SYNTHESIS OF TRITIUM LABELED MAZINDOL

Harry A. Dugger and Kenrick C. Tabot
Drug Metabolism Section, Sandoz Pharmaceuticals, Sandoz-Wander, Inc., East Hanover, New Jersey 07936 U.S.A.

Received on November 11, 1975
Revised on February 12, 1976

SUMMARY

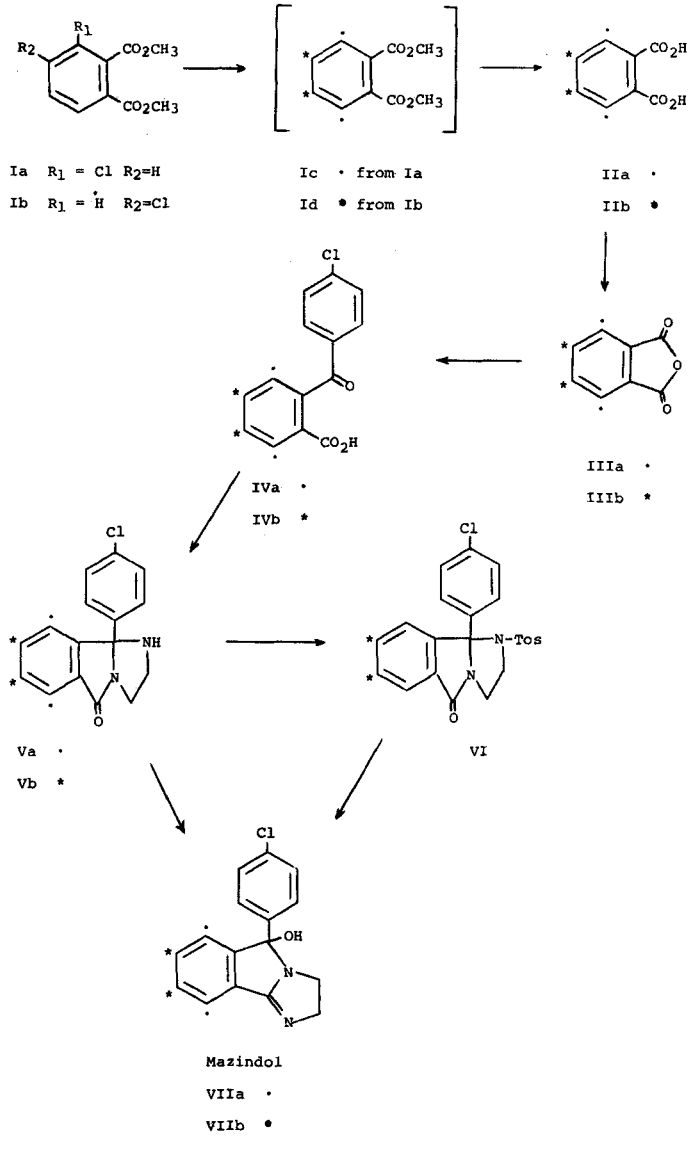
Mazindol was labeled with tritium in the 6,9 positions in low yield starting with dimethyl phthalate-3,6-^{3}H which was prepared by reduction of dimethyl 3-chloro-phthalate with tritium gas using 10% palladium on charcoal as a catalyst. The labeled dimethyl phathalate was hydrolisized with extensive loss of the label by exchange and after conversion to phthalic anhydride was condensed with chlorobenzene to give 2-(p-chlorobenzoyl) benzoic acid-3,6-^{3}H. This acid was converted in two steps into 5-hydroxy-5-(p-chlorophenyl)-2-3-dihydro-5H-imidazo [2,1-a] isoindol-6,9-^{3}H (mazindol-6,9-^{3}H). In a similar fashion, mazindol-7,8-^{3}H was prepared in higher yields beginning with dimethyl 4-chlorophthalate. There was no loss of the label during the hydrolysis of dimethyl phthalate-4,5-^{3}H.

