



BDG SYNTHESIS

Certificate of Analysis

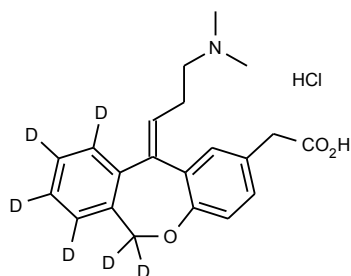
BDG Synthesis certifies that this reference material meets or exceeds the specifications stated herein.

Barry Dent

Barry R. Dent, PhD, Director
13 February 2014

Name: Olopatadine-d₆ HCl
CAS Number: 140462-76-6 (unlabelled)

Structure:



Molecular Weight: C₂₁H₁₇D₆NO₃·HCl = 379.91
Lot Number: BDG 9209
Appearance: White, crystalline solid
Corrected Purity: 99.7 % (HPLC) - 0.1 % (acetone) = 99.6 %
Isotopic Purity: Under 0.5 % d₀
Re-test Date: 13 February 2019
Storage and Handling: Temperature: refrigerate for prolonged storage; may be handled and shipped at ambient temperature.
Humidity: not believed to be hygroscopic; may be handled in normal laboratory atmosphere.
Light: protect from strong sunlight.
Caution: only experienced laboratory personnel should handle the material.

Identity and Purity

Proton NMR Spectrum

Identity: the signals are consistent with the proposed structure and in accord with literature where available.

Isotopic Labelling: signals at the sites of deuteration are greatly diminished, compared with the spectrum of unlabelled material, indicating clean deuteration.

Residual Solvents: a small amount of acetone (0.1 % w/w) is observed.

Impurities: no significant impurities are evident in the spectrum.

Carbon-13 NMR Spectrum

Identity: the signals are consistent with the proposed structure and in accord with literature where available.

Isotopic Labelling: signals at the sites of deuteration have collapsed to small multiplets compared with the spectrum of unlabelled material, indicating clean deuteration.

High-resolution Mass Spectrum (ESI+)

Found m/z 344.2125. $C_{21}H_{18}D_6NO_3$ $[M+H]^+$ requires m/z 344.2133. The deviation of 2.3 ppm is within normally accepted limits for the establishment of identity by HRMS. No signal for d_0 material was seen (detection limit about 0.5 %).

HPLC

A sharp, slightly tailing peak is observed (99.7 %). Note: in the absence of reference materials for preparing calibration curves, it is assumed that all peaks have the same detector response. Where possible, the conditions of analysis follow a pharmacopeial or literature method, or have been adapted from same.

Elemental Analysis

	Found:	C 66.61, H 5.05, D 3.37, N 3.68 %
$C_{21}H_{17}D_6NO_3 \cdot HCl$	Requires:	C 66.39, H 4.78, D 3.18, N 3.69 %

The elemental analyses fall within generally accepted limits for establishing the molecular formula given. The results may also be taken to imply the absence of significant quantities of water or inorganic salts (which have not been elsewhere tested for because of sample size limitations).

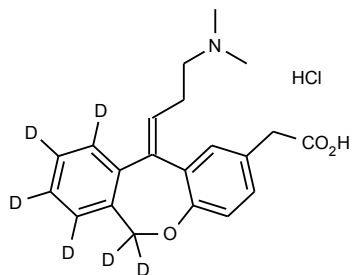
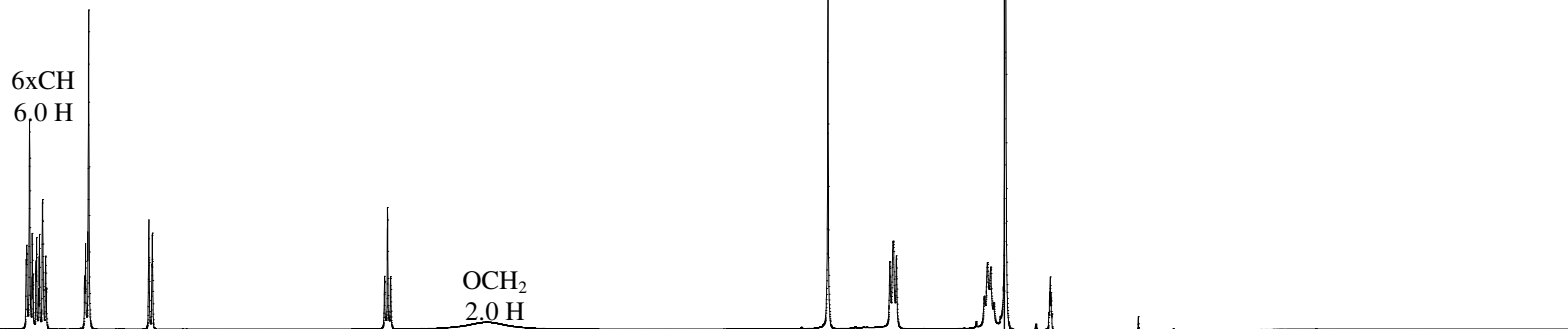
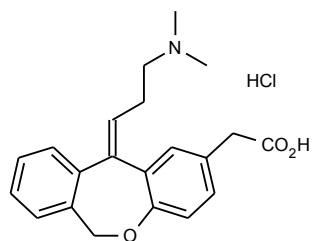
The available quantity of custom-synthesised material is always small, and this limits the extent and type of analytical data which can be obtained. This Certificate is presented in descriptive format for use by analytical chemists who are trained in the use of custom-synthesised materials. Custom materials often contain higher levels of residual solvents and/or water, and we urge you to use the corrected purity where needed rather than the raw HPLC purity. This compound is intended for use as an analytical reference material and it is not for human administration. Structures are shown with relative stereochemistry unless otherwise specified.

