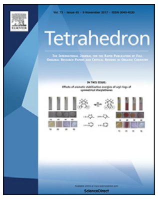
Tetrahedron 73 (2017) 6410–6420

Contents lists available at ScienceDirect

Tetrahedron

journal homepage: www.elsevier.com/locate/tet



Strategies for the synthesis of **HBGl$_3$**, a glutamic acid derived ligand bearing phenolic and azacarboxylate donor groups at the nitrogen atom

Raminder S. Mulla [a, **], Melissa T. Walden [a], Dmitry S. Yufit [a], Tanya Desa [b], Elena Lurie-Luke [b], J.A. Gareth Williams [a, *]

[a] *Department of Chemistry, Durham University, Durham, DH1 3LE, UK*
[b] *Procter and Gamble Technical Centres Limited, Rusham Park, Whitehall Lane, Egham, Surrey, TW2 9NW, UK*

ARTICLE INFO

Article history:
Received 6 June 2017
Received in revised form
5 September 2017
Accepted 15 September 2017
Available online 19 September 2017

Keywords:
Polydentate ligand
Polyaminocarboxylate ligand
Phenolate
Reductive amination
Lactam
Crystal structure

ABSTRACT

The development of a route applicable to the preparation of acyclic glutamic acid-based chelating ligands bearing two different auxiliary donor groups linked to the nitrogen atom by methylene spacers is described and applied to the synthesis of the new polydentate ligand **HBGl$_3$**, the first example of such a structure. The synthesis is accomplished using a strategy employing reductive amination and *t*-butyl ester protected intermediates. The most basic pK$_a$ values for the **HBGl$_3$** ligand have been estimated via potentiometric and UV–Visible titration techniques.

© 2017 Elsevier Ltd. All rights reserved.

1. Introduction

The naturally occurring glutamic acid motif (Fig. 1) has often been used as a coordinating unit, binding to metal ions via the amine and α-carboxylate to form a 5-membered chelate. Such a coordination mode leaves the ε-carboxylate non-coordinating and thus available, for example, to suppress the interaction of complexes of these ligands with anionic species[1,2] or to serve as a point of conjugation to other molecules through amide or ester formation.[3]

Another incentive for its incorporation into a ligand is that the presence of naturally occurring fragments in organic molecules can improve their biodegradability.[4] One such example is the well-known ligand **GLDA** {*N*,*N*-bis(carboxymethyl)glutamic acid or L-glutamic acid diacetic acid}, which has been shown to be readily biodegradable and is produced on a commercial scale (Fig. 2A).[5]

However, to the best of our knowledge, there has been no previous work on the preparation of acyclic glutamic acid-based chelating agents related to **GLDA** in which the donor groups pendent from the tertiary amine group are *(i)* different from one another, and *(ii)* linked to the amine group via methylene spacers. Owing to our interest in the development of biodegradable ligands that target both main group and first-row transition metal ions, we sought to prepare such a chelating agent that incorporates both aminocarboxylate *and* phenolate donors. We therefore chose the phenolic derivative **HBGl$_3$** as a synthetic target in the pursuit of a strategy towards the preparation of differentially N-substituted glutamic acid-based ligands; **HBGl$_3$** = (S)-2-{(carboxymethyl)[(*o*-hydroxyphenyl)methyl]amino}glutaric acid. In this compound, introduction of the phenol may be expected to increase the selectivity of the system for Fe^{3+} compared to **GLDA**, as well as adding a useful UV/Vis spectroscopic handle.

2. Results and discussion

2.1. Synthetic strategies

From the disconnections that would lead us back to L-glutamic

---

* Corresponding author.
** Corresponding author.
E-mail addresses: raminder.mulla@durham.ac.uk (R.S. Mulla), j.a.g.williams@durham.ac.uk (J.A.G. Williams).

https://doi.org/10.1016/j.tet.2017.09.032
0040-4020/© 2017 Elsevier Ltd. All rights reserved.

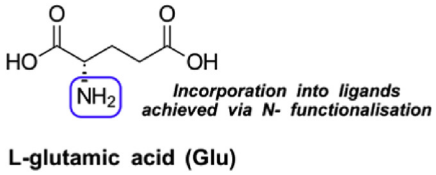

NH2
Incorporation into ligands achieved via N- functionalisation
L-glutamic acid (Glu)

Fig. 1. The structure of L-glutamic acid.

acid as a starting material (Fig. 2B), we first investigated the introduction of the amine as a separate fragment along disconnection **A**, which could be reacted with an α-haloglutarate alkylating agent. In this strategy, the use of separate amine fragments was considered advantageous since the fragments could also be used in other syntheses.

As such, the racemic α-haloglutarate alkylating compound **1a** was easily accessed from glutamic acid via diazotisation,[6] and could be subject to Finkelstein conditions to afford the more reactive iodo-compound **1b** (Scheme 1). So in order to evaluate the feasibility of a route based on disconnection **A**, **1a** was subjected to a test reaction with dibenzylamine which gave **2** in modest yields. Buoyed by this result, we prepared amine **3a** via reductive amination, for subsequent reaction with **1a**, but observed no consumption of starting materials.

Using the corresponding benzyl ether of **3a**, namely **3b**, we were able to obtain key intermediate **4** after a prolonged reaction period with the reactive electrophile **1b** (6 days). At this point, the low yields and racemisation along the route from the preparation of **1a**, together with the need for multiple deprotection steps, prompted us to abandon an approach based on disconnection **A**.

We then looked to reductive amination chemistry to introduce the salicylaldehyde unit onto glutamic acid (disconnection **B**) via intermediate **5** (Scheme 2),[7] since a route along this disconnection would involve fewer steps and retain chirality. Although **5** was easily accessible due to it being isolable through a simple precipitation-filtration purification procedure, N-alkylations of **5** attempted with either chloroacetic acid[8] or *t*-butyl bromoacetate under basic conditions (NaOH, $KHCO_3$ or Et_3N) in water, acetonitrile or DMF were all unsuccessful, giving recovered starting material only.

We therefore opted to protect the carboxyl groups of **5**, using a

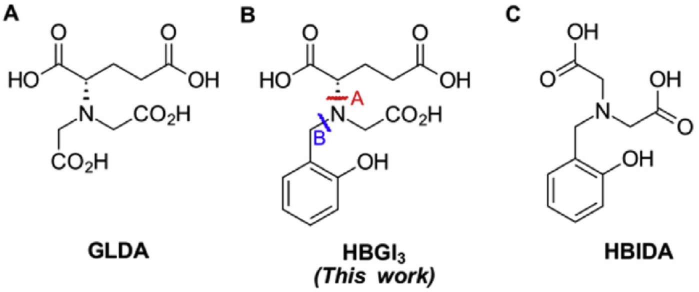
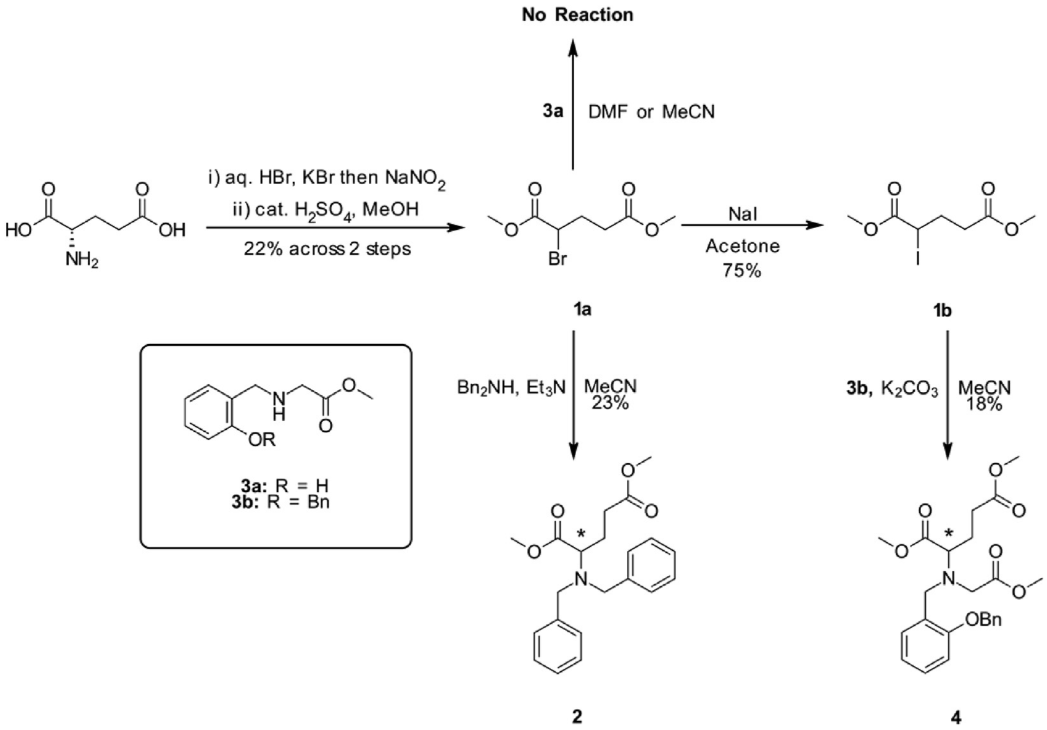
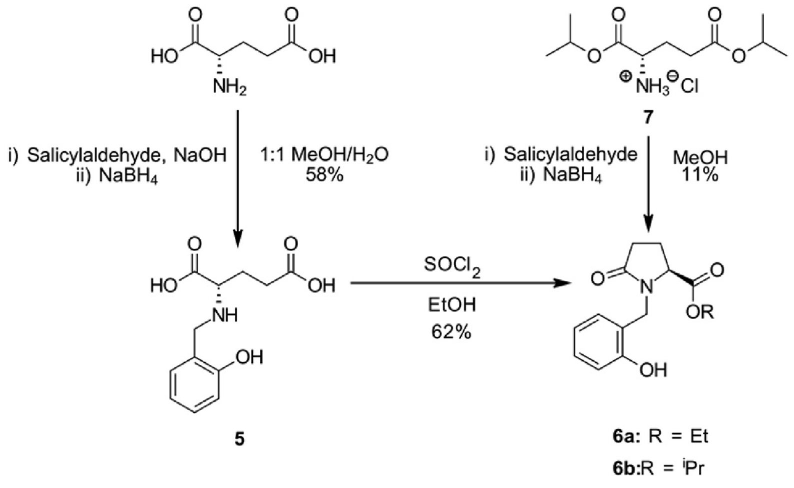

A
B
C
GLDA
HBGI3
(This work)
HBIDA

Fig. 2. Structures of the **GLDA**, **HBGI$_3$** and **HBIDA** ligands, with some potential disconnection points towards **HBGI$_3$** shown. (**HBIDA** = hydroxybenzyliminodiacetic acid).


