

SYNTHESIS OF ^3H - AND ^{14}C -LABELLED *o*-(β -MORPHOLINOETHOXY)-DIPHENYL-ETHER HYDROCHLORIDE.

Tohru HORIE and Takeshi FUJITA.

Department of pharmacology, Eisai Co., Ltd.

4-chome, Koishikawa, Bunkyo-ku,

Tokyo, Japan.

Received on February 29, 1972.

SUMMARY

*The methods for preparation of ^{14}C or ^3H labelled compound of *o*-(β -morpholinoethoxy)-diphenylether hydrochloride (MPE-HCl) have been investigated. (1) Starting from guaijacol and ^{14}C -bromobenzene (U), the synthesis of MPE-phenyl- ^{14}C (U)-HCl has been accomplished by a heterogeneous palladium-catalyzed exchange reaction in a yield of 60%.*

**o*-(β -Morpholinoethoxy)-diphenylether hydrochloride (MPE-HCl) has been found to be a new specific cardiovascular stimulant which acts possibly on a vasomotorcenter^(1,2). In order to fully study the metabolic transformation of MPE-HCl, the labelled compound was required.*

*In the preparation of ^{14}C -labelled MPE-HCl from ^{14}C -bromobenzene (U), a modified method reported by Toyoshima et al.⁽³⁾, as outlined in Figure 1, was used. *o*-Methoxydiphenylether-phenyl- ^{14}C (U) [II] was obtained by condensation of bromobenzene- ^{14}C (U) [I] and guaijacol. *o*-Hydroxydiphenylether-phenyl- ^{14}C (U) [III] derived from [II] was allowed to react with 2-morpholinoethylchloride*

hydrochloride. After purification of the crude product by silicic acid column chromatography, *o*-(β -morpholinoethoxy)-diphenylether-phenyl- ^{14}C (U) [IV] as a HCl salt, of which specific activity was $8.4 \mu\text{Ci}/\text{mg}$, was isolated in a yield of 21 % from [I].

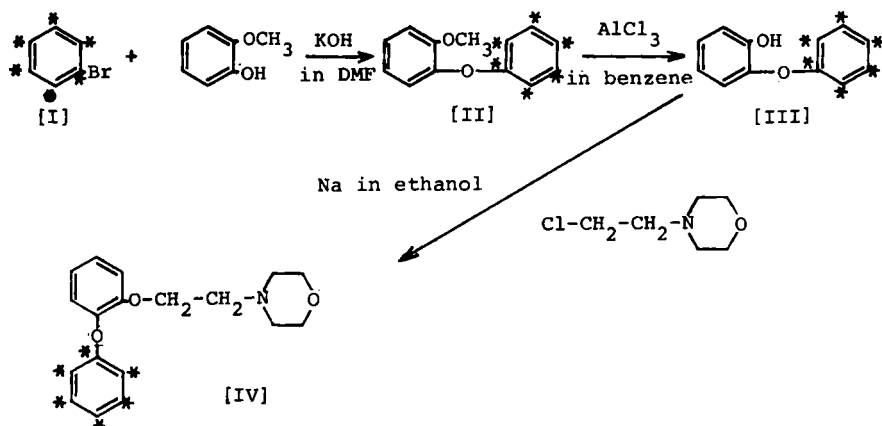


Fig. 1. Outline of synthesis of *o*-(β -morpholinoethoxy)-diphenylether.

star marks show position of C-14.

On the other hand, the catalytic tritiation of MPE-HCl with T_2O afforded pure ^3H -MPE-HCl, which had $33.3 \mu\text{Ci}/\text{mg}$ in a specific activity, following a purification of silicic acid column chromatography, in a yield of 60 %. This tritiation was found to be suitable for preparation of the high radioactive ^3H -MPE-HCl. This paper is concerned with the synthesis and purification of ^{14}C and ^3H labelled MPE-HCl.

