 6 months of treatment in 25% or more of the cases. It is generally agreed that 5–10% or more occur in the first few days after treatment.

In order to lessen or prevent the negative effects of PTCA described above, vasodilating substances such as nitroglycerine, sodium nitroprusside, and the like, as well as platelet inhibiting substances and substances preventing blood coagulation such as acetylsalicylic acid, dipyridamol, heparin, coumarin and warfarin, have been administered to the patient before and after the procedure. However, all these substances have actions that are either too potent to be safe in conjunction with the PTCA procedure, since bleeding complications from the catheter puncture sites may occur, or too weak or unpredictable to be fully effective.

Adenosine may be used effectively in conjunction with PTCA because it possesses a unique combination of beneficial properties which all work to antagonize the complicating reactions described above. It has a potent vasodilatory effect on the coronary circulation which enables good blood flow through the treated vessel, which in turn prevents platelet deposition on the traumatized vessel site. In addition, adenosine antagonizes the action of locally produced vaso-constrictor substances. Adenosine also has an inhibiting effect on platelet aggregation, which further inhibits the chances of clot formation in the treated vessel. These effects are further enhanced by the ability of adenosine to inhibit presynaptic neural mechanisms regulating the release of catecholamines from nerve endings of the sympathetic nervous system which, as is well known, have consequences that all work for clot formation.

The adenosine dosage anticipated to be effective in this context is typically from 10 to 100 microgram of adenosine per kilogram per minute. Since the PTCA procedure is performed in an awake patient higher doses will generally not be employed, since such doses produce symptoms such as facial flushing, neck and chest oppression, palpitation and increased rate of respiration in an awake patient which, although not harmful or life-threatening, nonetheless should be avoided because they may induce anxiety.

Adenosine is preferably administered into a peripheral vein such as the femoral vein or brachial vein. Preferably, administration is begun shortly, e.g., a few minutes, before PTCA and continued for the duration of the PTCA and for several hours, e.g., 24 hours after PTCA.

The adenosine treatment may be given to patients undergoing the PTCA treatment under current medication with other anti-anginal drugs, such as adrenergic blocking drugs, calcium antagonists, diuretics, digitalis glycosides, angiotension converting enzyme inhibitors, antihyperlipidemic drugs, nitrate compounds including nitroglycerin or other vasodilatory compounds.

EXAMPLE XII

ADENOSINE IN CORONARY THROMBOLYSIS

A recent advance in the treatment of acute myocardial infarction is by means of introducing substances in the bloodstream to dissolve the clot(s) in the coronary circulation, which in most cases are the cause of the diseased state. This procedure, which is generally refered to as coronary thrombolysis (CTL), is performed by introducing streptokinase, urokinase or tissue plasminogen activator, either intravenously or directly into the coronary circulation. All of the advantages noted above for use of adenosine in PTCA are also applicable to the CTL procedures now in use, or which may be used in the future, since the same vascular and platelet reactions which occur with PTCA also occur in CTL once the thrombolysis has been achieved. In the CTL context, however, it may be advantageous to administer the

adenosine concomitantly with the thrombolytic agent, either separately or premixed with it in a fixed solution, such premixed solution being a further aspect of this invention. When the two agents are administered separately, it is desirable but not essential that the initiation of administration of each be simultaneous. For example, administration of the adenosine may be initiated before administration of the thrombolytic agent.

IN CTL, as in PTCA, adenosine may be administered intravenously at the same dosage as with PTCA. It is recognized that, due to the very brief half-life of adenosine, the dose is dependent on the site of administration. For example, a 30-milligram dose of adenosine per kilogram per minute given in the right atrium of the heart provides very similar effects to a 50-milligram dose of adenosine per kilogram per minute given in a forearm vein. It is conceivable that in CTL procedures, adenosine may be given directly in the coronary circulation. The dose then needed to achieve the same effects as described with intravenous administration would then be expected to lie in the range of 5 to 30 microgram of adenosine per kilogram per minute.

EXAMPLE XIII

ADENOSINE IN THE DIAGNOSIS OF CORONARY HEART DISEASE BY RADIONUCLEIDE SCINTIGRAPHY

In the diagnosis of coronary heart disease, modern techniques include visualization of myocardial irrigation by means of injecting short-lived radio-isotopes, such as thallium-201, into the blood-stream and record, by means of a gamma-radiation detector, the activity over the heart muscle.