No Reaction
3a DMF or MeCN
i) aq. HBr, KBr then NaNO2
ii) cat. H2SO4, MeOH
22% across 2 steps
NaI
Acetone
75%
1a
1b
Bn2NH, Et3N MeCN 23%
3b, K2CO3 MeCN 18%
3a: R = H
3b: R = Bn
2
4

Scheme 1. Intermediates used towards the preparation of **HBGI$_3$** via the use of an α-haloglutarate electrophile.



Scheme 2. Reactions of glutamic acid derivatives leading to cyclisation.

Fischer-type esterification, since many reported ligand syntheses involving N-alkylation utilise protected carboxyl intermediates to prevent the possible detrimental influence of the carboxylate groups in subsequent reactions. In the first instance then, we chose to convert the carboxyl groups to their ethyl esters using thionyl chloride, in the hope that competing lactam formation would be suppressed by the protonation of the amine group in **5** under these conditions. This proved not to be the case. Indeed, lactam **6a** was the sole product isolated. It may be noted that the cyclisation proceeded with the preservation of the *S*-stereocentre of **5**, as verified by single-crystal X-ray crystallography (Fig. 3).

We then looked to prepare **7**, the di-isopropyl ester of L-glutamic acid as a reductive amination substrate, in the hope that the bulkier isopropyl esters would be base-labile but also large enough to inhibit lactam formation. Unfortunately, cyclisation was also observed using this approach, giving lactam **6b** as the main product.

Finally, we turned our attention to intermediates bearing the bulky *t*-butyl ester protecting group, which has shown the ability to prevent the attack of some strong nucleophiles on a given carboxyl group,[9] in the hope of preventing the cyclisation seen earlier (Scheme 2). This change to *t*-butyl esters allowed **HBGI$_3$** to be obtained according to Scheme 3.

Di-*t*-butyl-glutamate, **8**, is commercially available, and could be converted into protected intermediate **10** using reductive amination in either a one-pot process using NaBH(OAc)$_3$,[10] or a two-pot procedure via the isolated imine **9**. In our hands, the use of the two-pot procedure, using NaBH$_4$ as the reducing agent, offered much better reproducibility.

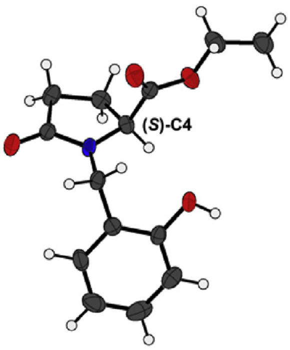


Fig. 3. Molecular structure in the crystals of **6a**. The stereocentre at C4 is shown for clarity. Thermal ellipsoids are shown at the 50% probability level.

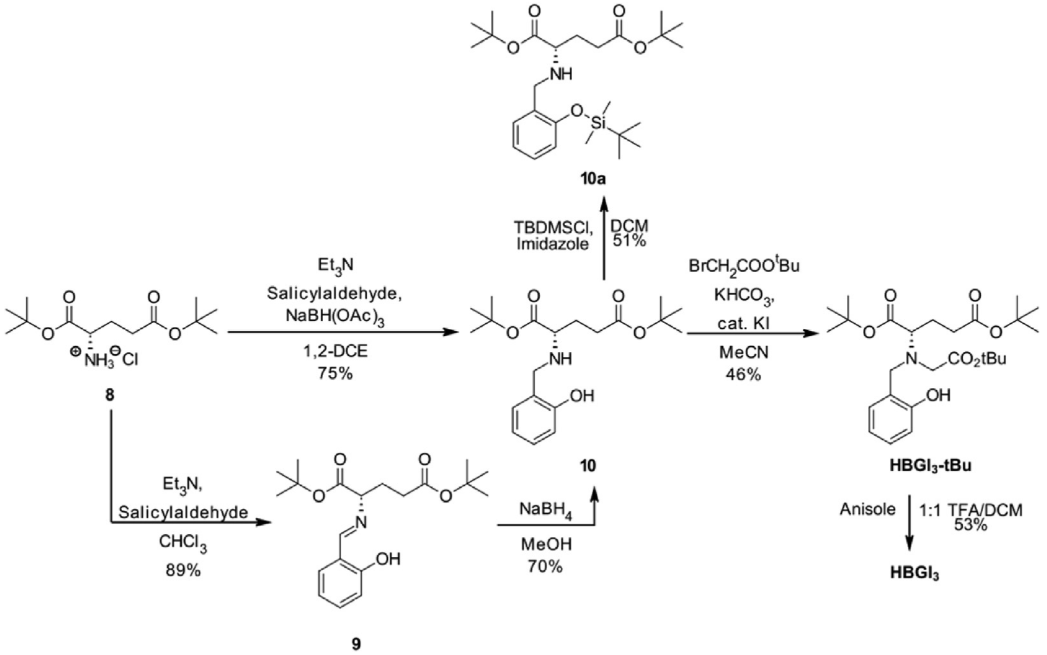
Alkylation of amine **10** with *t*-butyl bromoacetate to form **HBGI$_3$-tBu** was slow (>1 week for the disappearance of starting material by TLC), but afforded the compound in reasonable yields. Unfortunately, attempts to shorten the reaction time (*e.g.* using microwave irradiation and high boiling solvents like DMF) failed, often leading to complex mixtures even after repeated chromatographic purification.

HBGI$_3$-tBu was then treated with a mixture of TFA/DCM in the presence of anisole as a *t*-butyl cation scavenger to give **HBGI$_3$** in 15–18% cumulative yield, which is fairly typical for the synthesis of polyaminocarboxylate ligands.[11,12]

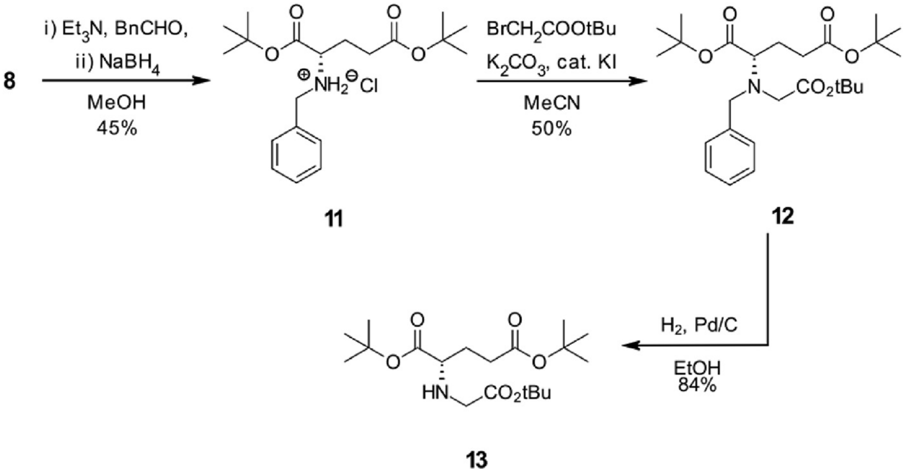
The low reaction rate for the alkylation of **10** to **HBGI$_3$-tBu** might be attributable either to steric hindrance of the amine group in **10**, or to strong intramolecular hydrogen bonding, and to determine the root cause we sought to investigate both of these possibilities. First, we investigated the role of sterics via the synthesis of the less hindered, benzylated intermediate **11**. The alkylation of **11** to form **12** proceeded readily within a 24-h period (Scheme 4), a much faster alkylation than that to form **HBGI$_3$-tBu**.

We also speculated as to whether an intramolecular hydrogen bond in **10** could be a contributing factor accounting for the slow rate of alkylation. The question was posed on the basis of the single crystal X-ray structure of **9**, which shows the presence of a strong intramolecular O–H···N bond, d(H···N) = 1.723(3) Å, see Fig. 4. We reasoned that the structure of **9** could help us predict the hydrogen bonding behaviour of **10** because the hydroxybenzyl group is unchanged in going from 9 to 10 and intramolecular hydrogen bonding in other hydroxybenzylamines is known.[13,14] As a control, we therefore prepared a TBDMS-ether of **10**, that is **10a** (Scheme 3), which would be unable to form an intramolecular hydrogen bond like that seen in **9**. Preliminary alkylation studies on **10a** were then performed and showed very slow conversion (**10a** was not fully consumed even after 7 days), indicating that the reason behind the slow alkylation of **10** is likely to be the presence of the *ortho*-substituted aromatic group hindering attack of an incoming electrophile as opposed to an intramolecular hydrogen bond.

To assess whether introduction of the salicylaldehyde moiety further along the route to **HBGI$_3$-tBu** would give improved yields and reaction times, **12** was debenzylated via catalytic hydrogenation to form tri-ester **13** (also accessible via **8** – see Experimental Section), which was in turn subjected to reductive amination with


10a
TBDMSCl, Imidazole
DCM 51%
Et3N
Salicylaldehyde,
NaBH(OAc)3
1,2-DCE
75%
8
10
BrCH2COOtBu
KHCO3,
cat. KI
MeCN
46%
HBGI3-tBu
Anisole
1:1 TFA/DCM
53%
HBGI3
Et3N,
Salicylaldehyde
CHCl3
89%
9
NaBH4
MeOH
70%

Scheme 3. Successful preparation of **HBGI₃** via *t*-butyl ester intermediates.