EXPERIMENTAL

(i) o-(8-Morpholinoethoxy)-diphenylether-phenyl-¹⁴C(U).

o-Methoxydiphenylether-phenyl-¹⁴C(U) [II].

A mixture of guaijacol (382 mg, 3.08 mmole) and powdered potassium hydroxide (191 mg, 3.41 mmole) was heated on a steam bath for three hours under reduced pressure. To the mixture, copper powder (20 mg, 0.32 mmole), copper iodide (20 mg, 0.11 mmole) and then [I] (224 mg, 1.43 mmole, 5 mCi obtained from Daichi Pure Chemicals Co., Ltd., Tokyo) dissolved in 2.5 ml of dimethylformamide were added in succession.

The mixture was stirred on oil bath at 140-150° for twenty hours under nitrogen stream. The reaction mixture was poured into 20 ml of water and the water phase was extracted three times with 100 ml of ethylether. The ethereal solution was washed with 0.5 N NaOH aqueous solution and then twice with 50 ml of water. Guaijacol was almost removed by such a procedure. After being dried on granulated anhydrous MgSO₄, the ethereal solution was evaporated in vacuo.

The residue dissolved in a small amount of chloroform was subjected to a column chromatography (2 x 40 cm) using silicic acid (100 mesh, obtained from Mallinckrodt Chemicals, Works) as an adsorbent and benzene as an elution solvent. Following evaporation of the fraction containing [II] alone, which was checked on TLC (Kieselgel G, 0.25 mm in thickness, solvent : benzene), 230 mg of o-methoxy-diphenylether-phenyl-¹⁴C(U) [II] was obtained in a yield of 82 % from [I].

o-Hydroxydiphenylether-phenyl- ^{14}C (U) [III].

A mixture of anhydrous aluminium chloride (400 mg, 3.01 mmole) and [II] (230 mg, 1.15 mmole) dissolved in 3 ml of benzene was refluxed for two hours with stirring under nitrogen stream. Following dryness of the reaction mixture in vacuo, 10 ml of 1 N HCl aqueous solution was added to the residue. The precipitates were collected and dried overnight on CaCl_2 in vacuo. 203 mg of *o*-hydroxydiphenylether-phenyl- ^{14}C (U) [III] was obtained in a yield of 95 % from [II].

o-(β -Morpholinoethoxy)-diphenylether-phenyl- ^{14}C (U) hydrochloride (^{14}C -MPE-HCl).

Metal sodium (143 mg, 6.23 mmole) was dissolved in 3 ml of ethanol containing [III] (203 mg, 1.09 mmole) with stirring. After 2-morpholinoethylchloride hydrochloride (532 mg, 3.56 mmole) was added, the mixture was refluxed for eight hours with stirring under nitrogen atmosphere, then evaporated in vacuo.

A solution of the residue dissolved in 10 ml of 1 N HCl was washed with 100 ml of ethylether, and alkalized with a small amount of solid sodium hydroxide, then extracted four times with 50 ml of ethylether. The ethereal solution was washed with water, and dried on granulated anhydrous MgSO_4 . The residue, which resulted from evaporation of the solution, was applied on a silicic acid column chromatography (2 x 40 cm) using a mixture of chloroform and acetone (6 : 4) as eluant. The effluent containing [IV] was collected and evaporated in vacuo.

112 mg of *o*-(β -morpholinoethoxy)-diphenylether-phenyl- ^{14}C (U) [IV] was obtained in a yield of 37 % from [III]. 10 ml of ethanol

saturated with HCl gas was added to 112 mg of [IV]. After evaporation of solvent, the residue was recrystallized from acetone. 124 mg of ^{14}C -MPE-HCl was obtained. Measurement of radioactivity was carried out using Aloka LSC-601 Type Liquid Scintillation Counter. ^{14}C -MPE-HCl had a specific activity of 8.4 $\mu\text{Ci}/\text{mg}$.