It has been shown that an injection of the vasodilator dipyridamole can augment the redistribution of flow within the heart muscle so that those areas irrigated by stenosed vessels may be better visualized. The mechanism behind this is the so-called "steal" phenomenon: with a generalized maximal vaso-dilation of the heart muscle, relatively more blood will flow through the vessels not stenosed or constricted in any other way, thereby "stealing" the flow from the area supplied through a stenosed vessel.

Dipryridamol is an adenosine uptake inhibitor, which means it prevents adenosine from crossing the cell/membranes of the red blood cells from the plasma to the interior (the normal, rapid main pathway for adenosine elimination from plasma), thereby increasing the adenosine levels in plasma. Most data concerning the mechanism of action of dipyridamole's vasodilatory effect in fact support the view that it is solely due to adenosine vasodilation.

In a further aspect of this invention, adenosine can be used instead of dipyridamole in the diagnostic test described. It would in fact be an advantage to use adenosine insofar as it can be dosed exactly and dose-titrated to a precise effect, whereas with dipyridamole, the adenosine levels are unpredictable. Thus, a safer and more reliable test can be expected if adenosine is used.

The exact dose will normally have to be titrated individually but should lie in the range of 10 to 150 micrograms per kilogram per minute.

This invention has been described in terms of specific embodiments set forth in detail herein, but it is to be understood that these are by way of illustration and the invention is not necessarily limited thereto. Modifications and variations will be apparent from the disclosure and may be resorted to without departing from the spirit of the invention as those of skill in the art will readily understand. Accordingly, such variations and modifications are considered to be within the purview and scope of the invention and the following claims.

What is claimed is:

1. A method of selectively vasodilating the arteries of a human patient without inducing significant venous dilation and without pretreatment with dipyridamole, comprising continuously administering into the blood stream of said patient adenosine at a rate of administration of 0.35 milligrams of adenosine per kilogram body weight per minute, or less.

2. A method according to claim 1 in which adenosine is administered to an anesthetized patient undergoing surgery.

3. A method of selectively vasodilating the arteries of a human patient without inducing significant venous dilation and without pretreatment with dipyridamole, comprising continuously administering into the blood stream of said patient by intravenous administration about 0.05 milligrams to about 0.30 milligrams of adenosine per kilogram body weight per minute.

4. A method according to claim 3 in which adenosine is administered to an anesthetized patient undergoing surgery.

5. In a surgical method carried out on a patient under general anesthesia the improvement comprising continuously administering into the blood stream of said patient adenosine in an amount sufficient to selectively vasodilate the arteries of said patient without pretreatment with dipyridamole, at a rate of administration of 0.35 milligrams of adenosine per kilogram body weight per minute, or less.

6. In a method as claimed in claim 5, the improvement further comprising continuously administering into the blood stream of said patient adenosine at a rate of from 0.05 to about 0.3 milligrams of adenosine per kilogram of body weight per minute.

7. A method of selectively vasodilating the arteries of a human patient without inducing significant venous dilation and without pretreatment with dipyridamole, comprising continuously administering into the blood stream of said patient adenosine at a rate of administration of 0.01 to 0.15 milligrams of adenosine per kilogram body weight per minute.

8. A method for selectively vasodilating the arteries of an anesthetized human patient without inducing significant venous dilation and without pretreatment with dipyridamole comprising continuously administering into the blood stream of said patient by intravenous administration about 0.2 milligrams to about 0.35 milligrams of adenosine per kilogram body weight per minute.

9. A method for inducing a reduced afterload in the vascular system of a human without reducing the preload and without pretreatment with dipyridamole, the method comprising continuously administering into the blood stream of said patient adenosine at a rate of administration of 0.35 milligrams of adenosine per kilogram body weight per minute, or less.

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EXHIBIT B

United States Patent [19]

Mohiuddin et al.

[11] **Patent Number: 5,070,877**

[45] **Date of Patent: Dec. 10, 1991**

[54] **NOVEL METHOD OF MYOCARDIAL IMAGING**

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[21] Appl. No.: **330,156**

[22] Filed: **Mar. 29, 1989**

Related U.S. Application Data

[63] Continuation-in-part of Ser. No. 231,217, Aug. 11, 1988, abandoned.

