Organic & Biomolecular Chemistry

ROYAL SOCIETY OF CHEMISTRY

PAPER

View Article Online
View Journal | View Issue

Cite this: *Org. Biomol. Chem.*, 2024, **22**, 590

Increasing the versatility of the biphenyl-fused-dioxacyclodecyne class of strained alkynes†

Sam Forshaw, [a] Jeremy S. Parker, [b] William T. Scott, [a,c] Richard C. Knighton, [a,d] Neelam Tiwari,[a] Samson M. Oladeji,[a] Andrew C. Stevens,[a] Yean Ming Chew, [a,c] Jami Reber, [a] Guy J. Clarkson, [a] Mohan K. Balasubramanian[c] and Martin Wills *[a]

Received 19th October 2023,
Accepted 8th December 2023

DOI: 10.1039/d3ob01712e

rsc.li/obc

Biphenyl-fused-dioxacyclodecynes are a promising class of strained alkyne for use in Cu-free 'click' reactions. In this paper, a series of functionalised derivatives of this class of reagent, containing fluorescent groups, are described. Studies aimed at understanding and increasing the reactivity of the alkynes are also presented, together with an investigation of the bioconjugation of the reagents with an azide-labelled protein.

Introduction

Bioconjugations[1] using Strain-Promoted Alkyne–Azide Cycloadditions (SPAAC)[2] are important reactions, due to their high reaction rates, the lack of a requirement for a catalyst,[3] and their bioorthogonal reactivity. A number of strained alkynes have been widely adopted for synthetic and biolabelling applications.[4] Early examples such as OCT **1**,[4a] and fluorinated derivatives such as DIFO **2**[4c] were followed by highly reactive, strained alkynes such as DIBO **3**,[4d,e] BCN **4**,[4f] DIBAC **5**[4g] and BARAC **6**[4h] (Fig. 1), which have been employed in numerous biolabelling applications. The second order rate constant for each alkyne with benzylazide provides a convenient means for comparison of their reactivity (Fig. 1). The high reactivity of the strained alkynes is reflected by the distorted sp bond angles. Derivatives of strained alkynes, loaded with a fluorescent group, also undergo cyclisations with azide-containing molecules both *in vitro* and *in vivo* without the need for a catalyst.

We,[5–7] and others,[8] recently reported the synthesis of strained alkynes of general structure **7**, where X/Y = O, NH, NTs,[9] as reagents for copper-free cycloaddition reactions with azides (Fig. 2). Specific examples of this class of alkyne are **8–14** and, although not as reactive as some of the well-established strained alkynes shown in Fig. 1, they benefit from the straightforward introduction of the alkyne through the reaction of a 2,2′-biphenol reagent with 1,4-ditosylbut-2-yne, and readily react with azides at concentrations above *ca.* 0.1 M. Alabugin *et al.*[8] described how the 'twisted' structure of this class of dioxacyclodecyne is alleviated upon approach of the azide. This effect generates improved reactivity when the heteroatom (X, Y) in the structure is an oxygen or a nitrogen atom.

Altering the heteroatoms X/Y in **7** influences their reactivity; Alabugin[8] studied biphenyl-fused-diazacyclodecyne derivative

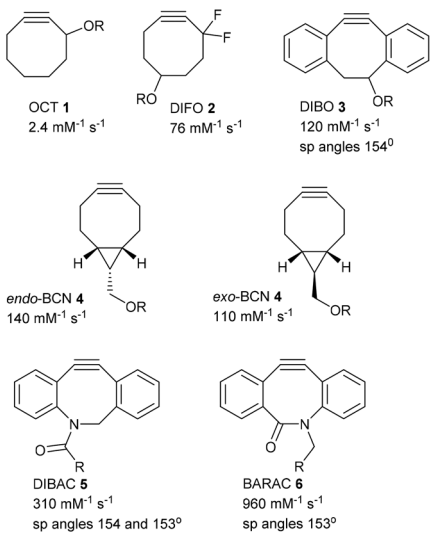


Fig. 1 Strained alkynes and their second order rate constants for addition to benzyl azide in MeCN, MeOH or MeCN/H_2O at rt.[2d] R = functional group.

