SYNTHESIS OF 8-CHLORO-6-PHENYL-4H-s-TRIAZOLO [4,3-A] [1,4]BENZODIAZEPINE-1-^{14}C.

Nobuyoshi Hayashi, Kazuo Shinozaki, Shinji Kato,
Kanji Meguro and Yutaka Kuwada.
Chemical Research Laboratories, Central Research Division,
Takeda Chemical Industries, Ltd.
Juso, Higashiyodogawa-ku, Osaka, Japan.
Received on October 5, 1973.

SUMMARY

8-Chloro-6-phenyl-4H-s-triazolo [4,3-a] [1,4] benzodiazepine (D-40TA)-1-^{14}C (V-^{14}C) was synthesized by the reaction of formic acid-^{14}C with 7-chloro-2-hydrazino-5-phenyl-3H-1, 4-benzodiazepine in the presence of N,N'-carbonylbis (2-methylimidazole). The overall radiochemical yield from formic acid-^{14}C to V-^{14}C was 57.8%.

8-Chloro-6-phenyl-4H-s-triazolo[4,3-a][1,4]benzodiazepine (D-40TA) (V), a new benzodiazepine derivative prepared in these Laboratories[1], has potent central nervous system depressant activity[2]. In order to investigate the metabolic fate of V in animals, the labelled compound was requested. According to the preliminary test on the stability, compound V was considerably stable in both acidic and alkaline media, though some cleavage of $N_{(5)}$-$C_{(6)}$ double bond was observed in acidic medium. These data indicated that the triazolylbenzophenone skeleton would be stable during metabolism of this drug. Based on these considerations, as well as those of synthetic route, yield, and economy, the $C_{(1)}$-position was chosen as the site of choice for ^{14}C label.

Although V can be prepared by treating 7-chloro-2-hydrazino-

5-phenyl-3H-1,4-benzodiazepine (III) with excess formic acid[1c], these reaction conditions are unfavorable for the synthesis of V-^{14}C due to consumption of large amount of radioisotope. If the amount of formic acid is decreased, however, the formation of

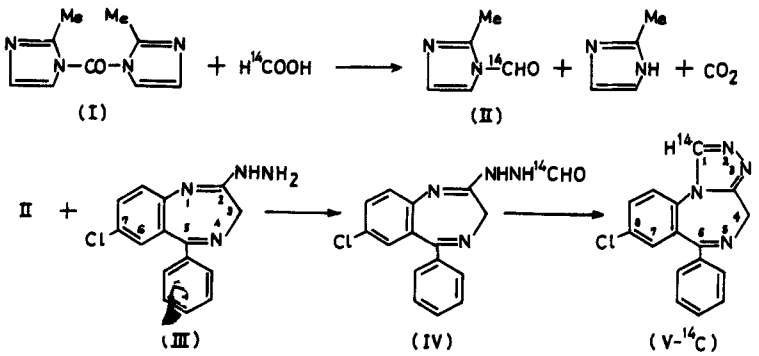

Chart 1.

a bis-type compound, N,N'-bis(7-chloro-5-phenyl-3H-1,4-benzodiazepin-2-yl)hydrazine[1] occurs; equimolar reaction of formic acid with III, for instance only gave the bis-type compound. In another trial run by Konig's method[3], i.e , reaction of III with a mixture of formic acid, DCC (dicyclohexylcarbodiimide) and 1-hydroxybenzotriazole, poor results were obtained. Finally V-^{14}C was synthesized by an application of Staab's method[4] using a formylimidazole as shown in Chart 1.

Formic acid-^{14}C in anhydrous THF (tetrahydrofuran) was reacted with N,N'-carbonylbis(2-methylimidazole) (I) at -20° to give

1-formyl-^{14}C-2-methylimidazole (II). Formylation of III with II in situ afforded 7-chloro-2-(2-formyl-^{14}C-hydrazino)-5-phenyl-3H-1,4-benzodiazepine (IV), which was cyclized to V-^{14}C by refluxing in pyridine The overall radiochemical yield from formic acid-^{14}C to V-^{14}C was 57.8% and the specific activity of V-^{14}C was 15.63 mCi/m.mol.

EXPERIMENTAL

Formic acid-^{14}C (specific activity, 59 mCi/m.mol.) was purchased from The Radiochemical Centre, Amersham, England.

Formic acid (special grade, 99%) used for dilution in experiment, was purchased from Wako Pure Chemical Industries, Ltd., Osaka, Japan.