[51] **Int. Cl.**[5] **A61B 6/00**

[52] **U.S. Cl.** **128/653.4**; 600/4; 424/9; 514/46; 128/654

[58] **Field of Search** ..... 600/3, 4; 128/659 C; 424/9; 514/46, 47

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Attorney, Agent, or Firm—Pennie & Edmonds

[57] **ABSTRACT**

The parenteral use of adenosine, functional adenosine receptor agonists which include 1-methyl-2-phenylethyladenosine, 5-ethyl carboxamide adenosine, cyclopentyl adenosine and 2-chloro adenosine; metabolic precursors or by-products of adenosine which include adenine and inosine; and phosphorylated derivatives of adenosine including adenosine monophosphate, adenosine diphosphate and adenosine triphosphate in conjunction with various invasive and noninvasive diagnostic techniques to detect the presence or assess the severity of vascular disease is a novel application (indication) for these compounds and forms the basis of this patent application.

48 Claims, No Drawings

NOVEL METHOD OF MYOCARDIAL IMAGING

This application is a continuation-in-part of application Ser. No. 231,217 filed Aug. 11, 1988 now abandoned.

BACKGROUND OF THE INVENTION

Several invasive and noninvasive techniques are used to assess patients with known or suspected coronary artery disease. Included among the noninvasive methodologies are electrocardiography, radionuclide angiography (first pass and equilibrium studies utilizing, for example, technecium 99 m labeled red blood cells), myocardial perfusion scintigraphy (utilizing positron emitting radiopharmaceuticals, for example, thallium-201, rubidium-82, nitrogen-13), and echocardiography (M mode and two dimensional). The manifestations of coronary artery disease are a function of the balance between myocardial oxygen supply and demand. Although these noninvasive procedures may be performed in a resting subject, there may not be sufficient imbalance between supply and demand to detect abnormalities at rest. Therefore, provocative studies are frequently performed to improve the predictive accuracy of these diagnostic proc dures. The most commonly employed provocative (stress) technique utilizes a standard exercise protocol. Under conditions of exercise myocardial oxygen demand is increased to exceed supply. This form of stress testing is commonly employed in conjunction with electrocardiography, radionuclide angiography, myocardial perfusion scintigraphy, echocardiography, and contrast ventriculography.

Recently, provocative studies have been developed utilizing pharmacological techniques designed to increase myocardial oxygen supply. Specifically, coronary vasodialators (e.g. nitrates, papavarine, dipyridamole, etc.) have been used for this purpose, although none have been approved by the FDA for this specific indication. While the mechanism is not clear, these agents may dilate normal vessels to a greater extent than diseased vessels, establishing a shunt or "myocardial steal". Pharmacological provocation may be particularly useful in patients who are unable to exercise, and may be equal to or superior to exercise provocation in patients capable of exercising. Furthermore, since exercise increases demand and coronary vasodilators increase supply, it is possible that the highest diagnostic yield will accrue when they are used in conjunction with one another.

Coronary arteriography is an invasive procedure which currently represents the "gold standard" for confirming the diagnosis of coronary artery disease. However, this procedure only establishes the anatomical severity of the disease and provides little information concerning the functional significance of visible lesions. Furthermore, small vessel disease may be present and beyond the resolution of currently available equipment. Recently, in an attempt to establish the functional significance of coronary lesions, coronary vasodilators have been administered by intracoronary injection or intravenous infusion and coronary blood flow is measured by one of several techniques, such as doppler flow catheters, videodensitometry, coronary sinus thermodilution, and radionuclide clearance of inert gases. These techniques are becoming more widely used to measure coronary flow reserve (i.e. reserve capacity) which provides important information concerning the functional significance of stenotic vessels. Although nitrates, papavarine, and dipyridamole have been used by some physicians for this purpose, no vasodilator has been approved by the FDA for this specific indication. The use of adenosine, 1-methyl-2-phenylethyl-adenosine, 5-ethyl carboxamide adenosine, cyclopentyl adenosine 2-chloro adenosine, adenine, inosine, adenosine monophosphate, adenosine diphosphate, or adenosine triphosphate, in conjunction with the above stated techniques to measure coronary flow reserve and assess the functional severity of stenotic vessels represents a novel application (indication) of our compound.