[a]*Department of Chemistry, The University of Warwick, Coventry, CV4 7AL, UK. E-mail: M.wills@warwick.ac.uk*
[b]*Early Chemical Development, Pharmaceutical Sciences, IMED Biotech Unit, AstraZeneca, Macclesfield, SK10 2NA, UK*
[c]*Warwick Medical School, The University of Warwick, Coventry, CV4 7AL, UK*
[d]*School of Chemistry, University of Southampton, SO17 1BJ, UK*

† Electronic supplementary information (ESI) available: Spectroscopic data, X-ray crystal structure and bioconjugation results. CCDC 2273606–2273609. For ESI and crystallographic data in CIF or other electronic format see DOI: https://doi.org/10.1039/d3ob01712e

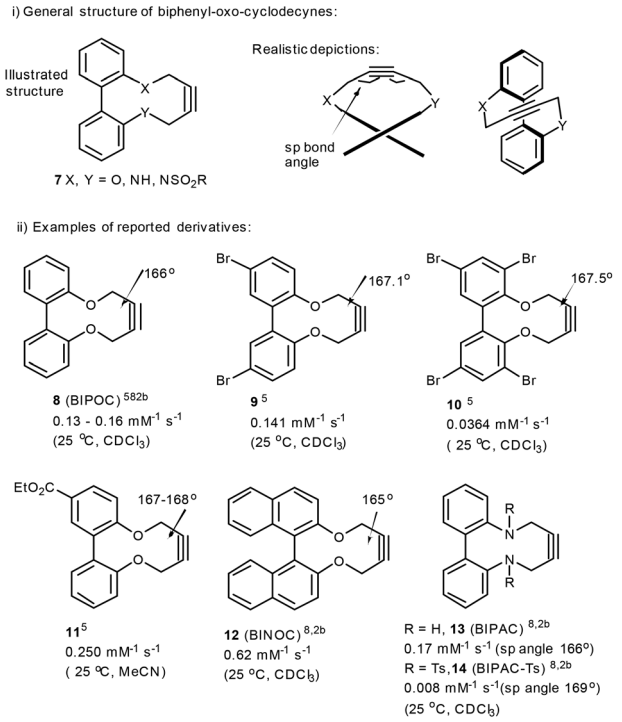


Fig. 2 Biphenyl-fused dioxacyclodecynes and second order rate constants for addition with benzylazide under the conditions shown. Bond angles were established by X-ray crystal structure analyses or DFT calculations.

13 and observed a similar rate constant to that of the unsubstituted biphenyl-dioxacyclodecyne **8**, when reacted with benzyl azide in $CDCl_3$ at rt. The *p*-toluenesulfonamide derivative **14** exhibited a lower rate of reaction, corresponding to its less distorted sp bond angle of 169° (Fig. 2).[8] This is less distorted than in the more reactive unsubstituted alkyne **8** which has an sp angle of *ca.* 166° and significantly less than for highly reactive alkynes such as DIBAC and BARAC (Fig. 1). In this paper, we describe our studies aimed at expanding the range of biphenyl-fused-dioxacyclodecyne reagents, and at increasing their reactivity in Cu-free click reactions.

Results and discussion

Fluorescent enone-containing derivatives

Enones can exhibit fluorescent properties. A report by Xing *et al.*[10] indicated that an effective pairing was a compound containing a methoxy group and a dimethylamino group *para*- to the ketone and aldehyde enone precursors respectively. Alkyne **15**[6] was reacted under basic conditions with 4-dimethylaminobenzaldehyde to give **16** in good yield (Fig. 3).

