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A Procedure for Preparing ^{3}H-Labeled Tertiary Amines. Synthesis of [^{3}H]-6,14-*endo*-Etheno-6,7,8,14-tetrahydrooripavine Derivatives

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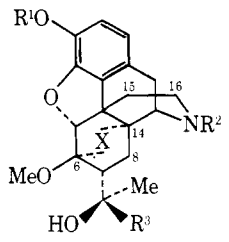
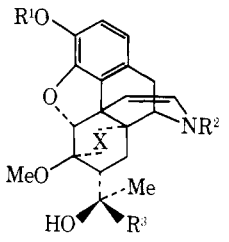
During radiosynthesis it is of paramount importance that the label is introduced at as late a stage as possible. The concept of a synthesis based on the unlabeled required product as starting material is very attractive; techniques such as the Wilzbach gas exposure method[1] and base-catalyzed exchange of aromatic protons[2] illustrate this approach. Both of these methods, however, suffer from distinct disadvantages, the former in that the labeling is unspecific and the latter in that exchange labeled material is susceptible to loss of activity by the reverse exchange process.

In the course of the study of the metabolism of the series of oripavine analgesics **1**, it has been necessary to develop methods of labeling these compounds with tritium. Initially, etorphine (**1a**), a very potent analgesic used for immobilization of animals,[3] was labeled at C-8 by a long inefficient route.[4] We here report a method of introducing a tritium label at carbon atoms α and/or β to a tertiary nitrogen by dehydrogenation to the enamine (*e.g.*, **2**) into which tritium is readily exchanged. The labeled tertiary base is regenerated by subsequent reduction with or without further tritiation.† Use of such a route has the advantages of high specific activity and cheapness inherent in exchange reactions, together with the regiospecificity and stability required for biological studies. Recently, Portoghese[6] has described a somewhat analogous method of labeling secondary amines in the α position by utilizing the acidity conferred on the α protons by the introduction of an *N*-nitroso function.



1



2

	R^1	R^2	R^3	X
a	H	Me	*n*-Pr	-CH=CH-
b	H	$CH_2CH=CH_2$	*n*-Pr	-CH=CH-
c	H	CH_2-c-C_3H_5	Me	$-CH_2CH_2-$
d[a]	Me	CH_2-c-C_3H_5	Et	-CH=CH-
e	H	CH_2-c-C_3H_5	Bu	$-CH_2CH_2-$

[a]Thebaine derivative.

†We have previously reported brief details of the synthesis of [15-^{3}H]etorphine from the 15,16-didehydro compound **2a**.[5]

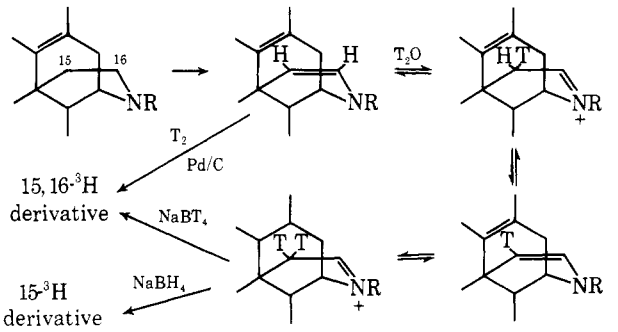
Table I. Specific Activities of Labeled Oripavine Derivatives

Labeling method	Compd	Product	Sp act., mCi/mmol
	1a	15-^{3}H	100, 220, 260
(i) T_2O (*ca.* 20 Ci)	**1b**	15-^{3}H	160
(ii) $NaBH_4$	**1c**	15-^{3}H	110
	1d	15-^{3}H	26[a]
	1e	15-^{3}H	230
(i) T_2O (*ca.* 20 Ci)	**1a**	15,16-^{3}H	900
(ii) $NaBT_4$ (250 mCi)	**1e**	15,16-^{3}H	1300
T_2 (*ca.* 10 Ci); Pd/C (10%)	**1a**	15,16-^{3}H	1500, 3600[b]

[a]T_2O used for this reaction was recovered from a previous experiment. [b]After isotope dilution.