SUMMARY OF THE INVENTION

Briefly, the present invention comprises a method of detecting the presence or assessing the severity of vascular disease which includes the administration to the human host of an effective dilating amount of adenosine; functional adenosine receptor agonists (e.g., 1-methyl-2-phenylethyladenosine, 5-ethyl carboxamide adenosine, cyclopentyl adenosine or 2-chloro adenosine); metabolic precursors or byproducts of adenosine (e.g., adenine and inosine); and phosphorylated derivatives of adenosine (e.g., adenosine monophosphate, adenosine diphosphate, or adenosine triphosphate), in conjunction with invasive or noninvasive techniques.

It is an object of this invention to provide a new diagnostic method to aid in the determination of the extent and severity of heart disease.

It is a further object of this invention to provide a new radioimaging technique for the coronary arteries.

More particularly, it is one object of this invention to provide an improved method of radioimaging the coronary arteries.

It is one significant object of this invention to provide wash out times for the radiolabeled agents used in stress-free cardiac imaging which are comparable to the wash out times presently attainable only in stress or exercise radioimaging tests.

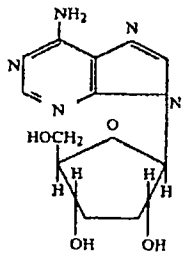
These and other objects and advantages will be apparent from the more detailed description which follows.

DETAILED DESCRIPTION OF THE INVENTION

Adenosine is chemically designated as 9-β-D-ribofuranosyl-9H-purine-6-amine; 6-amino-9-β-D-ribofuranosyl-9H-purine; 9-β-D-ribofuranosidoadenine; adenine riboside.

Adenosine is a nucleoside widely distributed in nature. factured from yeast nucleic acid. It is practically insoluble in alcohol. Crystals form from water, mp 234°–235°. $[\alpha]^{11}-61.7°$ (c=0.706 in water; $[\alpha]^{9}-58.2°$ (c=658 in water). uv max: 260 nm (ε15,100).

The structural formula is as follows:



-continued

$C_{10}H_{13}N_5O_4$	267.24
Empirical Formula	Molecular Weight

This invention utilized adenosine administration as a pharmacological stressor in conjunction with any one of several noninvasive diagnostic procedures available. For example, intravenous adenosine may be used in conjunction with thallium-201 myocardial perfusion imaging to assess the severity of myocardial ischemia. In this case, anyone of several different radiopharmaceuticals may be substituted for thallium-201 (e.g. rubidium-82, technitium 99m, derivatives of technitium 99m, nitrogen-13, iodine 123, etc.). Similarly, adenosine may be administered as a pharmacological stressor in conjunction with radionuclide angiography to assess the severity of myocardial dysfunction. In this case, radionuclide angiographic studies may be first pass or gated equilibrium studies of the right and/or left ventricle. Similarly, adenosine may be administered as a pharmacological stressor in conjunction with echocardiography to assess the presence of regional wall motion abnormalities. Similarly, adenosine may be administered as a pharmacological stressor in conjunction with invasive measurements of coronary blood flow such as by intracardiac catheter to assess the functional significance of stenotic coronary vessels.

This invention typically involves the administration of adenosine by intravenous infusion in doses which are effective to provide coronary artery dilation (approximately 20–200 mcg/kg/min). However, its use in the invasive setting may involve the intracoronary administration of the drug in bolus doses of 2-20 mcg. The adenosine used in this invention is normally admixed with any pharmaceutically suitable carrier or carriers such as saline, dextrose, water, or any other carrier customarily used for the type of administration intended. The solution may contain the active ingredient in a widely varying amount, for example, from about 1 mg/ml to about 12 mg/ml.

These doses increase coronary flow approximately 4–5 times resting values. Unlike papavarine which in this setting frequently causes QT interval prolongation, significant electrocardiographic or systemic hemodynamic abnormalities have not been observed. Adenosine is a superior vasodilator for this purpose.

The practice of this invention is applicable to radiopharmaceuticals generally, and specifically to those mentioned hereinabove.

Contemplated as equivalents of adenosine in the practice of this invention are analogues, derivatives, metabolic precursors or by-products or conjugates intended to function as agonists of the adenosine receptor responsible for mediating vasodilation. This appears to be the A_2 receptor subtype. Several analogues of adenosine have been developed which appear to have greater affinity or specificity for the A_2 receptor. These include primarily the N_6 substituted derivatives and the 2-carbon derivatives such as 1-methyl-2-phenylethyl-adenosine, 5-ethyl carboxamide adenosine, cyclopentyl adenosine, 2-chloro adenosine, etc.