Compound **16** exhibited strong fluorescence excitation and emission maxima at 420 nm and 536 nm respectively. The fluorescent data for the benzyl azide addition product **17** (formed as an inseparable 1 : 1 mixture of regioisomers) exhibited excitation and emission wavelengths essentially

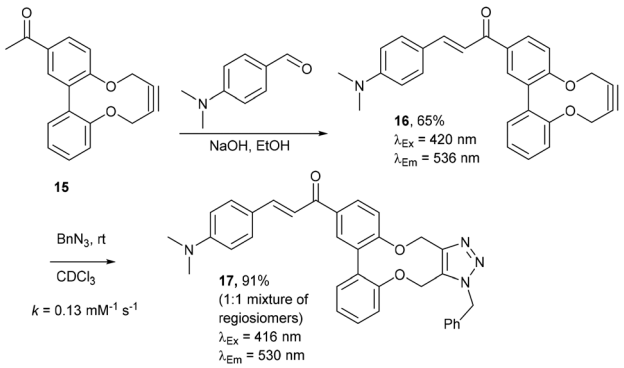


Fig. 3 Synthesis of enone alkyne, **16**, and subsequent cycloaddition of benzyl azide to form triazole **17**.

unchanged from alkyne **16**. The rate constant for the cycloaddition was 0.13 mM^{-1} s^{-1}, similar to that of biphenyl-fused-dioxacyclodecyne **8**. The reaction of 2,2′-biphenol with an excess of $AlCl_3$ and acetyl chloride gave **18** in moderate yield (47%). The ester groups were then hydrolysed using lithium hydroxide to give the diacetyl biphenol **19**, and its cyclisation with ditosylate **20** gave the strained diacetyl alkyne **21** (Fig. 4). Dienone **22** was then formed using the same conditions as for the synthesis of compound **17**, using two equivalents of 4-dimethylaminobenzaldehyde. Unexpectedly, the reaction rate for the reaction between dienone **22** and benzyl azide (k = 0.25 mM^{-1} s^{-1}) to give **23** was double that for enone **16**, possibly due to steric effects between the two large enone groups on the opposite side to the alkyne.

Two further derivatives, **24** and **25**, containing fluorescein and rhodamine groups respectively, were prepared through DCC couplings with known fluorescent precursors **26** and **27**, and the strained alkyne **28** (Fig. 5).[11,12] These were available

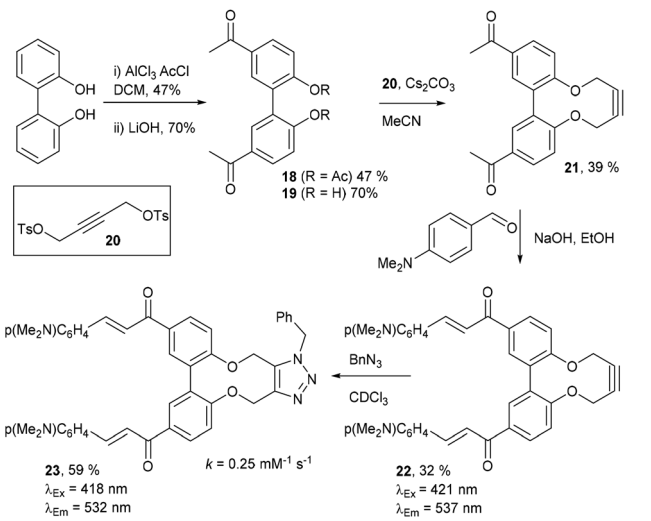


Fig. 4 Synthesis of dienone alkyne, **22**, and subsequent cycloaddition with benzyl azide to give triazole, **23**.