The enamines **2**, prepared by the dehydrogenation of the parent tertiary bases **1** with mercury(II) acetate,[5,7] are strong bases and the proton at C-15 equilibrates rapidly with water. Nmr studies with **2e** showed that in $CDCl_3$ the AB quartet (δ 4.26 and 5.93, J = 8 Hz) due to H-15 and H-16 collapsed to a singlet (δ 5.95) on addition of D_2O. Thus, treatment of 15,16-didehydroetorphine (**2a**) with ^3H_2O yielded the enamine specifically labeled at C-15. Subsequent reduction of the iminium salt with $NaBH_4$ yielded [15-^{3}H]etorphine. If the reduction step is carried out in the presence of ^3H_2O, the resulting product contains, in theory, two labeled atoms at C-15 (Scheme I). Other tritiated compounds in the series have been synthesized in this way. The use of NaB^3H_4 yielded 15,16-^{3}H derivatives which showed increased specific activity (Table I).

Scheme I



Etorphine was also labeled in the 15 and 16 positions by hydrogenation of the enamine double bond with $^3H_2$ gas, using 10% palladized charcoal as catalyst; the 6,14-etheno bridge is not reduced under these conditions.[8] The labeled etorphine produced by this route had a higher specific activity than the products of the previous experiments (Table I).

The limiting value for the specific activity of the product in the hydrogenation route is 58 Ci/mmol and though such activities would be difficult to obtain in practice it is apparent that the latter approach is the method of choice in view of the high pharmacological activity of the series and the consequent need for high specific activity.‡ It may not, however, be applicable to compounds containing

‡It would be possible, in fact, to increase further the activity of the hydrogenation product by preliminary equilibration of the enamine with tritiated water to yield, after reduction, a triply labeled product.

the cyclopropylmethyl group as these undergo reductive ring opening under hydrogenation conditions.§

The methods outlined above should prove to be of use in other series of compounds in which similar enamines can be generated and isolated, *e.g.*, meperidine and its analogs,[9] dihydromorphines,[10] morphinans, and benzomorphans,[11] provided such compounds contain no functional groups reactive toward both catalytic hydrogenation and borohydride reduction.

Experimental Section

Specific activities were determined in a Packard Tricarb 3003 scintillation spectrometer. Samples were counted in a cocktail containing naphthalene (100 g), 2,5-diphenyloxazole (5.0 g), and 1,4-bis[2-(5-phenyl)oxazolyl]benzene (0.05 g) in 1,4-dioxane (1000 ml). Chemical and radiochemical purities were estimated by tlc on Kieselgel 60 F_{254} plates (5 × 20 cm) supplied by Merck Ag using the following solvent systems: (i) *n*-BuOH-AcOH-H_2O (20:5:8), (ii) $CHCl_3$-MeOH-H_2O (8:5:1), (iii) EtOAc-MeOH (75:25). Distribution of radioactivity on the plates was determined with a Tracerlab 4π scanner. Radiochemical starting materials were supplied by the Radiochemical Centre, Amersham, England. Experimental details are given for etorphine only. Other compounds were labeled in an analogous manner.

15,16-Didehydrooripavine Derivatives. 15,16-Didehydrooripavine derivatives were prepared from the parent tertiary bases by the method of Lewis, *et al.*[5]

6,14-*endo*-Etheno-7α-(1-hydroxy-1-methylbutyl)tetrahydro-[15-^{3}H]oripavine ([15-^{3}H]Etorphine). 2a (100 mg, 0.24 mmol) was placed in a round-bottomed flask (25 ml) attached to a vacuum bridge. ^3H_2O (*ca.* 20 Ci, 80 μl) was frozen in liquid nitrogen, dry dioxane (3 ml) added, and the mixture vacuum transferred to the reaction flask. The vacuum was released to allow the addition of $NaBH_4$ (150 mg, 3.9 mmol). The reaction mixture was stirred at ambient temperature for 2 hr and taken to dryness *in vacuo*, and K_2CO_3 (0.5 g) in H_2O (3 ml) and dioxane (3 ml) was added. The mixture was heated at 90° for 10 min and cooled, and the volatile materials were again removed *in vacuo*. H_2O (5 ml) was added and the mixture was extracted with $CHCl_3$. The $CHCl_3$ solution was washed repeatedly with H_2O, dried (Na_2SO_4), and taken to dryness to yield the crude product (60 mg). A portion of this material (14 mg) was chromatographed on a silica column (30 × 1 cm; Merck Ag Kieselgel 60, 0.063–0.200 mm) with Et_2O as eluent. Fractions which contained only the required product were united and treated with a little ethereal HCl to yield [15-^{3}H]etorphine hydrochloride (6 mg, 16%): sp act. 260 mCi/mmol; radiochemical purity >98%.