Key Words: Mazindol, Tritium, Catalylic Reduction

Mazindol, 5-hydroxy-5-(p-chlorophenyl)-2,3-dihydro-5H-imidazo-[2,1-a]isoindole, is a new anorexic substance whose pharmacology has recently been reported by Gogerty et al. (1). The drug was labeled with tritium to facilitate studies of its metabolism and the pharmacokinetics of it and its metabolites by the two synthetic routes outlined in chart I. The sequence Ia —— IIa —— IIIa —— IVa —— Va —— VIIa (mazindol-6,9-^{3}H), in absence of metabolic data, was developed to place the label in what was hoped to be a relatively inert position in terms of metabolism. This route suffered from one disadvantage in that some or all of the label in IIa could be lost by exchange with the solvent during hydrolysis of the intermediate dimethyl phthalate-3,6-^{3}H. Under optimum conditions about one half of the label was lost. Later as more labeled mazindol was required and when data was available to show that little if any metabolism occurred in the ring containing the label in VIIa, a new route Ib —— IIb —— IIIb —— IVb —— Vb —— VIb —— VIIb (mazindol-7,8-^{3}H) was devised which had no loss of the label on hydrolysis of the ester and much improved yields overall.

The label was introduced by reductive dehalogenation of the corresponding 3-chloro or 4-chlorophthalic acid dimethyl ester. The conditions used to prepare 2-(p-chlorobenzoyl)benzoic acid were those described by Groggins and Newton (2). The preparation of V and its rearrangement to VII were carried out as described by Houlihan and Aeberli (3). The reaction of Vb with tosyl chloride to give VI and its further reaction to give VIIb proceeded as described by Sulkowski (4).

The overall radiochemical yield from the first synthesis (*) was 2.5% while that of the second one (*) was 17.5%. In each case, the product was shown to be chemically and radiochemically pure (96-99%).



The label in both cases was found to be stable to acid and base catalysed exchange and stable to metabolic attack in mouse, rat, rabbit, dog, monkey and man (5).

EXPERIMENTAL

Phthalic Acid-3,6-^{3}H (IIa) - 1.30 g of 3-chlorophthalic acid

dimethyl ester, 0.275 g 10% palladium on carbon, 0.95 g triethylamine and 25 ml dry tetrahydrofuran were combined in a reduction flask. The flask was evacuated and 15 curies of tritium and diluent hydrogen were introduced with shaking. After the uptake of hydrogen had ceased, the catalyst was removed by filtration and was washed with dry tetrahydrofuran. Exchangeable radioactivity was removed by successive additions of methanol followed by removal in vacuo. The residue was diluted with 2.3 g of non-labeled phthalic acid dimetnyl ester, 18 ml of 2N sodium hydroxide and 30 ml of acetone and heated at 60°C for one hour. About one half of the tritium appeared as tritium labeled water formed during the hydrolysis procedure. To minimize this loss the reaction time was kept as short as possible. The solution was evaporated to dryness in vacuo, and the residue was dissolved in water. After treatment with charcoal, the filtrate was acidified with concentrated hydrochloric acid and seeded to yield 1.99 g of phthalic acid-3,6-^{3}H. The chemical yield was 68% while the radiochemical yield was 4.15 curies, 23% (maximum theoretical yield was 50%).

Phthalic acid-4,5-^{3}H (IIb) - An analogous procedure to that described above beginning with 1.54 g of 4-chlorophthalic acid dimethyl ester but diluting the reduction product with only 1.0 g of non-labeled dimethyl phthalate gave 1.73 g of phthalic acid-4,5-^{3}H. In this case, the hydrolysis of the ester was carried out at 60° for 5 hours since no exchange of radioactivity occurred. The chemical yield was 88% and the radiochemical yield was 6.2 curies, 42% (maximum theoretical yield was 50%).

Phthalic Anhydride-3,6-^{3}H (IIIa) - A mixture of 1.99 g of phthalic acid-3,6-^{3}H, and 2 g of acetic anhydride was heated at reflux for 3 hours, cooled, and on dilution with ethyl acetate gave 1.37 g of phthalic anhydride-3,6-^{3}H in two crops. The radio-

chemical yield was 3.55 curies, 86%.