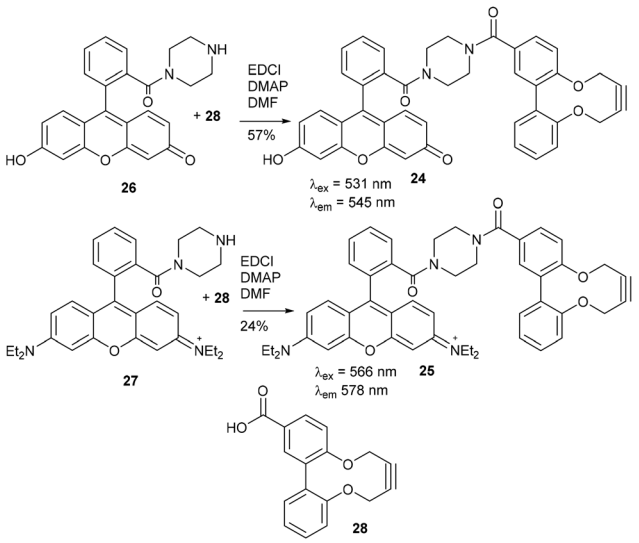


Fig. 5 Synthesis of fluorescein and rhodamine-containing strained alkynes.

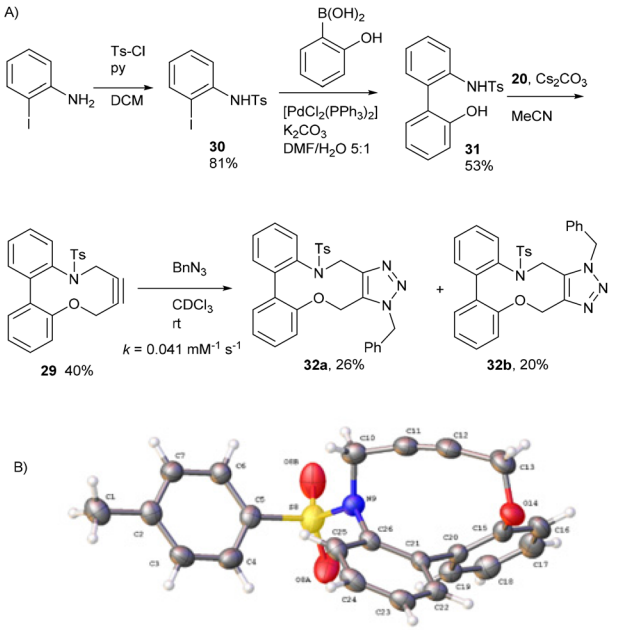


Fig. 6 (A) The synthetic route to the heterocyclic alkyne **29** and subsequent reaction with BnN_3. The regiochemistry of each isomer of **32** has not been confirmed. (B) X-ray crystal structure of **29**.

for subsequent testing with an azide-functionalised protein, which is described in a later section.

Further derivatives and attempts to increase the reactivity of the alkynes

To understand the significance of the heteroatoms on the reactivity, compound **29**, containing a combination of an oxygen and toluenesulfonamide heteroatoms, was prepared. 2-Iodoaniline was converted to **30** which was coupled under Suzuki conditions with 2-hydroxyphenylboronic acid to give the biphenyl **31**, and subsequently converted to **29** in moderate yield (Fig. 6a).

Alkyne **29** underwent cycloaddition with benzyl azide in $CDCl_3$ with a second order rate constant of just 0.041 mM^{-1} s^{-1}, to form **32**, as a mixture of isomers. This rate is lower than the same reaction of biphenyl-fused-dioxacyclodecyne, **8**, but higher than the reaction of biphenyl-NTs-alkyne, **14**. The X-ray crystal structure of **29** (Fig. 6b) revealed sp bond angles of *ca.* 169.5 and 167.4° respectively. The synthesis of other cyclic alkynes was considered, including the use of sulfur as a heteroatom. However, the introduction of sulfur atoms generally diminishes the reactivity of the alkyne due to the larger bond length of the sulfur–carbon bonds.[13,14] In an earlier result published by Wills *et al.*,[7] bisalkyne **33** (Fig. 7) was used in 'protein stapling' reactions. Analysis of the reaction by NMR, which featured direct formation of **34** without the monoadduct **35**, suggested that the first cycloaddition was rate limiting and that the second cycloaddition occurred much more rapidly. Molecular modelling confirmed that the transition state for the second cycloaddition had a lower energy barrier than the first. This increase in reactivity is likely caused by an increased distortion of the remaining alkyne bond.