The overall chemical and radiochemical yield based on bromobenzene and bromobenzene- $^{14}\text{C}(\text{U})$ were 26 % and 21 %, respectively. The chemical and radiochemical purities of ^{14}C -MPE-HCl were confirmed by UV spectrum and TLC on Kieselgel G using two solvent systems.

The UV spectrum of ^{14}C -MPE-HCl ($\lambda_{\text{max}}^{\text{EtOH}} = 271, 277 \text{ nm}$) was quite identical with that of the authentic sample, as shown in Figure 2.

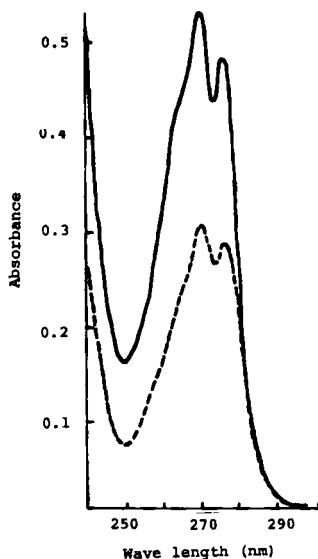


Fig. 2. UV spectra of *o*-(β -morpholinoethoxy)-diphenylether- $^{14}\text{C}(\text{U})$ hydrochloride and the authentic sample.

— : MPE-HCl 18.2 $\mu\text{g}/\text{ml}$ EtOH.

- - - : ^{14}C -MPE-HCl 10.5 $\mu\text{g}/\text{ml}$ EtOH.

The radiochromatograms of [IV] recorded using Aloka Thin Layer Chromatogram Scanner Model TLC-2B were indicated in Figure 3. A peak of radioactivity was located at the same region, where a coloured spot of the authentic sample was observed under iodine exposure, on TLC developed with the solvent systems of chloroform/acetone, 6/4 ($R_f = 0.68$) and water/n-buthanol/acetic acid, 5/4/1 ($R_f = 0.49$).

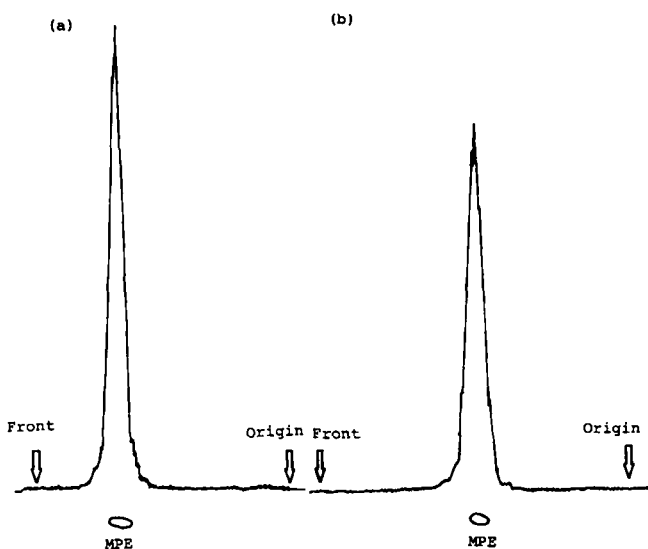


Fig. 3. Radioscans of *o*-(β -morpholinoethoxy)-diphenylether-phenyl- $^{14}\text{C}(\text{U})$ on TLC developed with (a) chloroform/acetone 6/4 (b) water/n-buthanol/acetic acid 5/4/1.

(ii) *o*-(β -Morpholinoethoxy)-diphenylether-³H(G) (³H-MPE-HCl).

Tritium water (0.2 ml, 1.0 Ci) and 10 % palladium on charcoal (50 mg) were added to a solution of MPE-HCl (100 mg, 0.30 mmole) dissolved in 1.0 ml of methanol. The mixture was heated at 120° for fifteen hours in a sealed ampule. Following evaporation of T₂O, labile tritium from the residue was removed by repeated addition of methanol and evaporation in vacuo. Crude ³H-MPE base was extracted with ethylether from a solution of the crude tritiated MPE-HCl dissolved in 0.1 N NaOH. At least two radioactive peaks except for that of ³H-MPE were found on radiochromatograms of crude ³H-MPE obtained, as shown in Figure 4.