The following methods are preferred embodiments of our invention.

The method comprising the use of an agent which is adenosine, functional adenosine receptor agonists, metabolic precursors or by-products of adenosine, or phosphorylated derivatives of adenosine as a substitute for exercise in conjunction with myocardial perfusion imaging to detect the presence and/or assess the severity of coronary arter disease in humans wherein myocardial perfusion imaging is performed by any one of several techniques including radiopharmaceutical myocardial perfusion imaging, planar (conventional) scintigraphy, single photon emission computed tomography (SPECT), positron emission tomography (PET), nuclear magnetic resonance (NMR)imaging, perfusion contrast echocardiography, digital subtraction angiography (DSA), or ultrafast x-ray computed tomography (CINE CT).

The method comprising the use of an agent which is adenosine, functional adenosine receptor agonists, metabolic precursors or by-products of adenosine, or phosphorylated derivatives of adenosine as a substitute for exercise in conjunction with imaging to detect the presence and/or assess the severity of ischemic ventricular dysfunction in humans wherein ischemic ventricular dysfunction is measured by any one of several imaging techniques including echocardiography, contrast ventriculography, or radionuclide angiography.

The method comprising the use of an agent which is adenosine, functional adenosine receptor agonists, metabolic precursors or by-products of adenosine, or phosphorylated derivatives of adenosine as a coronary hyperemic agent in conjunction with means for measuring coronary blood flow velocity to assess the vasodilatory capacity (reserve capacity) of coronary arteries in humans wherein coronary blood flow velocity is measured by any one of several techniques including Doppler flow catheter, digital subtraction angiography or other radiopharmaceutical imaging technique.

The following Examples are to illustrate the invention, and are not intended to limit the invention.

EXAMPLE I

As set forth in this example, the effects of intravenous adenosine as a pharmacological stressor in conjunction with thallium 201 scintigraphy were evaluated. In the first set of experiments, adenosine was compared to exercise in a crossover study design using planar (conventional) thallium 201 scintigraphy in a population of 20 healthy normal volunteers. In the second set of studies, adenosine was compared to dipyridamole in a crossover study design using planar (conventional) thallium 201 scintigraphy in a population of 26 subjects (12 healthy volunteers and 14 patients with angiographically documented coronary artery disease). In the third set of experiments, adenosine was evaluated using thallium 201 single-photon emission computed tomography (SPECT) in a population of 33 patients (18 normal subjects and 15 patients with angiographically documented coronary artery disease).

In the first set of experiments, 20 healthy normal volunteers (age 19–39 years) underwent planar (conventional) stress/redistribution thallium 201 scintigraphy twice (in a random crossover design). One study employed maximum treadmill exercise (Bruce protocol) as the method of stress and the other study employed an intravenous infusion of adenosine as the method of stress. Heart rate, blood pressure and a 12-lead electrocardiogram were monitored throughout the study. The exercise stress test was conducted in standard fashion. The adenosine stress test employed a constant infusion of adenosine initiated at 20 mcg/kg/min. The infusion was doubled at intervals to a maximum dose of 140 mcg/kg/min. The maximum tolerable dose was admin-

istered for at least 5 minutes prior to a single bolus injection of thallium 201 (approximately 2.0 mCi). Early (stress) imaging was performed 5–10 minutes after the thallium injection and delayed (redistribution) imaging was performed 3–4 hours after thallium injection. The adenosine infusion was continued to the end of early imaging. Early and delayed imaging each consisted of 3 sets of images (left arterior oblique, anterior and left lateral projections). The images were acquired and reconstructed in standard fashion. The adenosine infusion was well tolerated in all subjects. The exercise stress images and the adenosine stress images were interpreted as normal (i.e., no perfusion defect detected) in all subjects. This experiment indicates that adenosine compares favorably to exercise in detecting normalcy by planar thallium 201 scintigraphy.