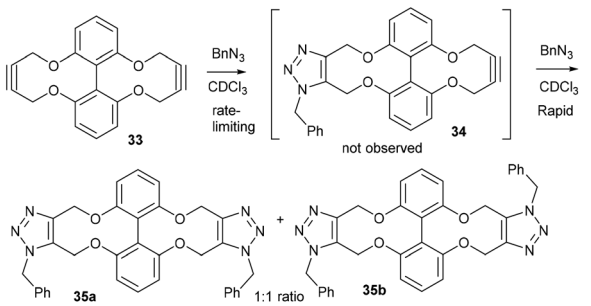


Fig. 7 The reaction between bisalkyne, **33**, and benzyl azide.

It was speculated that harnessing this effect into a strained alkyne could be beneficial. To achieve this, we studied the effects that functional groups at the 6 and 6′ positions have on the rates of cycloaddition. The known biphenol **36**,[15] was converted to 6,6′-dimethoxybiphenyl-dioxacyclodecyne **37** in low yield (Fig. 8) but sufficient material was isolated to test the addition reaction. The rate constant for the reaction of **37** with

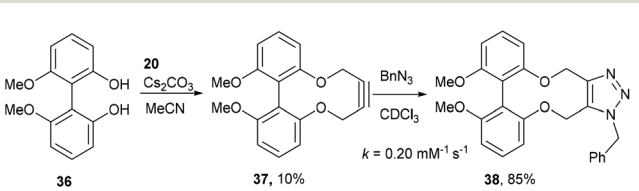


Fig. 8 Synthetic route to 6,6′-dimethoxybiphenyl-dioxacyclodecyne **37** and subsequent click reaction with benzyl azide in $CDCl_3$.

BnN_3, forming adduct **38**, was 0.20 mM^{-1} s^{-1}, indicating that methoxy groups at these positions have little effect on the rate of reaction or the structure of the alkyne and the distortion of the alkyne bond.

A route to asymmetric biphenols with a bridge between the 6 and 6′ positions has been reported using a removable chiral bridging group.[16,17] Using this approach, dimethylsulfonate, **39** was reacted with the known tetrol **40**[7] under the conditions reported by Harada *et al.*,[16] producing the ethyl bridged biphenol **41**,[18] in moderate yield. Cyclisation with 1,4-dibromobutane formed the bicyclic compound **42**. Lithium di-*tert*-butylbiphenyl (LiDBB) cleaved the more strained ethyl bridge in **42** selectively to produce **43** in good yield and this was then cyclised with alkyne **20** to give the 4C bridged strained alkyne **44** (Fig. 9).[8]

The 4C-oxo-bridged biphenyl-fused-dioxabiphenylcyclodecyne **44**, reacted with benzyl azide to give adduct **45**, with a rate constant of 2.1 mM^{-1} s^{-1}, representing an increase compared to analogous biphenyl-fused-dioxacyclodecyne **37** (k = 0.17 mM^{-1} s^{-1}). The increase in reactivity is likely caused by the 6,6′-4C bridge forcing the phenyl rings to lie in a more planar structure and providing more distortion to the alkyne bond angles. To increase this effect further, the synthesis of a three-carbon bridged derivative was attempted, however this was not successful. Given the promising result with **44**, an N-containing C4-bridged reagent was prepared. Ullmann homo-coupling of **46**[19] with activated copper was carried out to give protected biphenol **47** in high yield. An attempt at the Ullmann coupling of the unprotected analogue of **46** was unsuccessful. Deprotection of **47** gave biphenol **48** in high yield, which was then cyclised to the strained alkyne **49** (Fig. 10).