Phthalic Anhydride-4,5-^{3}H (IIIb) - Using the same procedure 1.73 g of phthalic acid-4,5-^{3}H was converted into 1.15 g of phthalic anhydride-4,5-^{3}H. Additional radioactive product, 0.40 g was diluted from the mother liquor with 0.50 g of non-labeled phthalic anhydride. Total radiochemical yield was 4.4 curies, 71%.

2-(p-Chlorobenzoyl)benzoic acid-3,6-^{3}H (IVa) - A solution of 1.34 g of phthalic anhydride-3,6-^{3}H in 37 ml of chlorobenzene was added to a slurry of 3.53 g of anhydrous aluminum trichloride in 5 ml of chlorobenzene at 95-100°C over a time period of 1.5 hours. The mixture was stirred overnight at 100°C, cooled, neutralized with 2N sodium hydroxide and diluted with water. This mixture was evaporated to dryness *in vacuo*, and the residue was partitioned between 5N hydrochloric acid and chloroform. After washing with water, the chloroform layers were combined, dried over anhydrous sodium sulfate, and evaporated to dryness *in vacuo*. Crystallization of the residue from ether:petroleum ether gave 1.63 g of 2-(p-chlorobenzoyl)benzoic acid-3,6-^{3}H. The radiochemical yield was 2.5 curies (70%).

2-(p-Chlorobenzoyl)benzoic acid-4,5-^{3}H (IVb) - Using the analogous procedure 1.63 g of phthalic anhydride-4,5-^{3}H gave 2.26 g of 2-(p-chlorobenzoyl)benzoic acid-4,5-^{3}H. The mother liquor was diluted with 3.0 g of non-labeled 2-(p-chlorobenzoyl)benzoic acid to yield a second crop of crystals, 2.41 g. The radiochemical yield was 4.4 curies (100%).

9b-(p-Chlorophenyl)-1,2,3,9b-tetrahydro-5H-imidazo-[2,1-a]isoindol-5-one-6,9-^{3}H (Va) - 1.616 g of tritiated 2-(p-chlorobenzoyl)-benzoic acid-3,6-^{3}H, 0.043 g of p-toluenesulfonic acid monohydrate, 0.55 g of ethylenediamine and 51 ml of toluene were combined in a

flask and warmed slowly over 3/4 hour to reflux temperature. After refluxing 24 hours, the reaction was filtered and the solvent removed in vacuo. The product was crystallized from 12 cc. of 2-propanol to give 1.35 g of Va, 1.58 curies (61% radiochemical yield).

9b-(p-Chlorophenyl)-1,2,3,9b-tetrahydro-5H-imidazo-[2,1-a] isoindol-5-one-7,8-^{3}H (Vb) - 2.253 g of 2-(p-chlorobenzoyl)benzoic acid-4,5-^{3}H was added to a stirred solution of 1.73 ml of ethylenediamine and 0.45 ml of water. This mixture was heated at reflux for 2.5 hours, cooled and diluted with 4.5 ml of water. The resultant precipitate was washed thoroughly with water, dried, and crystallized from 2-propanol to give 1.70 g of Vb in two crops. The radiochemical yield was 3.1 curies (77%).

5-Hydroxy-5-(p-chlorophenyl)-2,3-dihydro-5H-imidazo[2,1-a]isoindole-6,9-^{3}H (VIIa) - A solution of 1.347 g of Va in 15 ml of dry tetrahydrofuran was added dropwise during 1/2 hour to a slurry of 0.240 g of lithium aluminum hydride. The temperature was maintained at 25° or less. After stirring 6 hours at room temperature, the reaction was cooled to 10° and 0.5 ml of 2N NaOH and 0.7 ml of water were added. After stirring 1/4 hour, 5.0 g of anhydrous Na_2SO_4 were added and the resulting suspension was stirred an additional 1/4 hour at which time the solid material was removed by filtration and the filtrate concentrated in vacuo. The residue was taken up in methanol and refluxed for 1/2 hour. The methanol was in turn removed in vacuo and the residue was taken up in ether and seeded with pure non-labeled VII. After standing overnight, the product was removed by filtration and the process described above repeated to give other fractions. Total yield was 0.431 g of mazindol-6, 9-^{3}H (VIIa), 472 mc, 30%. The specific activity was 312 mc/mM or 1.09 curies/g.

