Journal of Labelled Compounds and Radiopharmaceuticals—Vol. XXXI, No. 10

Synthesis of 9-(2-Phosphonylmethoxy)ethyl-8-[^{14}C]adenine [^{14}C]PMEA

U. J. Haynes, J. E. Swigor and J. J. Bronson

Bristol-Myers Squibb Company
Pharmaceutical Research Institute
Syracuse, New York 13221-4755

and

Bristol-Myers Squibb Research Center
5 Research Parkway
Wallingford, Connecticut 06492-7660

SUMMARY

The synthesis of [^{14}C] PMEA (3) was achieved by coupling the sodium salt of 8-[^{14}C]adenine (1) with 2-(diisopropylphosphonymethoxy)ethanol methanesulfonate in N,N-dimethylformamide at 100°C to provide the diisopropyl ester (2). Deesterification with bromotrimethylsilane in acetonitrile, followed by concentration and then aqueous hydrolysis of the resulting silylated intermediate produced [^{14}C] PMEA (3) as a crystalline solid having a radiochemical purity of 99.1% and a specific activity of 88.7 μCi/mg in an overall yield of 30%.

KEY WORDS

9-(2-Phosphonylmethoxy)ethyl-8-[^{14}C]adenine, [^{14}C] PMEA, antiviral.

INTRODUCTION

9-(2-phosphonylmethoxy)ethyl adenine (PMEA) is a potent and selective inhibitor of retroviruses,[2,3] including human immunodeficiency virus (HIV), the causative agent of AIDS. In in vivo murine models of retrovirus

0362–4803/92/100837–04$07.00

Received 23 June, 1992
Revised 27 June, 1992

infection, PMEA is more efficacious then AZT.[3,4] PMEA is active in vitro against human cytomegalovirus,[1,3] a common opportunistic infection associated with AIDS.

During the development of PMEA, pharmacokinetic and drug disposition studies were essential to understand its absorption, tissue distribution, metabolism and elimination in various animal models used in the investigation of safety and efficacy.

This report describes the synthesis of [^{14}C]PMEA.

EXPERIMENTAL

Materials

8-[^{14}C]Adenine was purchased from Moravek Biochemicals, Inc. All other reagents were ACS Grade or the highest quality commercially obtainable. NMR spectra were obtained on a Bruker Spectrospin 360 MHz instrument, using tetramethylsilane as an internal standard. Radioactivity was measured by a Beckman LS9000 liquid scintillator. TLC Plates: Silica gel, 250μ GF (Analtech). Method: Mobile phase, as indicated; visualization, UV 254 nm.

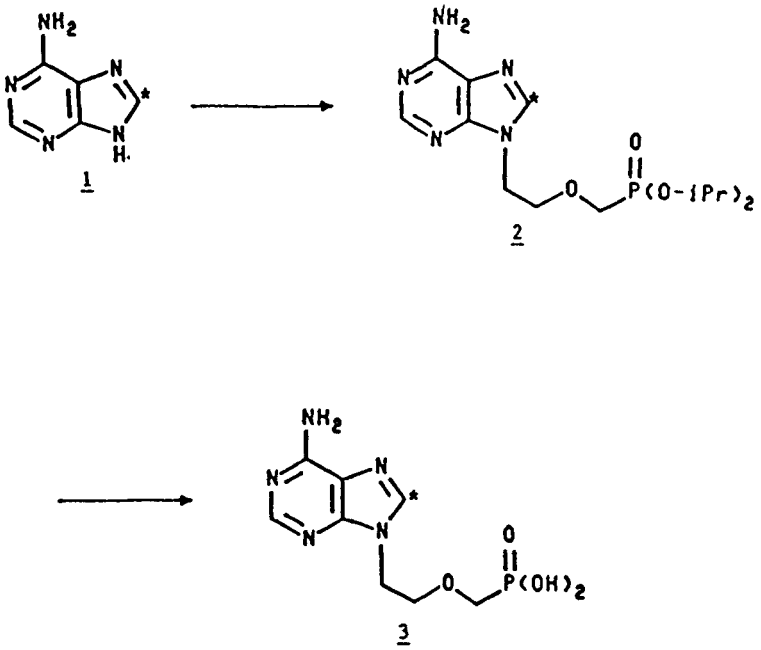
9-(2-Diisopropylphosphonylmethoxy)ethyl-8-[^{14}C]adenine (2).