It was found that the use of iron powder and ammonium chloride selectively reduced the nitro groups to amines to give **50**, leaving the alkyne intact (Fig. 10). 6,6′-Diamino-dioxacyclodecyne **50** was then reacted with toluenesulfonyl chloride, dansyl chloride and mesyl chloride under basic conditions to give **51** (Ts), **52** (Dns) and **53** (Ms) respectively. To create the anticipated more reactive derivatives, each bisulfonamide was reacted with 1,4-dibromobutane under basic conditions using a syringe pump to maintain a pseudo-dilute solution.[8] These studies afforded the bridged product, **54**, in moderate yield from the ditosylate precursor. Unfortunately, the bis-dansyl precursor **52** gave no corresponding product **55**, likely due to increased steric hindrance. Tests on the cyclisation of dimesylate **53** with varying equivalents of 1,4-dibromobutane revealed that the use of three equivalents of the dibromide gave an improved yield of C4-cyclised product **56** over the use of one equivalent. This was surprising as we were concerned that an excess of the dihalide would result in dialkylation of the dimesylate prior to intramolecular cyclisation. However, the improved yield indicates that the intramolecular step must outpace the second *N*-alkylation. The cycloadditions of the new alkynes with benzyl azide in $CDCl_3$ (at rt) were tested (Fig. 10). For **49**, the rate constant calculated for this reaction was 0.64 mM^{-1} s^{-1}; an improvement of about a factor of four compared to **37**, suggesting that the electron-withdrawing nitro group increases the reactivity. The X-ray crystal structure of **49** (Fig. 11a) indicates that the alkyne bond angles average 165.6°, more distorted than the biphenyl-fused-dioxacyclodecyne, **8**. For diamine **50**, the rate constant was 0.40 mM^{-1} s^{-1};

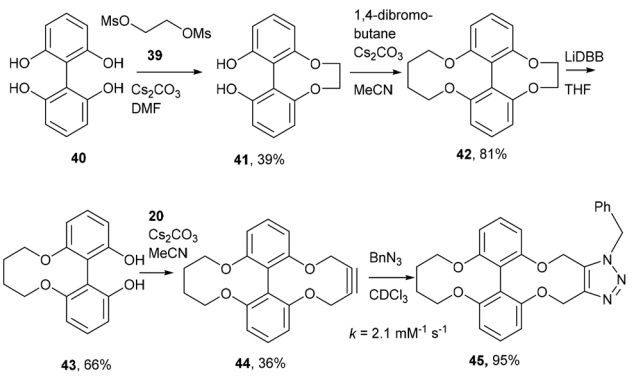


Fig. 9 Synthetic route to **44** and subsequent cycloaddition reaction with benzyl azide to produce triazole **45**.