The re-test date is assigned from experience gained with the material in the laboratory and/or on storage. It is not possible to perform formal storage studies because of the small amount of material available.

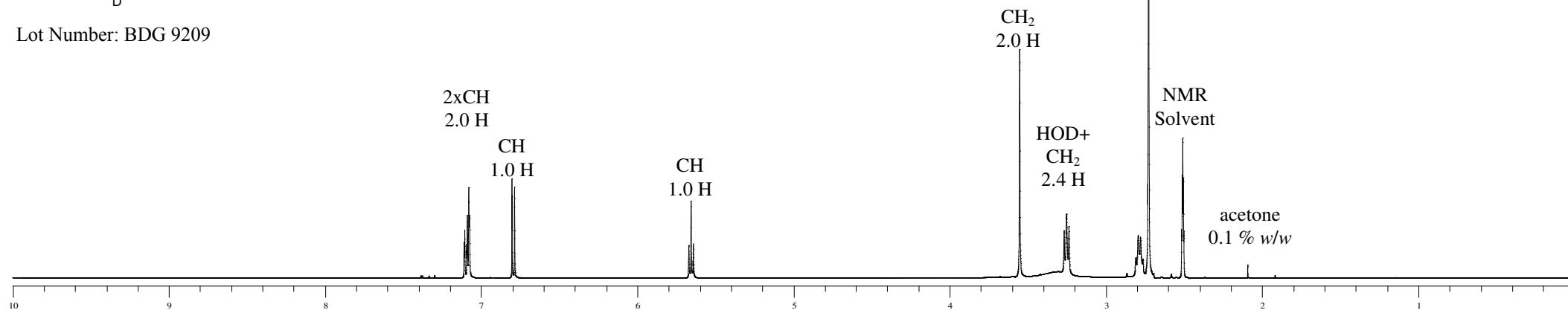


Proton NMR Spectrum of Olopatadine HCl (top) and Olopatadine-d₆ HCl (bottom) in DMSO-d₆

BDG SYNTHESIS



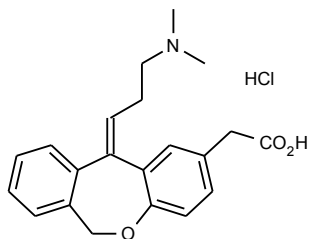
Lot Number: BDG 9209





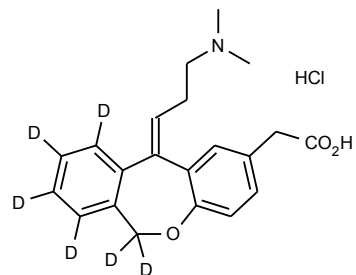
Carbon-13 NMR Spectrum of Olopatadine HCl (top) and Olopatadine-d₆ HCl (bottom) in DMSO-d₆