6,14-*endo*-Etheno-7α-(1-hydroxy-1-methylbutyl)tetrahydro-[15,16-^{3}H]oripavine ([15,16-^{3}H]Etorphine). Method a. [15,16-^{3}H]-Etorphine was synthesized by a route analogous to the method used for the 15-^{3}H derivative, NaB^3H_4 being used in the reduction step: sp act. 900 mCi/mmol; radiochemical purity of product >98%.

Method b. 2a (25 mg, 0.06 mmol) and 10% palladized charcoal (10 mg) in ethyl acetate (3 ml) was placed in a flask connected by a vacuum bridge to a break-seal ampoule containing $^3H_2$ gas (*ca.* 10 Ci; 4 ml at STP). The solution was cooled in liquid nitrogen and the system was evacuated and closed. The reaction mixture was exposed to $^3H_2$ gas by breaking the ampoule seal. The flask was allowed to warm to room temperature and the mixture stirred for 2 days, after which time hydrogen was introduced *via* the bridge to restore atmospheric pressure and the reaction was continued for a further 18 hr. The system was thoroughly evacuated, the catalyst removed by filtration, and the resulting solution was taken to dryness *in vacuo*. The crude product was chromatographed on a silica plate (20 × 20 × 0.2 cm) with *n*-BuOH-H_2O-AcOH (20:8:5) as eluent. The band corresponding to **1a** was eluted with CH_3OH (20 ml) and the resulting solution taken to dryness *in vacuo*. The residue was dissolved in cold 0.1 *N* HCl (5 ml) and the aqueous solution rapidly basified (K_2CO_3) and extracted with $CHCl_3$ (3 × 5 ml). The united $CHCl_3$ extracts were washed several times with water, dried (Na_2SO_4), and taken to dryness and the residue was dissolved in dry Et_2O (5 ml). Carrier **1a** (30 mg) was added and the solution was treated with a few drops of anhydrous ethereal HCl to yield [15,16-^{3}H]etorphine hydrochloride (45.8 mg, 48%): sp act. *ca.* 3.6 Ci/mmol; radiochemical purity >98%.

§H. P. Crocker, unpublished results.

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Synthesis and Biological Activity of 2′-Amino-2′-deoxy-5-fluorouridine

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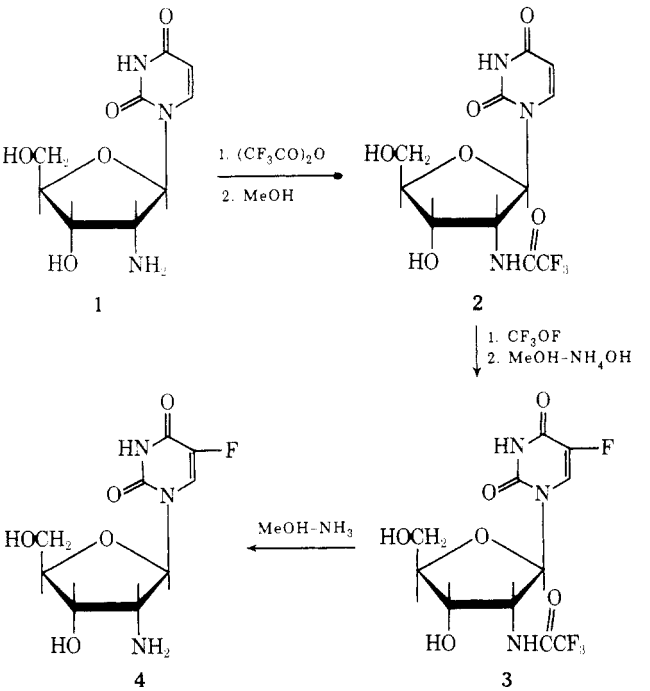
Recently, Verheyden, Wagner, and Moffatt[1] reported a useful synthesis of 2′-amino-2′-deoxyuridine and of its 5-iodo derivative. Although no information on the biological activity of these compounds has been published, it appeared worthwhile to us to prepare the 5-fluoro derivative of 2′-amino-2′-deoxyuridine, in view of the potent biological activity of other fluorinated pyrimidines.[2]



Chemical Results. Preparation of 2′-amino-2′-deoxy-5-fluorouridine (**4**) was first attempted by direct fluorination of 2′-amino-2′-deoxyuridine (**1**) with trifluoromethyl hypofluorite (CF_3OF), according to the methods described