8
i) Et3N, BnCHO,
ii) NaBH4
MeOH
45%
11
BrCH2COOtBu
K2CO3, cat. KI
MeCN
50%
12
H2, Pd/C
EtOH
84%
13

Scheme 4. Synthesis of the alkylation test substrate **11** and alternative **HBGI₃** precursor **13**.

salicylaldehyde in attempts to form **HBGI₃-tBu**. However these attempts were unsuccessful and only starting materials were recovered.

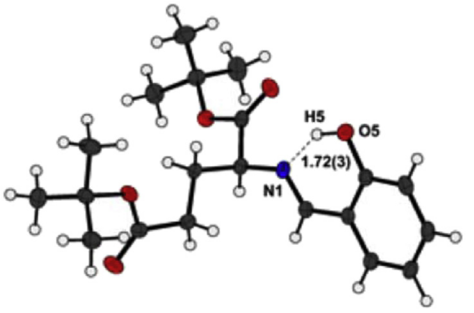

H5
O5
1.72(3)
N1

Fig. 4. Molecular structure in the crystal of **9**. The N-H distance is shown in Å. Thermal ellipsoids are shown at the 50% probability level.

2.2. *Spectroscopic properties and p*K_a

With **HBGI₃** in hand, we were able to estimate the pK_a values for the amine group and the phenol as being around 8 and 13 respectively. The value for the amine ($R_3NH^+ \rightleftharpoons R_3N$) equilibrium was determined from the pH titration curve and could also be followed by UV–Vis spectroscopy.

The largest bathochromic shift (15 nm) that occurs in the UV–Vis spectrum of **HBGI₃** as a function of increasing pH was used to determine the pK_a of the phenolic group by taking the ratio of absorbances at 277 and 291 nm. This bathochromic shift is assignable to the presence of smaller energy gaps between the molecular orbitals involved in the transitions of interest, evidenced by TD-DFT studies at the B3LYP/6-31G++ level of theory. Indeed, seen in Fig. 5C, there is a lower energy feature for the calculated spectrum of the phenoxide form of **HBGI₃** that is not present in the calculated spectrum of the phenolic form. For both the phenolic and phenoxide forms of **HBGI₃**, the molecular orbitals with the

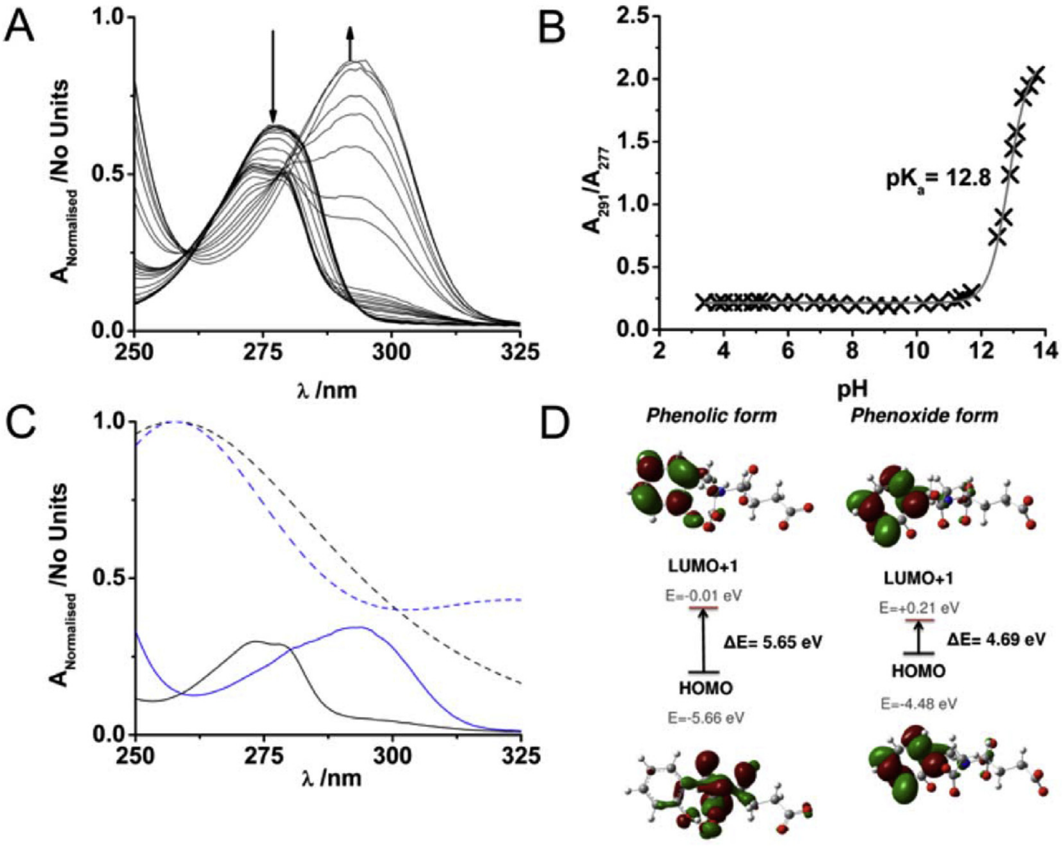


Fig. 5. **(A)** UV–Visible spectra for **HBGl₃** as a function of pH (T = 25 °C, I = 0.1 M KCl). **(B)** Plot of the ratio of absorbance values at 291 and 277 nm (A_{291}/A_{277}) as a function of pH. **(C)** Comparison of the experimental spectra (solid lines) of the phenolic (predominant at pH 10–12, black) and phenoxide (predominant at pH > 13, blue) forms of **HBGl₃** with those calculated (dashed lines) from TD-DFT calculations. **(D)** Main orbitals involved in transitions, HOMO and LUMO+1 for the **HBGl₃** ligand in the phenolic and phenoxide forms.

greatest contribution to the lowest energy bands (λ_{max} 277 and 291 nm for the phenolic and phenoxide forms respectively) are the HOMO and LUMO+1.

It is also noteworthy that the localisation of the HOMOs in the phenolic and phenoxide forms of **HBGl₃** are different: the phenolic form can be considered to have predominantly *n* character whereas the phenoxide HOMO possesses *π* character. It is clear that destabilisation of the HOMO, more so than the stabilisation of the LUMO+1, is responsible for the reduced energy gap between these two orbitals in the phenoxide form of **HBGl₃** (Fig. 5D).

The amine dissociation event is also observed when spectra obtained between pH 5 and 11 are considered in isolation and an absorbance ratio plotted from the pH titration data in Fig. 5A. A sigmoid curve results with a midpoint at pH = 8.2, which is in reasonable agreement with the value obtained from the potentiometric data (Fig. 6B).

At a first glance the value for the amine pK_a might appear rather low compared to other amine-bearing ligands like **IDA** or **NTA** (iminodiacetic acid and nitrilotriacetic acid respectively). Martell and others have reported similarly low pK_a values for the amino group of **HBIDA** (hydroxybenzyl-iminodiacetic acid) and related ligands,[15–17] and these values have usually been assigned to the formation of an intramolecular hydrogen bond between the free amine and the un-ionised phenol. This may also be the reason behind the relatively high phenol pK_a observed for **HBGl₃** compared to phenol itself, in that ionisation of the phenolic group

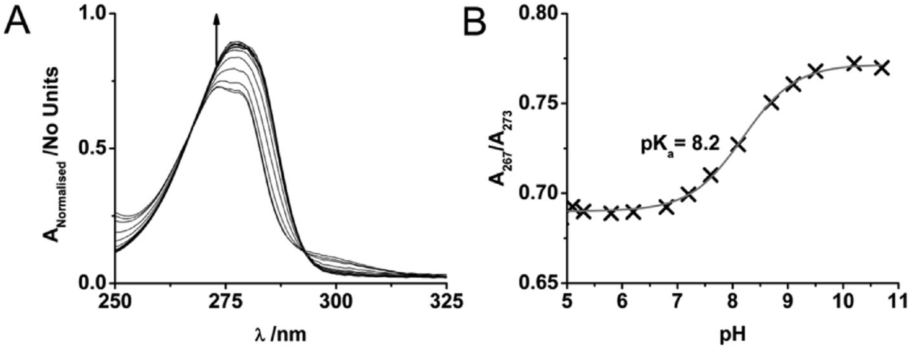


Fig. 6. **(A)** UV–Visible spectra (between pH 5 and 11) of **HBGl₃** (T = 25 °C, I = 0.1 M KCl) used to confirm the amine pK_a value obtained by pH-metric titration. **(B)** Plot of the ratio of absorbance values at 267 and 273 nm (A_{267}/A_{273}) as a function of pH.

to phenoxide in **HBGl$_3$** is penalised by loss of this intramolecular hydrogen bond.

3. Conclusions

A route has been devised to prepare the first acyclic glutamic acid based ligand that bears different chelating groups attached to the nitrogen atom via a methylene bridge, **HBGl$_3$**. The ligand is prepared using stepwise functionalisation at the nitrogen of a glutamic acid equivalent, as opposed to pre-forming an amine fragment for alkylation onto an α-haloglutarate type intermediate. *t*-Butyl ester protection is employed to prevent unwanted cyclisation reactions that occur when smaller ethyl or isopropyl protecting groups are used to mask the glutamic carboxyl groups on the synthetic intermediates. It is envisaged that the adoption of this route will facilitate the straightforward synthesis of other glutamic acid based chelating ligands.

4. Experimental

4.1. Materials

Reagents were obtained from commercial sources and used without further purification unless otherwise stated. Solvent extractions were performed in a 250 mL separating funnel with ca. 50 mL for each phase, unless otherwise stated. For procedures involving dry solvent, glassware was oven-dried for at least 8 h prior to use. Dedicated oxygen-free nitrogen or argon cylinders (BOC, UK) were used to provide an inert atmosphere.