In the second set of experiments, 12 healthy normal volunteers and 14 patients with angiographically documented coronary artery disease underwent planar (conventional) stress/redistribution thallium 201 scintigraphy twice (in a random crossover design). One study employed oral dipyridamole (300 mg) as the method of stress and the other study employed an intravenous infusion of adenosine as the method of stress. Dipyridamole stress imaging was performed in standard fashion and adenosine stress imaging was performed as described above. Again, the adenosine infusion was well tolerated in all subjects. The sensitivity, specificity and overall predictive accuracy for detection of coronary artery disease was 88.8%, 87.5% and 88.0%, respectively, with adenosine imaging, and 77.7%, 82.6% and 80.5%, respectively, with dipyridamole imaging. The positive predictive value of adenosine and dipyridamole imaging was 84.2% and 77.7% respectively. This study indicates that adenosine imaging is safe and may be superior to dipyridamole imaging for the accurate detection of angiographically significant coronary artery disease.

In the third set of experiments, 15 patients with angiographically documented coronary artery disease and 18 subjects with either angiographically normal coronary arteries (n=8) or healthy normal volunteers (n=10) underwent thallium 201 myocardial perfusion imaging using single photon emission computed tomography (SPECT). In all subjects, only an infusion of adenosine was employed as a method of stress. The adenosine infusion was initiated at 50 mcg/kg/min and titrated at 1 minute intervals by increments of 25 mcg/kg/min to a maximum dose of 140 mcg/kg/min. The maximum tolerable dose was maintained for at least 1 minute prior to and 3 minutes subsequent to a single bolus injection of thallium 201 (approximately 3.0 mCi). Early (stress) imaging was performed 5–10 minutes post-thallium and delayed (redistribution) imaging was performed 3–4 hours post-thallium. The SPECT images were acquired and reconstructed in standard fashion. Side effects occurred in 76% of the subjects, but were usually mild, did not require therapy and ceased instantly after discontinuing the adenosiene infusion. Chest pain occurred in 53%, headache in 34% and cutaneous flushing in 15%. Dose-dependent decreases is systolic blood pressure (hypotension) and reflex increases in heart rate were common. Perfusion defects were detected during adenosine stress imaging in all 15 patients with known coronary artery disease and these defects were reversible in 9 (sensitivity =100%). The adenosine stress images were interpreted as normal in 16 of 18 presumed healthy subjects (specificity =89%). This study indicates that adenosine-induced coronary vasodilation is a safe, convenient, and potent intervention to uncover perfusion defects during SPECT thallium scintigraphy in patients with coronary artery disease.

EXAMPLE II

As set forth in this example, the effects of intravenous adenosine as a pharmacological stressor in conjunction with echocardiography were evaluated.

Fifteen patients with a positive exercise (stress) SPECT thallium 201 tomogram were selected for this study. The tomographic perfusion defect was fixed (irreversible) in 6 subjects and reversible in 9 subjects. Subsequently, these patients underwent standard 2-dimensional echocardiographic studies under conditions of rest (baseline) and during an intravenous infusion of adenosine as previously described (Example I, 3rd set of experiments). Echocardiographic studies were performed over a 1 minute period prior to the adenosine infusion (baseline), during maximum adenosine infusion (140 mcg/kg/min), and 3 minutes after the cessation of the adenosine infusion. All echocardiographic studies included parasternal views (long axis and short axis at the level of the mitral valve, papillary muscles and apex) and apical views (4-chamber, 2-chamber and apical long axis). All echocardiogrpahic images were interpreted by standard qualitative and quantitative techniques. The echocardiographic images obtained at rest were interpreted as normal in all subjects. However, left ventricular wall motion abnormalities were detected during adenosine (stress) studies in all 6 patients with fixed thallium perfusion defects. Left ventricular wall motion remained normal during the adenosine infusion in all patients with reversible thallium perfusion defects. This study indicates that adenosine may be a useful pharmacological stressor for the detection if ischemic ventricular dysfunction as assessed by echocardiography.

EXAMPLE III

As set forth in this example, the effects of intravenous and intracoronary adenosine as a pharmacological stressor in conjunction with measurements of coronary blood flow reserve (CBFR) were evaluated at the time of coronary arteriography using a Doppler flow catheter.

