

Short Research Article

Syntheses of isotopomers of SK&F-S-104864, topotecan HCl[†]

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Introduction

SK&F-S-104864-A **1**, topotecan HCl, is a topo-isomerase-I inhibitor marketed as Hycamtin.

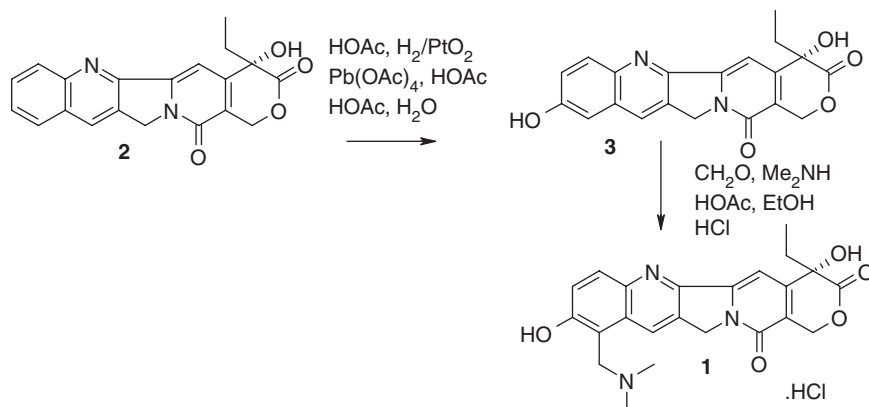
Topotecan¹ is a semi-synthetic derivative of camptothecin **2**, an alkaloid first isolated² in 1966. Camptothecin¹ is converted into the 10-hydroxy derivative **3** by a reduction/oxidation sequence and a subsequent Mannich reaction gives the desired SK&F-S-104864 (Scheme 1).

Results and discussion

Carbon-14 labelled SK&F-S-104864 was required for ADME studies. 10-Hydroxycamptothecin **3** is readily available from the synthesis of bulk drug. *N*-demethyl-

ation of the dimethylaminomethyl side chain is a known metabolic pathway,³ however, the exo-methylene carbon remains untouched so we chose to label with carbon-14 as shown in Scheme 2. 10-Hydroxycamptothecin was suspended in glacial acetic acid, dimethylamine added followed by [¹⁴C]formaldehyde. The mixture was stirred at 75°C until no further reaction was observed (HPLC). The crude SK&F-S-104864 was purified by reverse phase HPLC, the relevant fractions pooled and c. HCl added. The product was isolated by lyophilization giving [¹⁴C]SK&F 104864-A in 68% yield from formaldehyde.

[³H]Topotecan was required for mechanistic studies and is prepared in a directly analogous manner using [³H]formaldehyde as the source of radiolabel. The



Scheme 1

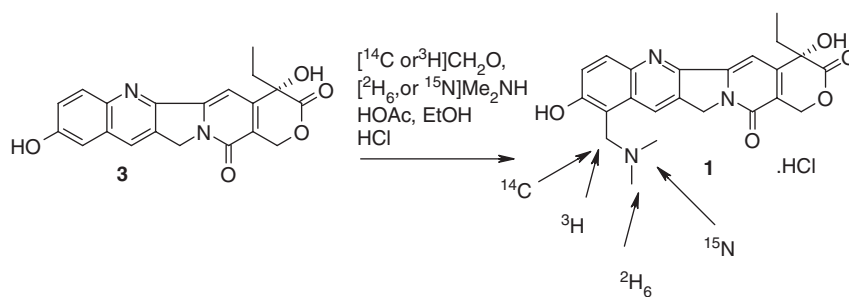
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crude product is purified by HPLC (C18, acetonitrile/water/TFA) and isolated as the TFA salt by lyophilization. The specific activity was 2.8 Ci/mmol.

Stable isotope labelled topotecan was required as an internal standard in LC/MS/MS assays. 10-Hydroxycamptothecin **3** was reacted in glacial acetic acid with aqueous formaldehyde and [²H₆] dimethylamine



Scheme 2

hydrochloride (3 equivalents) at 55°C until the reaction was complete. The residue was purified by medium pressure chromatography (RP18 silica, methanol/water 1:4 (v/v)). The product was dissolved in DCM/methanol (2:1 (v/v)) and a calculated amount of 10M HCl in IPA added. The product slowly precipitated. Typical yields from 10-hydroxycamptothecin were 50–60%. $[^{15}\text{N}]$ SK&F-S-104864 was required for NMR studies in support of pharmaceutical formulation, and was synthesised in a directly analogous manner to $[^2\text{H}_6]$ SK&F-S-104864.

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