4.2. Instrumentation

NMR: Routine ^{1}H (400 MHz) and ^{13}C NMR (101 MHz) spectra were acquired on Bruker Avance 400, or Varian Mercury 400 NMR spectrometers. Two-dimensional NMR (COSY, HSQC and HMBC), and certain ^{1}H/^{13}C NMR spectra, were acquired by the solution state NMR service at Durham University on Varian VNMRS-600 (600 MHz) or VNMRS-700 (700 MHz) instruments. Where visible, signals corresponding to CDCl$_3$ were referenced to $\delta = 7.26$ ppm in ^{1}H NMR spectra and $\delta = 77.2$ ppm for ^{13}C spectra.

4.3. Mass spectrometry

ES-MS data (positive and negative ionisation modes) were obtained on a Waters TQD mass spectrometer interfaced with an Acquity UPLC system. GC-MS (EI ionisation) was performed on an Agilent instrument equipped with a 5973 model quadrupole mass spectrometer.

4.4. FT-IR

All infra-red spectra were recorded on a Perkin-Elmer Spectrum 90 spectrometer equipped with an ATR stage. Substances for analysis were used neat unless otherwise indicated.

4.5. Chromatography

TLC was performed on silica using Merck foil-backed TLC plates. Column chromatography on silica was undertaken in one of two ways. Method i) involved the use of a standard glass column and silica sourced from Fluorochem Limited and elution was performed according to the method of Clark Still.[18] Method ii) involved the use of a Teledyne ISCO CombiFlash instrument equipped with RediSep R$_f$ silica cartridges to perform automated gradient elution. Analytical and preparative HPLC was performed by the chromatography service at Durham University.

4.6. Elemental analysis

Elemental analyses were performed using an Exeter CE-440 Elemental Analyser device.

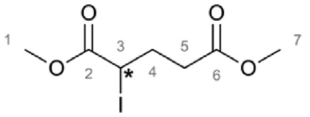
4.7. X-ray crystallography

The X-ray single crystal data for compounds **6a** and **9** have been collected on a Bruker D8Venture (λCuKα radiation, $\lambda = 1.54178$ Å, Photon100 CMOS detector, IμS-microsource, focusing mirrors, ω- and φ-scan, 1.0°/frame) and Bruker SMART CCD 6000 (λMoKα radiation, $\lambda = 0.71073$ Å, fine-focus sealed tube, graphite monochromator, MonoCap optics, ω-scan, 0.3°/frame) diffractometers equipped with a Cryostream (Oxford Cryosystems) open-flow nitrogen cryostats at temperatures 120.0(2)K. All structures were solved by direct method and refined by full-matrix least squares on F^2 for all data using Olex2[19] and SHELXTL[20] software. All non-hydrogen atoms were refined anisotropically, hydrogen atoms in structures **3a** and **9** were refined isotropically, the hydrogen atoms in structure **6a** were placed in the calculated positions and refined in riding mode.

The compounds **1a**, **5** and **7** were prepared using previously reported conditions, as summarised in the Supporting Information.

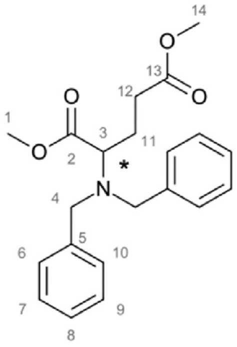
4.8. Synthesis and characterisation data for individual compounds

*4.8.1. Dimethyl 2-iodoglutarate, **1b***



Sodium iodide (2.22 g, 13.4 mmol) was partially dissolved in acetone (12 mL) and ester **4a** (0.66 g, 2.76 mmol) was added at once forming a yellow-orange solution that was refluxed for 2 h. Volatiles were then removed *in vacuo* and the resulting brown solid was triturated in diethyl ether and then filtered. Concentration of the filtrate *in vacuo* afforded the product as a brown oil (0.59 g, 2.06 mmol, 75%). δ_H (400 MHz, CDCl$_3$) 4.46 (1H, dd, *J* 8.5, 6.0 Hz, H^3), 3.75 (3H, s, H^1), 3.68 (3H, s, H^7), 2.47–2.43 (2H, m, H^5), 2.29–2.25 (2H, m, H^4); δ_C (101 MHz, CDCl$_3$) 172.1 (C^6), 171.3 (C^2), 52.8 (C^1), 51.7 (C^7), 33.2 (C^3), 30.8 (C^5), 18.9 (C^4); *m/z* (GC-EI$^+$) 286.0 (M$^{\bullet+}$, M = C$_7$H$_{11}$IO$_4$), 256.0 (M−2Me$^{\bullet+}$).

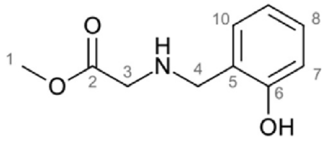
*4.8.2. Dimethyl 2-[bis(benzyl)amino]glutarate, **2***



A solution of haloester **1a** (400 mg, 1.69 mmol) in acetonitrile

(4 mL, dried over molecular sieves) was prepared and allowed to stir at room temperature under nitrogen in a Schlenk tube prior to the addition of triethylamine (0.5 mL, 3.61 mmol) at once followed by the dropwise addition of dibenzylamine over 15 min (0.39 mL, 2.03 mmol). The brown solution was heated at 60 °C for 40 h under nitrogen leading to the formation of a precipitate that was filtered off after cooling to room temperature. The filtrate was concentrated *in vacuo* and partitioned between diethyl ether and water; the organic layer was washed with brine, dried over magnesium sulphate and concentrated *in vacuo* prior to purification of the yellow residue via column chromatography (method ii, dichloromethane:methanol 100:0 → 90:10) to afford the product as a yellow oil (140 mg, 0.394 mmol, 23%). δ_H (600 MHz, $CDCl_3$) 7.37–7.27 (8H, m, H^6, H^7, H^9, H^{10}), 7.21–7.13 (2H, m, H^8), 3.89 (2H, d, *J* 14.0 Hz, H^4), 3.77 (3H, s, H^1), 3.54 (3H, s, H^{14}), 3.52 (2H, d, *J* 14.0 Hz, H^4), 3.38–3.33 (1H, m, H^3), 2.47–2.41 (1H, m, H^{11}), 2.34–2.27 (1H, m, H^{11}), 2.05–2.00 (2H, m, H^{12}); δ_C (151 MHz, $CDCl_3$) 173.5 (C^{13}), 172.9 (C^2), 139.4 (C^5), 129.1, 128.4, 127.2 (C^6, C^7, C^8, C^9), 59.8 (C^3), 54.6 (C^{14}), 51.6 (C^4), 51.3 (C^1), 30.5 (C^{11}), 24.5 (C^{12}); *m/z* (ES-MS$^+$) 356.2 (100%, $[M+H]^+$); HRMS (ES-MS$^+$) MH^+ found 356.1854, $[C_{21}H_{26}NO_4]^+$ requires 356.1862.

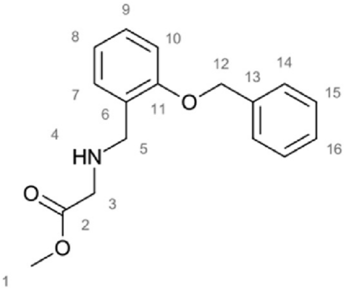
4.8.3. Methyl {[(o-hydroxyphenyl)methyl]amino}acetate, **3a**



Glycine methyl ester hydrochloride (1.01 g, 8.01 mmol) was added to a solution of triethylamine (1.2 mL, 8.60 mmol) in 1,2-dichloroethane (30 mL) followed by the addition of salicylaldehyde (0.87 mL, 8.89 mmol). The reaction mixture was subsequently stirred under nitrogen for 10 min prior to the addition of sodium triacetoxyborohydride (2.10 g, 9.91 mmol) and monitored via TLC (90:10 dichloromethane:methanol) and ES-MS techniques until the absence of starting material and derivative imines were observed (<4 h). The mixture was then quenched with aqueous, saturated potassium bicarbonate (30 mL) and the biphasic mixture was stirred for a few minutes prior to extraction of the organic layer with dichloromethane (2 × 30 mL). The combined extracts were dried with magnesium sulphate and concentrated *in vacuo* to a yellow solid which was purified by column chromatography (method ii, dichloromethane:methanol, compound eluted in 90:10 v/v dichloromethane:methanol) to give the title compound as a crystalline sold (0.36 g, 1.84 mmol, 23%). Slow evaporation of a sample from acetonitrile gave crystals suitable for analysis by X-ray crystallography. δ_H (400 MHz, $CDCl_3$) 7.22–6.75 (4H, m, H^7, H^8, H^9, H^{10}), 4.00 (2H, s, H^4), 3.76 (3H, s, H^1), 3.45 (2H, s, H^3); δ_C (101 MHz, $CDCl_3$) 172 (C^2), 158 (C^5), 130, 129, 122, 120, 117 (C^6, C^7, C^8, C^9, C^{10}), 53 (C^4), 52.5 (C^1), 49 (C^3); *m/z* (ES-MS$^+$) 196 (100%, $[M+H]^+$).

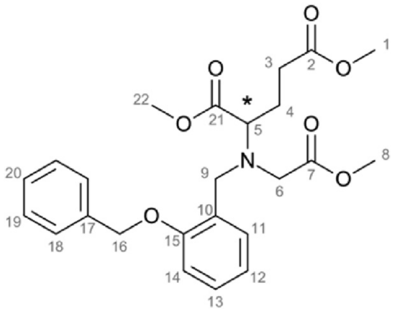
*Crystal data for **3a***: $C_{10}H_{13}NO_3$, M = 195.21, orthorhombic, space group P bca, a = 9.2513(8), b = 9.0842(8), c = 22.8517(19) Å, U = 1920.5(3) Å^3, F(000) = 832, Z = 8, D_c = 1.350 mg m^{-3}, μ = 0.10 mm^{-1}. 18663 reflections were collected (2Θ 3.56–60°) yielding 2799 unique data (R_{merg} = 0.0546). Final $wR_2(F^2)$ = 0.1146 for all data (179 refined parameters), conventional R_1 (F) = 0.0404 for 1966 reflections with $I \geq 2\sigma$, GOF = 0.987. CCDC-1551895.