BDG SYNTHESIS



7xCH+C

OCH₂



Lot Number: BDG 9209

3xCH+
4xCD+C

CH

NCH₂

N(CH₃)₂

NMR Solvent
+CH₂

CH₂

C=O

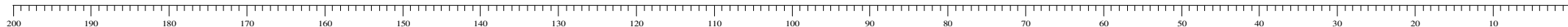
C-O

2xC

C

C

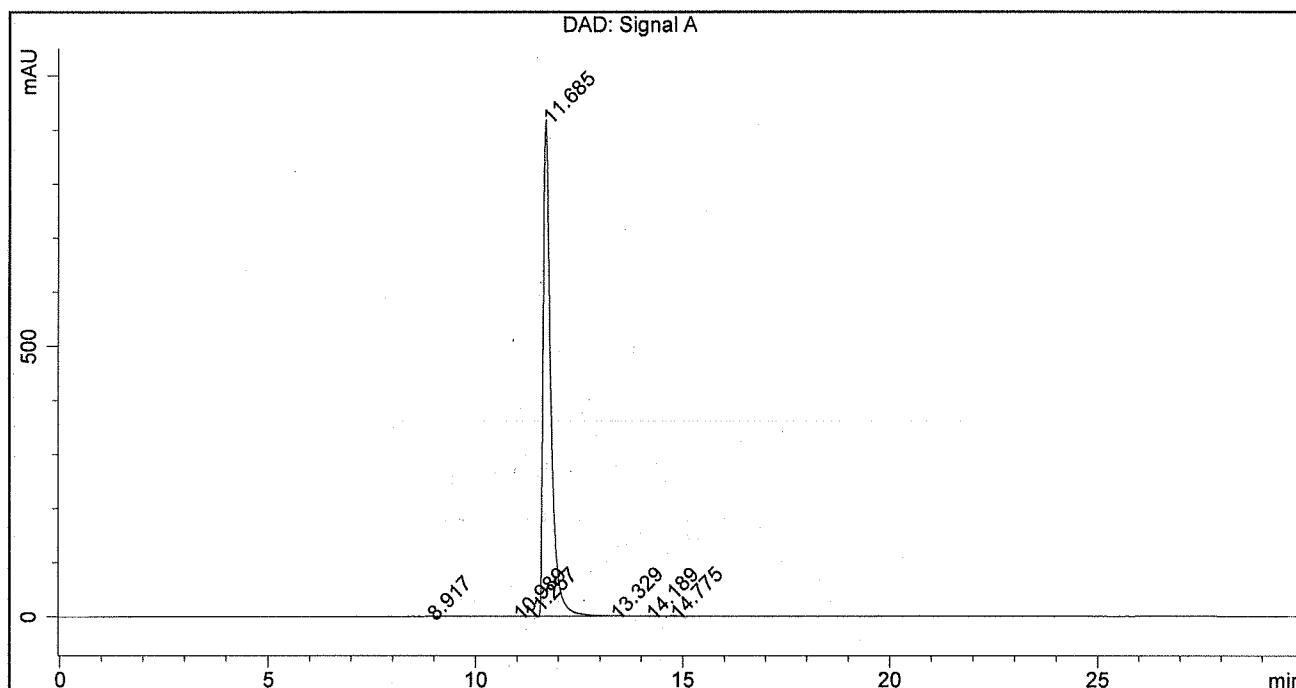
OCD₂



BDG - Analysis of olopatadine derivatives

Column : Phenomenex Luna C18(2) 5um 250 x 4.6 mm
 Guard : Phenomenex Security Guard C18 RP 4 x 3 mm
 Mobile Phase A : 10 mM diPotassium Hydrogen Phosphate pH = 7.0
 Mobile Phase B : Acetonitrile
 Gradient (A:B) : T0=80:20, T20=50:50, T24=50:50, T27=80:20, T30=80:20
 Flow Rate : 1.0 mL/min Sample Solvent : 2:1 Water : Acetonitrile
 Column Temperature : 20C Injection Volume : 10 uL Detection : UV at 254 nm

Sample Name	BDG 9209	Instrument	AnalyticalLC01
Acquisition	13/02/2014, 13:32:47	Method (rev.)	LC10016I (5)
Sequence	BDG_13Feb2014a	Vial Position	2
Operator	solvation010\cerityadmin	Injection	1 of 1



Area Percent Report

Peak#	RT	Peak Height	Peak Area	Width	Area %
1	8.92 min	0.3177	4.3256	0.1828 min	0.037 %
2	10.99 min	0.3428	3.3314	0.1594 min	0.028 %
3	11.26 min	0.4899	4.7752	0.1557 min	0.041 %
4	11.69 min	916.1073	11682.1557	0.1888 min	99.728 %
5	13.33 min	0.4866	6.0348	0.1627 min	0.052 %
6	14.19 min	0.3798	2.8735	0.1123 min	0.025 %
7	14.77 min	0.8063	10.5326	0.2024 min	0.090 %