The product was shown to be chemically pure by comparison of its infrared spectrum measured in KBr with that of an authentic sample. The preparation was shown to be radiochemically pure by radio-thin-layer chromatography on silica gel in two solvent systems: chloroform-methanol (8:2) and butanol-water-acetic acid (4:1:5). In the first system, 99% of the label was found within $\pm$ 0.1 R_f unit of the center of the spot due to the standard substance. The corresponding amount was 96% in the second system.

5-Hydroxy-5-(p-chlorophenyl)-2,3-dihydro-5H-imidazo[2,1-a]isoindole-7,8-^{3}H (VIIb) - A mixture of 1.678 g of 9b-(p-chlorophenyl)-1,2,3,9b-tetrahydro-5H-imidazo-[2,1-a]isoindol-5-one-7,8-^{3}H, 1.678 g of p-toluenesulfonyl chloride, and 205 g of pyridine was refluxed for 15 hours. The solvent was removed *in vacuo*, and at 20° the residue was triturated with 13 ml of water. After stirring for 15 minutes, the slurry was filtered. The residue was washed with three 12 ml aliquots of water, two 3 ml aliquots of 95% ethanol and dried *in vacuo* at 50° to give 2.414 g of 9b-(4'-chlorophenyl)-1,2,3,9b-tetrahydro-1-(p-tolylsulfonyl)-5H-imidazo-[2,1-a]isoindol-5-one-7,8-^{3}H (VI), which was added in portions to 8.1 ml of 96.5% sulfuric acid. After stirring for 1 hour at room temperature, the reddish brown solution was poured slowly into 144 ml of water at a temperature range of 20-25°. After vacuum filtration, the filtrate was diluted with 35 g of ice and was basified to pH 8 with 50% sodium hydroxide solution. The white slurry formed was stirred for 15 minutes, filtered, and the precipitate was washed with five 5.5 ml aliquots of water followed by 5 ml of acetone. After drying *in vacuo* at 50°, this precipitate was diluted with 1.06 g of non-labeled VII and, following treatment with charcoal, was crystallized from tetrahydrofuran: methanol to give 2.25 g of mazindol-7,8-^{3}H (VIIb) in two crops. The radiochemical yield was 2.35 curies (76%) and the specific activity was 295 mc/mM or 1.04 curies/g.

The chemical purity of this product was also demonstrated by comparison of its infrared spectrum measured in KBr with that of an authentic sample. The radiochemical purity of 97-98% was shown by radio-thin-layer chromatography on Silica Gel G in the solvent system methanol:chloroform (1:3). Radiochemical purity of 97-98% also was demonstrated by reverse isotope dilution.

Stability of the Label to Exchange in Acidic or Basic Medium - VIIa or VIIb, (2 mg of labeled material and 20 mg of non-labeled material) was refluxed 1/2 hour in 50 ml of ethanol-water, 4:1, and 0.2N formic acid or sodium hydroxide. The solvent was removed in vacuo and the radioactivity of the residue and the distillate was measured. In both cases, there was less than 0.01% of the radioactivity in the distillate.

REFERENCES

1. J.H. Gogerty, C. Penberthy, L.C. Iorio, and J.H. Trapold, Arch. intern. pharmacodynamic, in press; R.G. Engstrom, L.A. Kelly and J.H. Gogerty, ibid, in press.
2. P.H. Groggins and H.P. Newton, Ind. and Eng. Chem. 21,369 (1929).
3. W.J. Houlihan and P. Aeberli, Private communication.
4. T.S. Sulkowski, Belgian Patent #712,784, Published July 31, 1968.
5. H.A. Dugger, H.J. Schwarz and J.H. Heider, Drug Metab. Disp., in press.