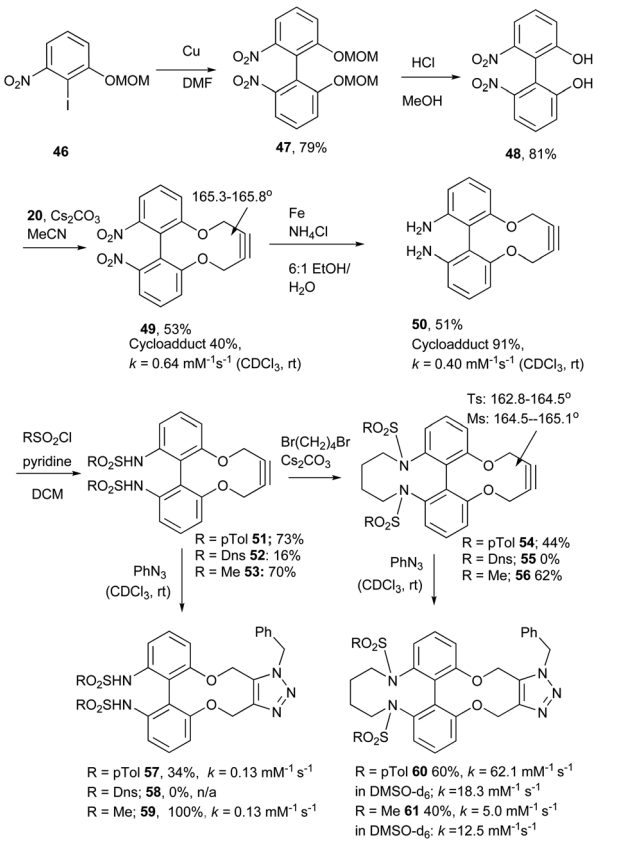


Fig. 10 Synthetic route to the 6,6′-dinitrobiphenyl-dioxacyclodecyne **49** and derivatives, with rate constraints for subsequent cycloadditions with benzyl azide in $CDCl_3$(unless otherwise indicated) Where sp bond angles are given, these were determined by X-ray crystallography (Fig. 11).

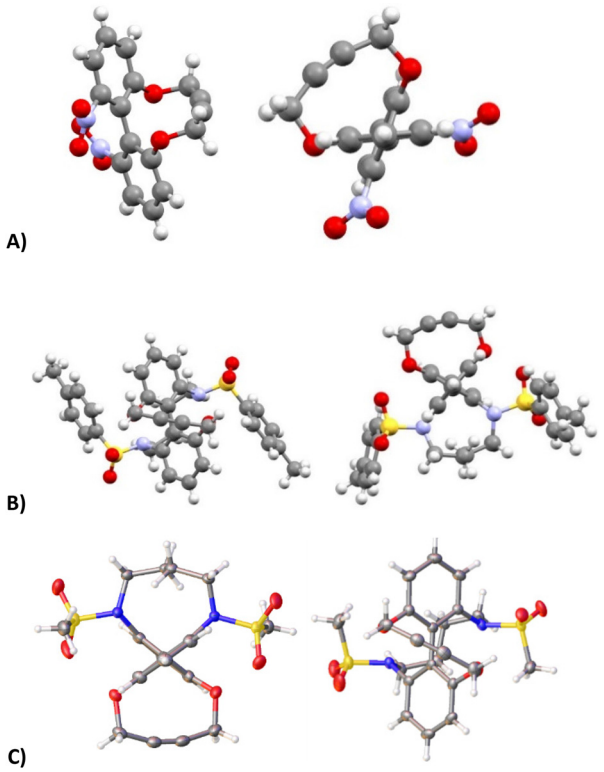


Fig. 11 (A) Two views of the X-ray crystal structure of 6,6′-dintrobiphenyl-dioxacyclodecyne **49**. (B) Two views of the X-ray crystal structure of bridged alkyne **54**. (C) Two views of the X-ray crystal structure of bridged alkyne **56**.

slightly lower than that for **49**, but slightly higher than that observed for the reaction with biphenyl-fused-dioxacyclodecyne **37**. Bisulfonylated compounds **51** and **53** produced the triazole products **57** and **59** respectively upon reaction with benzyl azide, however due to the low solubility of alkyne **52**, a rate constant could not be accurately determined and the anticipated product **58** was not isolated. The reaction between **51** (Ts) and **53** (Ms) and benzyl azide gave rate constants of 0.13 mM^{-1} s^{-1} in each case, similar to that of biphenyl-fused-dioxacyclodecyne **8**. However, the corresponding cycloaddition reactions of **54** and **56** proceeded with significantly higher rate constants of 62.1 and 5.0 mM^{-1} s^{-1} respectively, in $CDCl_3$ to give products **60** and **61** respectively. The reaction of **54** in an NMR tube was substantially complete within 5 hours ([alkyne] = 0.04 mM), representing a step change in reactivity for this class of strained alkynes. X-ray structures of **54** and **46** (Fig. 11b and c), revealing the alkyne sp bond angles to be 163.7° and 162.8° in **54** and 164.0°/165.1° in **56**. The difference in average alkyne bond angle between compound **54** and compound **49** is only 2.3°, which shows even small changes to the bond angle can have a great influence on the reaction rate. For **56** (diMs) the angles were intermediate, and this was reflected in its reactivity.