4.8.4. Methyl ({[o-(benzyloxy)phenyl]methyl}amino)acetate, **3b**



o-(Benzyloxy)benzaldehyde (510 mg, 2.41 mmol) was dissolved in 1,2-dichloroethane (10 mL) and methyl glycinate hydrochloride (330 mg, 2.63 mmol) was suspended in the mixture prior to the addition of triethylamine (0.5 mL, 3.6 mmol) and sodium triacetoxyborohydride (1.01 g, 4.77 mmol). The reaction mixture was then stirred at room temperature under nitrogen for 17 h prior to the addition of a solution of saturated sodium carbonate. Dichloromethane was then added to the biphasic mixture and the organic layer extracted, dried over magnesium sulphate and the solvent removed *in vacuo* to afford the title compound (0.50 g, 1.75 mmol, 74%) as a colourless oil. δ_H (700 MHz, $CDCl_3$) 7.47–7.42 (2H, m, H^{14}), 7.43–7.34 (2H, m, H^{15}), 7.36–7.28 (1H, m, H^{16}), 7.28 (1H, dd, *J* 7.5, 2.0 Hz, H^7), 7.22 (1H, dd, *J* 8.0, 2.0 Hz, H^9), 6.98–6.88 (2H, m, H^8, H^{10}), 5.08 (2H, s, H^{12}), 3.89 (2H, s, H^5), 3.63 (3H, s, H^1), 3.40 (2H, s, H^3), 2.64 (1H, s, H^4); δ_C (176 MHz, $CDCl_3$) 172.7 (C^2), 156.8 (C^{11}), 137.1 (C^{13}), 130.0 (C^7), 128.6 (C^{15}), 128.5 (C^9), 127.9 (C^{16}), 127.9 (C^6), 127.2 (C^{14}), 120.8 (C^8), 111.8 (C^{10}), 69.9 (C^{12}), 51.6 (C^1), 50.0 (C^3), 48.5 (C^5).

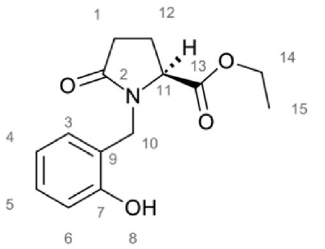
4.8.5. Dimethyl-2-({[o-(benzyloxy)phenyl]methyl}(methoxycarbonylmethyl)amino) glutarate, **4**



Alkyl iodide **4b** (181 mg, 0.625 mmol) and amine **6a** (170 mg, 0.596 mmol) were dissolved in acetonitrile (3 mL, dried over molecular sieves) and potassium carbonate (0.17 g, 1.23 mmol) added. The reaction mixture was then heated at 60 °C under nitrogen for 6 days. Once cooled to room temperature, solids were filtered off and the solvent removed from the filtrate *in vacuo*. The residue was then purified by column chromatography (method ii, ethyl acetate:-hexanes 0:100 → 100:0 compound eluted in ca. 30:70 v/v ethyl acetate:hexanes) to afford the title compound (47.0 mg, 0.108 mmol, 18%) as a colourless oil. ν_{max} (ATR) 1730 (ester C=O st), 1200 (ester C–O st); δ_H (600 MHz, $CDCl_3$) 7.46 (1H, dd, *J* 7.5, 2.0 Hz H^{11}), 7.42 (2H, dd, *J* 7.5, 1.5 Hz, H^{19}), 7.38 (2H, dd, *J* 7.5, 1.0 Hz, H^{18}), 7.33–7.28 (1H, m, H^{20}), 7.23–7.15 (1H, m, H^{13}), 6.95 (1H, dd, *J*

7.5, 1.0 Hz, H^{12}), 6.89 (1H, dd, *J* 8.0, 1.0 Hz, H^{14}), 5.07 (2H, s, H^{16}), 4.04 (1H, d, *J* 14.0 Hz, H^{9}), 3.84 (1H, d, *J* 14.0 Hz, H^{9}), 3.62 (3H, s) and 3.61 (3H, s, H^{8}, H^{22}), 3.59 (3H, s, H^{1}), 3.56 (1H, s H^{6}), 3.51–3.44 (2H, m, H^{5}, H^{6}), 2.65–2.36 (2H, m, H^{3}), 2.15–1.86 (2H, m, H^{4}); δ_C (151 MHz, $CDCl_3$) 173.9 (C^{2}), 173.3, 172.3 (C^{7}, C^{21}), 157.1 (C^{15}), 137.7 (C^{17}), 130.8 (C^{11}), 128.6 (C^{18}), 128.4 (C^{13}), 127.9 (C^{20}), 127.3 (C^{19}), 127.2 (C^{10}), 120.9 (C^{12}), 111.9 (C^{14}), 70.1 (C^{16}), 62.1 (C^{5}), 51.9 (C^{6}), 51.5 (C^{8}, C^{22}), 51.5 (C^{1}), 51.3 (C^{8}, C^{22}), 50.0 (C^{9}), 30.3 (C^{3}), 25.0 (C^{4}); *m/z* (ES-MS$^+$) 444.5 (100%, [M+H]$^+$); HRMS (ES-MS$^+$) MH$^+$ found 444.2026, $[C_{24}H_{30}NO_7]^+$ requires 444.2022.

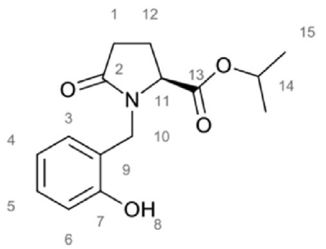
4.8.6. *Ethyl (2S)-1-[(o-hydroxyphenyl)methyl]-5-oxo-2-pyrrolidinecarboxylate, **6a***



Amino acid **5** (420 mg, 1.66 mmol) was suspended in ethanol (7 mL) and thionyl chloride (0.6 mL, 8.27 mmol) was added dropwise. The suspension was then refluxed for 48 h under argon, cooled and then filtered. The filtrate was then concentrated *in vacuo* to give a white solid that was triturated with ethanol using ultrasound (3 × 5 mL), and then diethyl ether (3 × 5 mL), the solvent being removed *in vacuo* between cycles. Dichloromethane was used to suspend the resulting solid which was filtered through Celite® and the solvent removed from the pink filtrate to afford the title compound (270 mg, 1.03 mmol, 62%) as a pink crystalline solid. Single crystals suitable for structural and stereochemical determination via X-ray crystallography were grown from this solid via slow evaporation from acetone. mp 111–112 °C; ν_{max} (ATR) 3069 (br, phenol O–H st), 2978 (sp^3 C–H st), 1739 (ester C=O st), 1660 (amide C=O st), 1203 (C–O st); δ_H (600 MHz, $CDCl_3$) 8.76 (1H, s, H^{8}), 7.22 (1H, dd, *J* 7.5, 1.5 Hz, H^{5}), 7.02 (1H, dd, *J* 7.5, 1.5 Hz, H^{3}), 6.94 (1H, d *J* 7.5 Hz, H^{6}), 6.79 (1H, dd, *J* 7.5, 1.5 Hz, H^{4}), 4.64 (1H, d, *J* 15.0 Hz, H^{10}), 4.26–4.28 (1H, m, H^{13}), 4.18–4.11 (m, 2H, H^{13}, H^{14}), 4.06 (1H, dq, *J* 11.0, 7.0 Hz, H^{14}), 2.65–2.52 (1H, m, H^{1}), 2.49–2.37 (1H, m, H^{1}), 2.35–2.25 (1H, m, H^{12}), 2.17–2.06 (1H, m, H^{12}), 1.22 (3H, t, *J* 7.0 Hz, H^{15}); δ_C (151 MHz, $CDCl_3$) 177.5 (C^{2}), 171.3 (C^{13}), 155.9 (C^{7}), 131.0 (C^{3}), 130.3 (C^{5}), 121.3 (C^{9}), 119.6 (C^{4}) 117.8 (C^{6}), 61.8 (C^{14}), 60.4 (C^{11}), 43.0 (C^{10}), 29.2 (C^{1}), 23.0 (C^{12}), 14.0 (C^{15}); *m/z* (ES-MS$^+$) 264.0 (100%, [M+H]$^+$); HRMS (ES-MS$^+$) MH$^+$ found 264.1243, $[C_{14}H_{18}O_4]^+$ requires 264.1236.

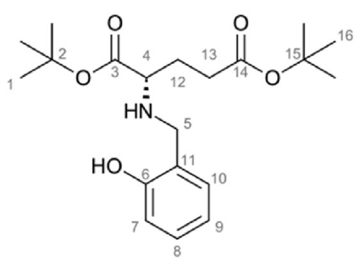
*Crystal data for **6a***: $C_{14}H_{17}NO_4$, M = 263.29, triclinic, space group P 1, a = 6.8353(3), b = 7.5669(3), c = 7.7552(3) Å, α = 96.5965(19), β = 112.7236(19); γ = 106.441(2)°, U = 343.27(2) Å^3, F(000) = 140, Z = 1, D_c = 1.274 mg m^{-3}, μ = 0.775 mm^{-1}. 2382 reflections were collected (2Θ 12.6–139.9°) yielding 1722 unique data (R_{merg} = 0.0554). Final wR_2(F^2) = 0.2218 for all data (174 refined parameters), conventional R_1 (F) = 0.0767 for 1666 reflections with I ≥ 2σ, GOF = 1.086, Hooft −0.1(3). CCDC-1551896.