7-Chloro-2-hydrazino-5-phenyl-3H-1,4-benzodiazepine (III)[1]:

Compound III was prepared by the reaction of 80% hydrazine hydrate with 2-amino-7-chloro-5-phenyl-3H-1,4-benzodiazepine[5,6] at 5 ~ 10° or with 7-chloro-1,3-dihydro-5-phenyl-2H-1,4-benzodiazepine-2-thione[7] at room temperature. III was obtained as colorless prisms after recrystallization from CH_2Cl_2-benzene, mp. 170° (browning), 202-204° (decomp.). Anal. Calcd. for $C_{15}H_{13}N_4Cl$: C, 63.27; H, 4.60; N, 19.68. Found: C, 63.43; H, 4.48; N, 19.27.

8-Chloro-6-phenyl-4H-s-triazolo[4,3-a][1,4]benzodiazepine-1-^{14}C (V-^{14}C):

To a solution of 45 mCi of formic acid-^{14}C and 0.082 ml of formic acid in 7.5 ml of anhydrous THF which was cooled at -20° in

a dry ice-acetone bath, 710 mg of N,N'-carbonylbis(2-methylimidazole) was added. After stirring for 30 min, a solution of 910 mg of III in 15 ml of anhydrous THF was added dropwise to the mixture and stirring continued for another 40 min at -20°. The solution was then warmed to room temperature and stirred for an additional 30 min in order to complete the reaction. The solvent was removed in vacuo and the residue dissolved in 5 ml of pyridine. The resulting solution was refluxed for 45 min and then evaporated in vacuo. After addition of water to the residue, the mixture was extracted with chloroform. The chloroform layer was washed with water, dried over sodium sulfate and concentrated in vacuo. The crystalline residue was washed with ether and chromatographed over 20 g of a silica gel column using chloroform-methanol (19:1, v/v) as an eluent. Recrystallization from acetone-n-hexane afforded 490 mg of radiochromatographycally pure V-^{14}C, *mp* 227-228°, specific activity 15.63 mCi/m.mol, radiochemical yield 57.8%, which was identical in every respect with an authentic unlabelled sample of V[1].

Radioactivity

A liquid scintillator was prepared by dissolving 4 g of PPO (2,5-diphenyloxazole) and 100 mg of POPOP (1,4-bis(5-phenyloxazol-2-yl)benzene) in 1 L of toluene. A sample solution was prepared by dissolving 0.1 mg of counting sample in 100 ml of toluene. A mixture of 15 ml of the liquid scintillator and 1 ml of the sample solution was counted in Alkoa LSC-502 liquid scintillation spectrometer with five channel systems and automatic channel ratio computor. Absolute counting efficiency was determined by comparison

with a standard calibration curve by the external standard method. The radiochemical purity of V-^{14}C was checked radiochromatographically by means of Alkoa thin-layer chromatogram scanner (Model TRM-1B) in the following system : TLC; Spotfilm, Silicagel f, Tokyokasei Ltd., Tokyo, Japan, developing solvent; $CHCl_3$:AcOEt: MeOH (19:1:2, v/v).

ACKNOWLEDGEMENT. The authors wish to acknowledge to Dr. S. Tatsuoka, Director of this Division, for his encouragement.

REFERENCES.

1) a) MEGURO, K. and KUWADA, Y. - Tetrahedron Letters, 1970, 4039.

b) idem - Heterocycles V, Chem. Pharm. Bull. (Tokyo), in press.

c) MEGURO, K., TAWADA, H., MIYANO, H., SATO, Y. and KUWADA, Y. - Heterocycles VI, ibid., in press.

2) NAKAJIMA, R., HATTORI, C. and NAGAWA, Y. - Japan. J. Pharmacol., 21, 489 (1971); NAKAJIMA, R., TAKE, Y., MORIYA, R., SAJI, Y., YUI, T. and NAGAWA, Y. - ibid., 21, 497 (1971).

3) KÖNIG, W. and GEIGER, R. - Chem. Ber., 103, 788 (1970).

4) STAAB, H. A. and POLENSKI, B. - Ann. Chem., 655, 95 (1962).

5) STERNBACH, L. H., REEDER, E., KELLER, O. and METLESICS, W. - J. Org. Chem., 26, 4488 (1961).

6) MEGURO, K., TAWADA, H. and KUWADA, Y. - Heterocycles II, Yakugaku Zasshi, 93, 1253 (1973).

7) ARCHER, G. A. and STERNBACH, L. H. - J. Org. Chem., 29, 231 (1964).