Ten patients with an angiographically normal left coronary artery were studied at the time of diagnostic coronary arteriography. A 3F Doppler catheter was positioned in the left coronary artery to measure coronary blood flow velocity (CBFV), and mean arterial pressure, heart rate and the ECG were simultaneously recorded. Following repeated measures of baseline CBFV, incremental doses of intracoronary papaverine (8–12 mg boluses), intracoronary adenosine (4-14 mcg boluses) and intravenous adenosine (70–140 mcg/kg/min infusions) were administered in crossover fashion. Each drug was titrated to the maximum coronary hyperemic response. While the ECG intervals were unchanged during adenosine administration, papaverine routinely prolonged the QT interval (mean 96±18 msec). Relative to papaverine, maximum coronary hyperemic responses (4–5 fold increases in CBFV) were achieved with 14 mcg intracoronary bolus doses of adenosine, as well as 140 mcg/kg/min intravenous infusions of adenosine. Compared to papaverine, maximal coronary hyperemia occurred sooner with adenosine (10 vs 20 seconds) and resolved sooner with adeno-

19. A method of detecting the presence and assessing the severity of coronary artery disease in a human comprising the steps of:

(a) administering by an intracoronary route to said human about 2 mcg to about 20 mcg of an adenosine receptor agonist sufficient to provide coronary artery dilation;

(b) administering a radiopharmaceutical agent into said human; and

(c) performing radiopharmaceutical myocardial perfusion imaging on said human in order to detect the presence and assess the severity of coronary artery disease.

20. The method of claim 17 or 19, wherein said adenosine receptor agonist is selected from the group consisting of adenosine, 1-methyl-2-phenylethyl-adenosine, 5-ethyl carboxamide-adenosine, cyclopentyl adenosine, 2-chloro adenosine, adenosine, inosine, adenosine monophosphate, adenosine diphosphate and adenosine triphosphate.

21. The method of claim 17 or 19, wherein said radiopharmaceutical agent is selected from the group consisting of thallium-201, technetium-99m, derivatives of technetium-99m, nitrogen-13, rubidium-82 iodine-123 and oxygen-15.

22. The method of claim 17 or 19, wherein said radiopharmaceutical myocardial perfusion imaging is selected from the group consisting of scintigraphy, single photon emission computed tomography (SPECT), positron emission tomography (PET), nuclear magnetic resonance (NMR) imaging, perfusion contrast echocardiography, digital subtraction angiography (DSA) and ultrafast X-ray computed tomography (CINE CT).

23. The method of claim 17, 19 or 18, wherein said adenosine receptor agonist is adenosine.

24. The method of claim 20 wherein said adenosine receptor agonist is adenosine.

25. The method of claim 21 wherein said radiopharmaceutical agent is thallium-201.

26. The method of claim 22 wherein said radiopharmaceutical myocardial perfusion imaging is scintigraphy.

27. A method of detecting the presence and assessing the severity of ventricular dysfunction caused by coronary artery disease, in a human, comprising the steps of:

(a) administering by an intravenous route to said human about 20 mcg/kg/minute to about 200 mcg/kg/minute of an adenosine receptor agonist sufficient to provide coronary artery dilation;

(b) performing a ventricular function imaging technique on said human; and

(c) determining the presence and assessing the severity of ventricular dysfunction.

28. A method of detecting the presence and assessing the severity of ventricular dysfunction caused by coronary artery disease, in a human, comprising the steps of:

(a) administering by an intravenous route to said human about 2 mcg to about 20 mcg of an adenosine receptor agonist sufficient to provide coronary artery dilation;

(b) performing a ventricular function imaging technique on said human; and

(c) determining the presence and assessing the severity of ventricular dysfunction.

29. The method of claim 27 or 28, wherein said adenosine receptor agonist is selected from the group consisting of adenosine, 1-methyl-2-phenylethyl-adenosine, 5-ethyl carboxamide-adenosine, cyclopentyl adenosine, 2-chloro adenosine, adenine, inosine, adenosine monophosphate, adenosine diphosphate and adenosine triphosphate.

30. The method of claim 27 or 28, wherein said ventricular function imaging technique is selected from the group consisting of echocardiography, contrast ventriculography and radionuclide angiography.

31. The method of claim 27 wherein said adenosine receptor agonist is administered by intravenous infusion in a dosage of about 140 mcg/kg/minute.

32. The method of claim 27, 28 or 31, wherein said adenosine receptor agonist is adenosine.

33. The method of claim 29 wherein said adenosine receptor agonist is adenosine.

34. The method of claim 30 wherein said ventricular function imaging technique is echocardiography.

35. A method of determining the difference between the coronary blood flow through disease free coronary vessels and stenotic coronary vessels in a human comprising the steps of:

(a) administering by an intravenous route to said human about 20 mcg/kg/minute to about 200 mcg/kg/minute of an adenosine receptor agonist sufficient to provide coronary artery dilation;

(b) performing a method for measuring coronary blood flow velocity on said human in order to assess the vasodilatory capacity of disease free coronary vessels as opposed to stenotic coronary vessels.