4.8.7. *Isopropyl 1-[(o-hydroxyphenyl)methyl]-5-oxo-2-pyrrolidinecarboxylate, **6b***



Diisopropyl glutamate hydrochloride **7** (610 mg, 2.28 mmol) and salicylaldehyde (280 mg, 2.28 mmol) were dissolved in methanol (6 mL) and stirred under nitrogen at 40 °C for 1 h, and then at room temperature overnight. Then, sodium borohydride (410 g, 10.9 mmol) was then added portionwise and the mixture stirred at room temperature for 6 h. Solids were then filtered off and the filtrate concentrated *in vacuo* prior to re-dissolution in dichloromethane and the removal of precipitates via filtration. The filtrate was then washed with aqueous hydrochloric acid, extracted, dried using magnesium sulphate and the solvent removed *in vacuo* to provide an oil that was purified via column chromatography (method ii, ethyl acetate:hexanes compound eluted in 30:70 v/v ethyl acetate:hexanes) to afford the title compound (87.0 mg, 0.31 mmol, 11%) as a pink oil. ν_{max} (ATR) 3182 (br, phenol O–H st), 1735 (ester C=O st), 1660 (amide C=O st), 1103 (C–O st); δ_H (700 MHz, $CDCl_3$) 8.79 (1H, s, H^{8}), 7.25–7.18 (1H, m, H^{5}), 7.04–6.97 (1H, m, H^{3}), 6.96–6.91 (1H, m, H^{6}), 6.83–6.77 (1H, m, H^{4}), 5.05–4.97 (1H, m, *J* 6.5 Hz, H^{14}), 4.68 (1H, d, *J* 15.0 Hz, H^{10}), 4.17 (1H, dd, *J* 9.3, 3.3 Hz, H^{11}), 4.10 (1H, d, *J* 15.0 Hz, H^{10}), 2.65–2.54 (1H, m, H^{1}), 2.43–2.36 (1H, m, H^{1}), 2.36–2.38 (1H, m, H^{12}), 2.13–2.03 (1H, m, H^{12}), 1.28 (3H, d, *J* 6.5 Hz, H^{15}), 1.16 (3H, d, *J* 6.5 Hz, H^{15}); δ_C (176 MHz, $CDCl_3$) 178 (C^{2}), 171 (C^{13}), 156 (C^{7}), 131 (C^{3}), 130.5 (C^{5}), 121 (C^{9}), 120 (C^{4}), 118 (C^{6}), 70 (C^{14}), 61 (C^{11}), 43 (C^{10}), 29 (C^{1}), 23 (C^{12}), 22 (C^{15}), 21.6 (C^{15}); *m/z* (ES-MS$^+$) 278.0 (100%, [M+H]$^+$); HRMS (ES-MS$^+$) MH$^+$found 278.1412, $[C_{15}H_{20}NO_4]^+$ requires 278.1392.

4.8.8. *Di-tert-butyl (2S)-2-{[(o-hydroxyphenyl)methyl]amino} glutarate, **10***

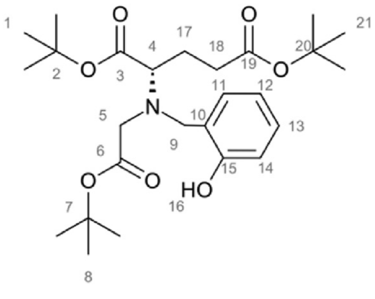


4.8.8.1. One-pot method. L-Glutamic acid di-tert butyl ester hydrochloride, **8** (500 mg, 1.69 mmol) was suspended in 1,2-dichloroethane (10 mL) and triethylamine (0.26 mL, 1.86 mmol) was added to the stirred mixture under nitrogen prior to the addition of salicylaldehyde (0.175 mL, 1.65 mmol). The resulting pale yellow suspension was stirred under nitrogen for 20 min and sodium triacetoxyborohydride (780 mg, 3.68 mmol) was added. After 3 h of stirring at room temperature, a further equivalent of sodium triacetoxyborohydride (800 mg, 3.77 mmol) was added and the reaction mixture was allowed to stir for a further 36 h under nitrogen, at which point the reaction was judged to be complete by TLC. A saturated aqueous potassium bicarbonate solution (20 mL) was then used to quench the reaction mixture, and the resulting mixture extracted into dichloromethane (3 × 50 mL). The organic extracts were recombined and washed with brine prior to solvent removal *in vacuo*. Purification of the resulting oil via column chromatography (silica, 0 → 20% ethyl acetate:hexanes) led to the isolation of the title molecule **10** as a colourless oil (450 mg, 1.23 mmol, 75%).

*4.8.8.2. Via imine **9**.* L-Glutamic acid di-tert butyl ester hydrochloride, **8** (1.00 g, 3.38 mmol) was suspended in chloroform (10 mL) and triethylamine (0.94 mL, 6.74 mmol) added. The reaction mixture was then heated at 50 °C for 90 min and then transferred into a separating funnel and washed with water and then brine. The yellow organic layer was then dried over magnesium sulphate and concentrated to afford **9** as a yellow solid (1.10 g, 3.03 mmol, 89%). Recrystallisation of a sample of **9** from hot ethanol gave single crystals suitable for analysis via X-ray crystallography. *Crystal data for **9***: $C_{14}H_{17}NO_4$, M = 363.44, orthorhombic, space group $P2_12_12_1$, a = 5.8887(2), b = 10.4967(4), c = 32.3124(13) Å, U = 1997.3(1) Å^3, F(000) = 784, Z = 4, D_c = 1.209 mg m^{-3}, μ = 0.703 mm^{-1}. 22828 reflections were collected (2Θ 5.47–141.89°) yielding 3816 unique data (R_{merg} = 0.0286). Final $wR_2(F^2)$ = 0.0628 for all data (351 refined parameters), conventional R_1 (F) = 0.0247 for 3752 reflections with $I \geq 2\sigma$, GOF = 1.092, Hooft = 0.05(4). CCDC-1551897.

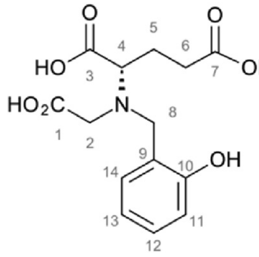
Imine **9** (1.10 g, 3.03 mmol) was then dissolved in methanol (12 mL) and sodium borohydride (230 mg, 6.05 mmol) was added at once **CAUTION**- gas evolution. The reaction mixture was stirred at room temperature for 4 h, after which the solvent was removed *in vacuo* and the residue taken up in chloroform (50 mL) and washed in saturated ammonium chloride (50 mL) to give an emulsion. The emulsion was extracted, dried over magnesium sulphate and concentrated *in vacuo*. Dichloromethane (50 mL) was added to the residue, which was then washed in brine (50 mL). The organic layer was extracted, dried over magnesium sulphate and concentrated *in vacuo* to afford **10** (775 mg, 2.11 mmol, 70%) as a yellow oil. $[\alpha]_D^{20} = -6.4$ (c = 0.28, DCM); ν_{max} (ATR) 2976 (sp^3 C–H), 1724 (ester C=O st), 1149 (ester C–O st); δ_H (700 MHz, $CDCl_3$) 7.18 (1H, dd, *J* 8.0, 1.5 Hz, H^8), 6.96 (1H, d, *J* 7.5, 1.5 Hz, H^{10}), 6.85 (1H, dd, *J* 8.0, 1.0 Hz, H^7), 6.78 (1H, dd, *J* 7.5, 1.0 Hz, H^9), 4.04 (1H, d, *J* 13.5 Hz, H^5), 3.74 (1H, d, *J* 13.5 Hz, H^5), 3.24 (1H, dd, *J* 6.0, 1.5 Hz H^4), 2.34–2.28 (2H, m, H^{12}), 2.03–1.91 (1H, m, H^{13}), 1.92–1.81 (1H, m, H^{13}), 1.50 (9H, s, H^1), 1.42 (9H, s, H^{16}); δ_C (176 MHz, $CDCl_3$) 173.0 (C^3), 172.0 (C^{14}), 157.9 (C^6), 129.2 (C^8), 128.8 (C^{10}), 122.3 (C^{11}), 119.4 (C^8), 116.7 (C^7), 82.3 (C^2), 80.9 (C^{15}), 60.0 (C^4), 51.1 (C^5), 31.9 (C^{13}), 28.5 (C^{12}), 28.3 (C^1), 28.2 (C^{16}); *m/z* (ES-MS$^-$) 366.2 (100%, [M–H]$^-$); HRMS (ES-MS)$^-$ MH$^-$ found 366.2288, $[C_{20}H_{32}NO_5]^-$ requires 366.2280.