Sodium hydride (55 mg, 80% oil dispension, 1.8 mmol) was added in one portion to a rapidly stirred slurry of adenine (113 mg) and 8-[^{14}C] adenine (130 mg, 50 mCi, 52.3 mCi/mmol, total of 1.8 mmol) in N,N-dimethylformamide (7 ml) in a 25 ml 3-necked, round-bottom flask equipped with magnetic stirrer and a nitrogen inlet adapter. The reaction mixture was heated at 80°C for 2 h to give a white slurry. A solution of 2-(diisopropylphosphonylmethoxy)ethanol methanesulfonate (573.6 mg, 1.8 mmol) in N,N-dimethylformamide (0.5 ml) was then added via a syringe to the hot slurry and the reaction temperature was raised to 100°C. The reaction mixture was stirred at 100°C for 5 h, allowed to cool at room temperature, and filtered to remove insoluble material. The filtrate was concentrated in vacuo and the gummy residue was suspended in 10% isopropanol in methylene chloride (5 ml). The mixture was filtered, and the filtrate was concentrated in vacuo to give an orange oil. Purification by column chromatography on silica gel (Silica CC-7, 60 g) eluting with a gradient of 3%-->5%-->7% methanol/methylene chloride afforded 350 mg of product (2) as a white crystalline solid.

9-(2-Phosphonylmethoxy)ethyl-8-[^{14}C]adenine (3).

A solution of (2) (350 mg, 1 mmol) in anhydrous acetonitrile (5 ml) was treated with bromotrimethylsilane (0.9 ml, 9 mmol) and the resulting clear solution was stirred at room temperature under nitrogen for 16 h. The reaction mixture was concentrated *in vacuo* and the yellow residue was placed under high vacuum for 2 h. Water (2 ml) was added, causing immediate formation of a white precipitate. Acetone (2 ml) was added and the white slurry was stirred at room temperature for 2 h. The solid was collected by filtration, washing twice with acetone (2 ml) and anhydrous ether (2 ml) and dried *in vacuo*. This produced 150 mg of product (3) having a radiochemical purity of 99.1% and specific activity of 88.7 μCi/mg.

SYNTHETIC PATHWAY



*** position of radiolabel**

RESULTS AND DISCUSSION

Synthesis of [^{14}C]PMEA (3) was achieved by coupling of 2-(diisopropylphosphonylmethoxy)ethanol methanesulfonate with 8-[^{14}C]adenine followed by deprotection of the phosphoric acid moiety. The alkylation reaction was carried out by treatment of the sodium salt of 8-[^{14}C]adenine with 2-(diisopropylphosphonylmethoxy)ethanol methanesulfonate in N,N-dimethylformamide at 100°C to provide the diisopropyl ester. Final deprotection was accomplished by treatment with bromotrimethylsilane in acetonitrile. Concentration of the reaction mixture, followed by treatment of the residue with water and acetone afforded [^{14}C]PMEA (3) as a crystalline solid having a radiochemical purity of 99.1% and a specific activity of 88.7 μCi/mg in an overall yield of 30%. All experimental conditions were optimized using non-radiolabeled materials.

REFERENCES

1. De Clercq, E., T. Sakuma, M. Baba, R. Pauwels, J. Balzarini, I. Rosenberg, and A. Holy. (1987). Antiviral activity of phosphonylmethoxyalkyl derivatives of purine and pyrimidines. Antiviral Res., 8:261-272.

2. Pauwels, R., J. Balzarini, D. Schols, M. Baba, J. Desmyter, I. Rosenberg, A. Holy, and E. De Clercq. (1988) Phosphonylmethoxyethyl purine derivatives, a new class of anti-human immunodeficiency virus agents. Antimicrob. Agents Chemother., 32:1025-1030.

3. Bronson, J.J., C.U. Kim, I. Ghazzouli, M.J.M. Hitchcock, E.R. Kern, and J.C. Martin. (1989) Synthesis and antiviral activity of phosphonylmethoxyethyl derivatives of purine and pyrimidine bases. In "Nucleotide Analogues as Antiviral Agents" (Martin, J.C., ed). American Chemical Society, Washington, D.C.

4. Balzarini, J., L. Naesens, P. Herdewijn, I. Rosenberg, A. Holy, R. Pauwels, M. Baba, D.G. Johns, and E. De Clercq. (1989) Marked in vivo antiretrovirus activity of 9-(2-phosphonylmethoxyethyl) adenine, a selective anti-human immunodeficiency virus agent. Proc. Natl. Acad. Sci. USA, 86:332-336.