36. The method of claim 35 wherein said adenosine receptor agonist is administered by intravenous infusion in a dosage of about 140 mcg/kg/minute.

37. A method of determining the difference between the coronary blood flow through disease free coronary vessels and stenotic coronary vessels in a human comprising the steps of:

(a) administering by an intracoronary route to said human about 2 mcg to about 20 mcg of an adenosine receptor agonist sufficient to provide coronary artery dilation;

(b) performing a method for measuring coronary blood flow velocity on said human in order to assess the vasodilatory capacity (reserve capacity) of disease free coronary vessels as opposed to stenotic coronary vessels.

38. The method according to claim 35 or 37, wherein said adenosine receptor agonist is selected from the group consisting of adenosine, 1-methyl-2-phenylethyl-adenosine, 5-ethyl carboxamide-adenosine, cyclopentyl adenosine, 2-chloro adenosine, adenine, inosine, adenosine monophosphate, adenosine diphosphate and adenosine triphosphate.

39. The method of claim 35 or 37, wherein said method for measuring coronary blood flow velocity is selected from the group of Doppler flow catheter, digital subtraction angiography and radiopharmaceutical imaging techniques.

40. The method of claim 35, 37 or 36, wherein said adenosine receptor agonist is adenosine.

41. The method of claim 38 wherein said adenosine receptor agonist is adenosine.

42. The method of claim 39 wherein said method for measuring coronary blood flow velocity is doppler flow catheter.

43. A method of detecting the presence and assessing the severity of coronary artery disease in a human comprising the steps of:

(a) administering to said human by intravenous infusion about 20 mcg/kg/minute to about 200 mcg/kg/minute of adenosine in order to provide coronary artery dilation;

(b) administering thallium-201 to said human; and

(c) performing the scintigraphy on said human in order to detect the presence and assess the severity of coronary artery disease.

44. A method of detecting the presence and assessing the severity of ventricular dysfunction in a human comprising the steps of:

(a) administering to said human by intravenous infusion about 20 mcg/kg/minute to about 200 mcg/kg/minute of adenosine in order to provide coronary artery dilation;

(b) performing an echocardiography on said human; and

(c) determining the presence and assessing the severity of ventricular dysfunction.

45. A method of determining the difference between coronary blood flow through disease free coronary vessels and stenotic coronary vessels in a human comprising the steps of:

(a) administering to said human by intracoronary bolus injection about 2 mcg to about 20 mcg of adenosine, in order to provide coronary artery dilation;

(b) measuring the difference between coronary blood flow through disease-free coronary vessels and stenotic coronary vessels in said human using a doppler flow catheter in order to assess the vasodilatory capacity (reserve capacity) of disease-free coronary vessels as opposed to stenotic coronary vessels.

46. A method of detecting the presence and assessing the severity of coronary artery disease in a human comprising the steps of:

(a) administering to said human by intracoronary bolus injection about 2 mcg to about 20 mcg of adenosine in order to provide coronary artery dilation;

(b) administering thallium-201 to said human; and

(c) performing scintigraphy on said human in order to detect the presence and assess the severity of coronary artery disease.

47. A method of detecting the presence and assessing the severity of ventricular dysfunction in a human comprising the steps of:

(a) administering to said human by intracoronary bolus injection about 2 mcg to about 20 mcg of adenosine in order to provide coronary artery dilation;

(b) performing an echocardiography on said human; and

(c) determining the presence and assessing the severity of ventricular dysfunction.

48. A method of determining the difference between coronary blood flow through disease free coronary vessels and stenotic coronary vessels in a human comprising the steps of:

(a) administering to said human by intravenous infusion about 20 mcg/kg/minute to about 200 mcg/kg/minute of adenosine, in order to provide coronary arter dilation;

(b) measuring the difference between coronary blood flow through disease-free coronary vessels and stenotic coronary vessels in said human using a Doppler flow catheter in order to assess the vasodilatory capacity (reserve capacity) of disease-free coronary vessels as opposed to stenotic coronary vessels.

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