4.8.9. *Di-tert-butyl (2S)-2-{[(o-hydroxyphenyl)methyl] (tert-butoxycarbonylmethyl) amino}glutarate, HBGl$_3$-tBu*



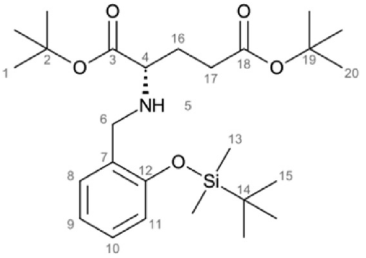
Aminoester **10**, (775 mg, 2.11 mmol) was dissolved in acetonitrile (10 mL, dried over molecular sieves) and potassium hydrogen carbonate (0.43 g, 4.30 mmol) was suspended in the stirred solution. A catalytic amount of potassium iodide (35.0 mg, 0.211 mmol) was then added followed by *t*-butyl bromoacetate (0.34 mL, 3.00 mmol). The mixture was then heated to 55 °C under argon for 12 days. Upon cooling to room temperature, the reaction mixture was filtered, concentrated *in vacuo* and the target compound isolated by column chromatography (ethyl acetate:hexanes 0:100 → 30:70 compound eluted in ca. 15:85 v/v ethyl acetate:-hexanes) as a colourless oil (470 mg, 0.981 mmol, 46%). $[\alpha]_D^{20} = -46.7$ (c = 0.094, DCM); ν_{max} (ATR) 3340 (O–H st), 2977 (sp^3 C–H st), 1722 (ester C=O st); 1137 (C–O st); δ_H (700 MHz, $CDCl_3$) 9.12 (1H, s H^{16}), 7.17 (1H, dd, *J* 7.5, 2.0 Hz, H^{13}), 6.96 (1H, dd, *J* 7.5, 2.0 Hz, H^{11}), 6.86 (1H, dd, *J* 7.5, 1.0 Hz, H^{14}), 6.75 (1H, ddd, *J* 7.5, 1.0 Hz H^{12}), 3.91 (1H, d, *J* 13.5 Hz, H^9), 3.73 (1H, d, *J* 13.5 Hz, H^9), 3.59 (1H, d, *J* 18.0 Hz, H^5), 3.26 (1H, d, *J* 18.0 Hz, H^5), 3.22 (1H, dd, *J* 9.0, 6.0 Hz, H^4), 2.37–2.19 (1H, m, H^{18}), 2.17–2.02 (1H, m, H^{18}), 2.02–1.84 (1H, m, H^{17}), 1.84–1.68 (1H, m, H^{17}), 1.48 (9H, s, H^{21}), 1.47 (9H, s, H^8), 1.35 (9H, s H^1); δ_C (176 MHz, $CDCl_3$) 172.3 (C^{19}), 172.0 (C^6), 171.2 (C^3), 157.3 (C^{15}), 130.2 (C^{11}), 129.5 (C^{13}), 121.6 (C^{10}), 119.3 (C^{12}), 116.6 (C^{14}), 82.4 (C^{20}), 81.8 (C^7), 80.4 (C^2), 60.5 (C^4), 55.9 (C^9), 51.5 (C^5), 31.8 (C^{18}), 28.3 (C^{21}), 28.1 (C^8), 28.0 (C^1), 24.7 (C^8); *m/z* (ES-MS$^+$) 480.5 (100%, [M+H]$^+$); HRMS (ES-MS$^+$) MH$^+$ found 480.2948, $[C_{26}H_{42}NO_7]^+$ requires 480.2961.

4.8.10. *(S)-2-{(Carboxymethyl)[(o-hydroxyphenyl)methyl]amino} glutaric acid, HBGl$_3$*



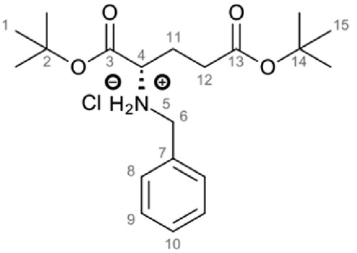
HBGl$_3$-tBu (110 mg, 0.229 mmol) was dissolved in dichloromethane (1 mL) and anisole (0.05 mL, 0.462 mmol) was added, followed by trifluoroacetic acid (1 mL). The mixture was stirred at room temperature for 24 h and the solvent removed *in vacuo*. The residue was then taken up in dichloromethane (ca. 5 mL) and the solvent was then removed *in vacuo*. The residue was triturated with hexane (ca. 25 mL) using ultrasonic agitation to afford a white precipitate that was collected via centrifugation (4000 rpm, 5 min). This solid was then dissolved in aqueous hydrochloric acid (1 M, 10 mL), transferred to a round bottom flask and the solvent removed *in vacuo* to give **HBGl$_3$** as an off-white solid (37.9 mg, 0.122 mmol, 53%). $[\alpha]_D^{20} = -14.3$ (c = 0.028, MeOH); t_R (H_2O:MeOH, 95:5 H_2O:MeOH to 5:95 H_2O:MeOH = 5 min, MS detection); 1.8 min; ν_{max} (ATR) 2960 (O–H st), 1730 (acid C=O st), 1220 (acid C–O st); δ_H (700 MHz, D_2O, pD ~ 2); 7.40 (1H, dd, *J* 8.0, 1.5 Hz, H^{12}), 7.36 (1H, dd, *J* 8.0, 1.5 Hz, H^{14}), 7.00 (1H, dd, *J* 8.0, 1.5 Hz, H^{13}), 6.98 (1H, dd, *J* 8.0, 1.5 Hz, H^{11}), 4.61 (1H, d, *J* 13.0 Hz, H^8), 4.51 (1H, d, *J* 13.0 Hz, H^8), 4.11 (1H, t, *J* 7.0 Hz, H^4), 4.09 (2H, s, H^2), 2.81–2.69 (1H, m, H^6), 2.67–2.61 (1H, m, H^6), 2.35–2.23 (2H, m, H^5); δ_C (176 MHz, D_2O, pD ~ 2) 176.1 (C^7), 170.7 (C^3), 169.4 (C^1), 155.4 (C^{10}), 132.5 (C^{14}), 132.2 (C^{12}), 120.7 (C^9), 115.6 (C^{13}), 115.5 (C^{11}), 64.4 (C^4), 54.7 (C^8), 52.6 (C^2), 30.2 (C^6), 21.25 (C^5); *m/z* (ES-MS$^+$) 312.1 (100%, [M+H]$^+$); HRMS (ES-MS$^+$) MH$^+$ found 312.1079, $[C_{14}H_{18}NO_7]^+$ requires 312.1083.

*4.8.11. Di-tert-butyl (2S)-2-{[(o-tertbutyldimethylsiloxyphenyl) methyl]amino}glutarate, **10a***



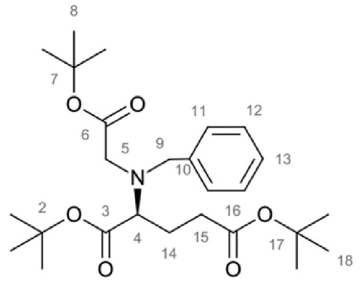
Aminoester **10** (0.283 g, 0.775 mmol) was dissolved in anhydrous dichloromethane (4 mL) and cooled in an ice bath prior to the addition of imidazole (160 mg, 2.35 mmol) and TBDMSCl (150 mg, 0.995 mmol) in rapid succession. The ice bath was then removed and the reaction mixture allowed to stir for 6 h at room temperature. The reaction mixture was then poured into a separating funnel and washed with water, and the organic layer was then extracted, dried over magnesium sulphate. Purification of the residue via column chromatography (method i, diethyl ether:hexanes 0:100 → 20:80 compound eluted in ca. 20:80 v/v diethyl ether:hexanes) gave **10a** as a colourless oil (190 mg, 0.396 mmol, 51%). δ_H (600 MHz, $CDCl_3$) 7.33 (1H, d, *J* 7.5, 1.5 Hz, H^{11}), 7.09 (1H, ddd, *J* 7.5, 1.5 Hz, H^9), 6.90 (1H, ddd, *J* 7.5, 1.0 Hz H^{10}), 6.76 (1H, dd, *J* 7.5, 1.0 Hz, H^8), 3.78 (1H, d, *J* 14.0 Hz, H^6), 3.65 (1H, d, *J* 14.0 Hz, H^6), 3.21–3.05 (1H, m, H^4), 2.41–2.31 (2H, m, H^{17}), 1.95–1.77 (3H, m, H^5, H^{16}), 1.44 (9H, s, H^1), 1.42 (9H, s, H^{20}), 1.00 (9H, s, H^{15}), 0.23 (6H, s, H^{13}); δ_C (151 MHz, $CDCl_3$) 174.4 (C^3), 172.7 (C^{18}), 153.6 (C^{12}), 130.8 (C^7), 129.4 (C^{11}), 127.7 (C^9), 121.1 (C^{10}), 118.5 (C^8), 81.0 (C^2), 80.2 (C^{19}), 61.0 (C^4), 47.1 (C^6), 32.1 (C^{17}), 28.9 (C^{16}), 28.2 (C^1), 25.9 (C^{20}), 18.2 (C^{15}), −4.05 (C^{13}, C^{14}); *m/z* (ES-MS$^+$) 480.1 (100%, $[M+H]^+$); HRMS (ES-MS$^+$) MH$^+$ found 480.3143, $[C_{26}H_{46}NO_5Si]^+$ requires 480.3145.

*4.8.12. Di-tert-butyl (2S)-2-benzylaminoglutarate, hydrochloride salt, **11***



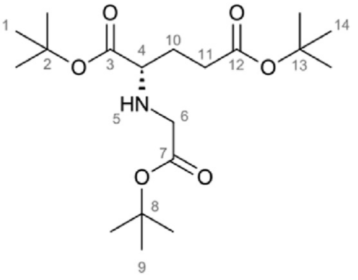
L-Glutamic acid di-tert butyl ester hydrochloride, **8** (500 mg, 1.69 mmol) and triethylamine (0.28 mL, 3.82 mmol) were added to methanol (8 mL), resulting in the formation a white precipitate. Benzaldehyde (0.18 mL, 1.78 mmol) was then added and the mixture stirred under nitrogen for 2.5 h. Sodium borohydride (230 mg, 6.07 mmol) was subsequently added portionwise **CAUTION-** gas evolution and the mixture was allowed to stir under nitrogen for a further 3 h. Volatiles were then removed *in vacuo* and the residue partitioned between dichloromethane and aqueous hydrochloric acid (1 mol dm^{-3}) and the organic layer extracted, washed with brine, dried with magnesium sulphate and the solvent removed *in vacuo*. The resulting white solid was triturated in hexane to afford the title compound (290 mg, 0.751 mmol (based on molecular weight of the monohydrochloride salt), 45%) as a white powder. mp 140–142 °C; ν_{max} (ATR); 2982 (sp^3 C–H st), 2631 (N^+–H st), 1725 (ester C=O st), 1149 (C–O st); δ_H (600 MHz, $CDCl_3$) 10.81 (1H, s, H^5), 9.84 (1H, s, H^5), 7.63 (2H, d, *J* 7.0 Hz, H^8), 7.42–7.31 (3H, m, H^9,H^{10}), 4.24 (2H, dd, *J* 22.0, 11.5 Hz, H^6), 3.62 (1H, s, H^4), 2.63–2.22 (4H, m, H^{11}, H^{12}), 1.51 (9H, s, H^{15}), 1.35 (9H, s, H^1); δ_C (151 MHz, $CDCl_3$) 171.3 (C^{13}), 166.7 (C^3), 131.0 (C^8), 130.0 (C^7), 129.7, 129.3 (C^9, C^{10}), 84.9 (C^{14}), 81.3 (C^2), 57.5 (C^4), 49.7 (C^6), 31.3, 28.2 (C^{15}), 28.1 (C^1) 25.2 (C^{11}, C^{12}); *m/z* (ES-MS$^+$) 350.2 (100%, $[M+H]^+$); HRMS (ES-MS$^+$) MH$^+$ found 350.2330, $[C_{20}H_{32}NO_4]^+$ requires 350.2331.