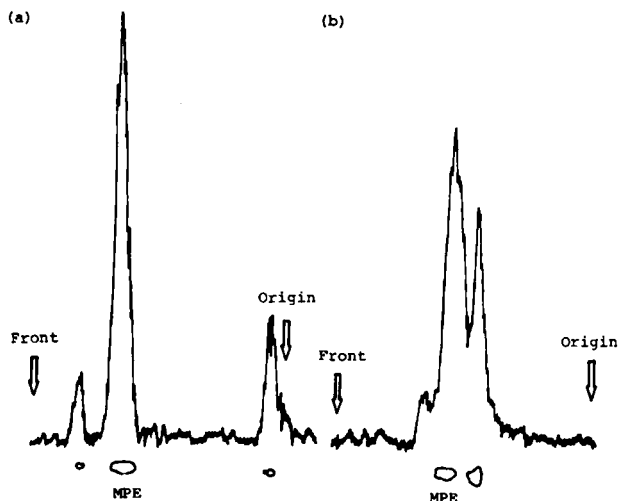


Fig. 4. Radioscans of the crude *o*-(β -morpholinoethoxy)-diphenylether-³H(G) on TLC. TLC : Kieselgel G (0.25 mm in thickness). Solvent system : (a) : chloroform/acetone = 6/4. (b) : H₂O/n-butanol/acetic acid = 5/4/1.

Purification of ^3H -MPE was achieved by silicic acid column chromatography, eluting with a mixed solvent of chloroform and acetone (6 : 4), as described above. ^3H -MPE isolated was confirmed to be pure on TLC, as shown in Figure 5. 60 mg of ^3H -MPE as a hydrochloride salt was obtained and has a specific activity of 33.3 $\mu\text{Ci}/\text{mg}$

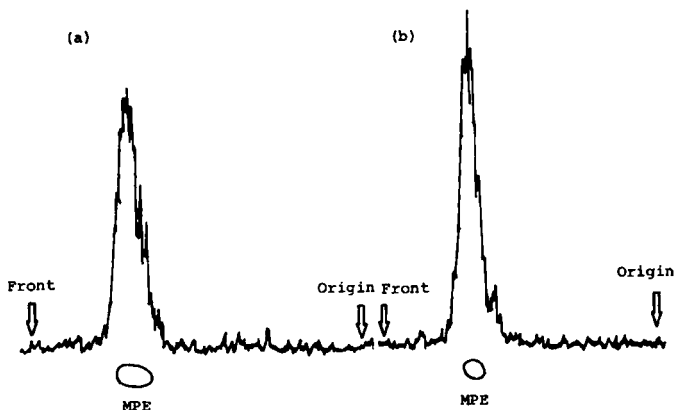


Fig. 5. Radioscans of *o*-(β -morpholinoethoxy)-diphenylether- ^3H (G) on TLC.
 TLC : Kieselgel G (0.25 mm in thickness).
 Solvent system :
 (a) : chloroform/acetone = 6/4.
 (b) : $\text{H}_2\text{O}/n$ -buthanol/acetic acid = 5/4/1.

ACKNOWLEDGMENTS.

The authors are grateful to Dr K. Miyao, director of the department of pharmacology, for his kind encouragement and Mr. S. Tanaka for his valuable advice in this research.

REFERENCES.

1. IGARASHI, T. and NAKAJIMA, Y.- *Autonomic Nervous System*, 5 : 59 (1968).
2. IGARASHI, T. and HASHIMOTO, K.- *Folia pharmacologica Japonica*, 66 :131 (1970).
3. TOYOSHIMA, S., TANAKA, S., OHGOH, T. and HASHIMOTO, K., -*Yakugaku zasshi*, 89 : 1417 (1968).