*4.8.13. Di-tert-butyl (2S)-2-[(1R)-(benzyl)(tert-butoxycarbonylmethyl)amino]glutarate, **12***



Aminoester **11** (400 mg, 1.04 mmol) was added to acetonitrile (3.5 mL, Aldrich anhydrous) and potassium carbonate (570 mg, 4.13 mmol), potassium iodide (30.0 mg, 0.181 mmol) salts were added along with *t*-butyl bromoacetate (0.23 mL, 1.57 mmol) and the mixture was heated at 60 °C under nitrogen. TLC (ethyl acetate:hexanes 20:80 v/v) indicated that consumption of starting material was achieved within 6 h, but heating was maintained for a further 16 h prior to cooling to room temperature and removal of solids by filtration. The filtrate was concentrated *in vacuo* and subjected to column chromatography (method ii, diethyl ether:hexanes 0:100 → 40:60 compound eluted in ca. 15:85 v/v diethyl ether:hexanes) to afford the title compound (240 mg, 0.518 mmol, 50%) as a colourless oil. ν_{max} (ATR); 2974 (sp^3 C–H st), 1719 (ester C=O st), 1137 (ester C–O st); δ_H (700 MHz, $CDCl_3$); 7.40–7.22 (5H, m, H^{11}, H^{12}, H^{13}), 3.95 (1H, d, *J* 13.5 Hz, H^5), 3.74 (1H, d, *J* 13.5 Hz, H^5), 3.43 (1H, d, *J* 17.0 Hz, H^9), 3.34–3.20 (2H, m, H^9, H^4), 2.48 (1H, ddd, *J* 16.5, 9.5, 6.0 Hz, H^{15}), 2.26 (1H, ddd, *J* 16.5, 9.5, 6.0 Hz, H^{15}), 1.96–1.85 (2H, m, H^{14}), 1.50 (9H, s, H^8), 1.43 (9H, s, H^{18}), 1.40 (9H, s, H^1); δ_C (176 MHz, $CDCl_3$); 172.9 (C^{16}), 171.9 (C^6), 170.9 (C^3), 129.3, 128.4, 127.4 (C^{10}, C^{11}, C^{12}, C^{13}, two of which are overlapping), 81.5 (C^{17}), 80.9 (C^7), 80.2 (C^2), 62.4 (C^4), 56.7 (C^5), 52.7 (C^9), 32.0 (C^{15}), 28.5 (C^{18}), 28.3 (C^8), 28.2 (C^1), 25.2 (C^{14}); *m/z* (ES-MS$^+$) 464.3 (100%, $[M+H]^+$); HRMS (ES-MS$^+$) MH$^+$ found 464.3009, $[C_{26}H_{42}NO_6]^+$ requires 464.3012.

*4.8.14. Di-tert-butyl (2S)-2-[(tert-butoxycarbonylmethyl)amino] glutarate, **13***



4.8.14.1. Via intermediate ***12****.* Ethanol (25 mL) was used to dissolve **12** (80.0 mg, 0.172 mmol) and the colourless solution was evacuated and back-filled with nitrogen 3 times prior to the addition of 5% palladium on charcoal (7 mg, 3 μmol with respect to Pd). Once the catalyst was added, the reaction mixture was evacuated and back-filled with hydrogen gas from a balloon three times prior to stirring at room temperature. Reaction progress was monitored by the use of ^{1}H NMR which indicated that debenzylation was achieved within 4 h. The reaction mixture was subsequently allowed to stand for a few minutes under a flow of nitrogen prior to catalyst removal via filtration over Celite®. Concentration of the filtrate *in vacuo*, followed by re-dissolution of the residue in chloroform and a second solvent removal *in vacuo* led to isolation of the title molecule (51 mg, 0.144 mmol, 84%) as a colourless oil.

4.8.14.2. Direct synthesis from commercially available starting materials. L-Glutamic acid di-tert butyl ester hydrochloride, **8** (1.50 g, 5.07 mmol) was stirred at room temperature in chloroform under argon in the presence of activated zinc dust (3.00 g, 46.1 mmol) for 1 h. The zinc dust was then filtered off, and the solvent removed from the filtrate *in vacuo* to afford the free base. The resulting oil was dissolved in acetonitrile (28 mL, dried over molecular sieves). When the solution was nearly homogeneous, anhydrous triethylamine (1.21 mL, 8.68 mmol) was added at once, followed by the dropwise addition of *t*-butyl bromoacetate (0.89 mL, 6.07 mmol). Finally, a catalytic amount of potassium iodide (ca. 0.1 g) was added and the resulting mixture stirred at room temperature under argon for 24 h. After the solvent was removed *in vacuo*, the oily residue was partitioned between water and dichloromethane (50 mL of each) and the combined organic extracts were subsequently washed in brine prior to being dried with magnesium sulphate and the solvent being removed *in vacuo*. The resulting oil was dissolved in the minimum volume of dichloromethane and subjected to column chromatography (method i, silica, ethyl acetate:hexanes 30:70 → 60:40) to afford the pure compound (0.44 g, 1.18 mmol, 23%) as a colourless oil.

ν_{max} (ATR) 2970 (N–H st), 2930 (sp^3 C–H), 1731 (ester C=O st), 1149 (ester C–O st); δ_H (700 MHz, CDCl$_3$) 3.29 (1H, d, *J* 17.0 Hz, H^6), 3.22 (1H, d, *J* 17.0 Hz, H^6), 3.14 (1H, dd, *J* 7.5, 6.0 Hz, H^4), 2.34 (2H, t, *J* 7.5 Hz, H^{11}), 1.92–1.89 (2H, m, H^5, H^{10}), 1.88–1.76 (1H, m, H^{10}), 1.46 (9H, s, H^1), 1.44 (9H, s, H^{14}), 1.43 (9H, s, H^9); δ_C (176 MHz, CDCl$_3$) 173.8 (C^3), 172.8 (C^{12}), 171.3 (C^7), 81.7 (C^2), 81.5 (C^{13}), 80.5 (C^8), 60.8 (C^4), 50.2 (C^6), 32.2 (C^{11}), 28.6 (C^{10}), 28.4 (C^1, C^9, C^{14}); *m/z* (ES-MS$^+$) 374.2 (100%, [M+H]$^+$); HRMS (ES-MS$^+$) MH$^+$ found 374.2538, [C$_{19}$H$_{36}$NO$_6$]$^+$ requires 374.2543.

Notes

Crystallographic data for the structures in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication numbers 1551895–1551897.

Acknowledgements

We thank P&G and Durham University for support of this work.

Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.tet.2017.09.032.

References

1. Messeri D, Lowe MP, Parker D, Botta M. *Chem Commun*. 2001:2742–2743.
2. Pal R, Parker D. *Org Biomol Chem*. 2008;6:1020.
3. Elemento EM, Parker D, Aime S, Gianolio E, Lattuada L. *Org Biomol Chem*. 2009;7:1120.
4. Boethling RS, Sommer E, DiFiore D. *Chem Rev*. 2007;107:2207–2227.
5. Itrich NR, McDonough KM, van Ginkel CG, et al. *Environ Sci Technol*. 2015;49:13314–13321.
6. Henig J, Tóth É, Engelmann J, Gottschalk S, Mayer HA. *Inorg Chem*. 2010;49:6124–6138.
7. Sreenivasulu B, Vittal JJ. *Angew Chem Int Ed*. 2004;116:5893–5896.
8. Frost AE, Martell AE. *J Am Chem Soc*. 1950;72:3743–3746.
9. Greene TW, Wuts PGM. *Protective Groups in Organic Synthesis*. third ed. New York: Wiley; 1999.
10. Abdel-Magid AF, Carson KG, Harris BD, Maryanoff CA, Shah RD. *J Org Chem*. 1996;61:3849–3862.
11. Price EW, Cawthray JF, Bailey GA, et al. *J Am Chem Soc*. 2012;134:8670–8683.
12. Bechara G, Leygue N, Galaup C, Mestre-Voegtlé B, Picard C. *Tetrahedron*. 2010;66:8594–8604.
13. Koll A, Melikova SM, Karpfen A, Wolschann P. *J Mol Struct*. 2001;559:127–145.
14. Labby KJ, Xue F, Kraus JM, et al. *Bioorg Med Chem*. 2012;20:2435–2443.
15. Harris WR, Motekaitis RJ, Martell AE. *Inorg Chem*. 1975;14:974–978.
16. Cui X, Cabral MF, Costa J, Delgado R. *Inorg Chim Acta*. 2003;356:133–141.
17. Jarjayes O, Mortini F, du Moulinet d'Hardemare A, Philouze C, Serratrice G. *Eur J Inorg Chem*. 2005;2005:4417–4424.
18. Still WC, Kahn M, Mitra A. *J Org Chem*. 1978;43:2923–2925.
19. Dolomanov OV, Bourhis LJ, Gildea RJ, Howard JAK, Puschmann H. *J Appl Crystallogr*. 2009;42:339–341.
20. Sheldrick GM. *Acta Crystallogr A*. 2008;64:112–122.