When comparing the rate of the reaction between alkyne **54** and benzyl azide with previously published strained alkynes it displays similar reactivity to the difluorinated cyclooctynes, which display rate constants between 42–76 mM^{-1} s^{-1}.[2d] The comparative rate constants of the novel compounds in this study give an insight into how electronic and structural effects can combine to produce more reactive alkynes. Although using electron withdrawing groups at the 6 and 6′ position in **49** did improve the reactivity, the largest increases in reactivity came with the addition of the 6,6′-4C bridge, *i.e.* in **44**, **54** and **56**. The reason for the difference in reactivity between **54** and **56** may stem from an increase strain created by the bulky tosyl groups creating increased distortion in the alkyne bond.

To establish whether the new alkynes may be compatible with biomolecules, attempts were made to react the novel alkynes with glutathione *S*-transferase (GST) containing an azidophenylalanine at position 52. In an initial series of tests, an earlier-reported BoDIPY-containing strained alkyne, fluorescein **24** and rhodamine derivative **25** were reacted and a gel indicated that conjugation had occurred in most cases. However, MS analyses of the adducts indicated that this was only the case for the fluorescein derivative **24**, hence there may be non-covalent, non-specific interactions between protein and dye in the other cases (see the ESI†). In a second round of tests of non-fluorescent compounds, the dimesylated compound **56** gave an addition product when analysed by mass spectrometry, although the more reactive ditosylated **54** did not. Examination of the second order rate constant for the reaction with BnN_3 in DMSO-d_6 (*i.e.* reflecting more closely the conditions used in the enzyme reactions where DMSO/H_2O was used) gave k values of 18.3 and 12.5 mM^{-1} s^{-1} for **54** and **56** respectively. Hence the rates of each compound were closer in DMSO-d_6 than in $CHCl_3$. Coupled to a potential lower solubility of the larger molecule in the water/DMSO mixture used with the enzyme may account for differences in the observed results. Compound **44**, bearing a 4C aliphatic linking group, added to the protein, but at a low level. See the ESI† for full details of these tests.

Conclusion

The development of novel strained alkynes for use in bioconjugation is still a focus of international research. Biphenyl-fused-dioxacyclodecynes react with azides without the need for a catalyst. The current investigation into the reactivity of this class of strained alkyne has led to the synthesis of a variety of 6,6′-functionalised biphenyl-fused-dioxacyclodecyne derivatives with rate constants in the region of 0.13–0.64 mM^{-1} s^{-1}. This inspired the synthesis of a four-carbon bridged class of biphenyl-fused-dioxacyclodecynes, which are more reactive towards azides, with rate constants between 2.13–62.1 mM^{-1} s^{-1}. There is potential for functionalisation of the sulfonamide groups of the C4-bridged alkynes, *e.g.* with fluorescent groups, which could provide a reactive, fluorescent strained biphenyl alkyne for use in bioimaging. Further studies of the value of the alkynes, of which **56** represents a promising candidate for further applications, are ongoing.

Experimental section

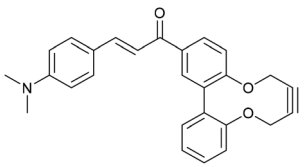
Solvents and reagents were degassed before use and all reactions were carried out under a nitrogen atmosphere using vacuum line apparatus. Reactions were monitored by TLC using aluminium backed silica gel 60 (F254) plates, visualized using UV 254 nm and phosphomolybdic acid or potassium permanganate as appropriate. Flash column chromatography was carried out routinely on silica gel. Reagents were used as received from commercial sources unless otherwise stated. Dry solvents were purchased and used as received. ^{1}H NMR spectra were recorded on a Bruker DPX (300, 400 or 500 MHz) spectrometer. Chemical shifts are reported in δ units, parts per million relative to the singlet at 7.26 ppm for chloroform and 0.00 ppm for TMS. Coupling constants (J) are measured in hertz. Mass spectra for analysis of synthetic products were recorded on a Bruker Esquire 2000 or a Bruker MicroTOF mass spectrometer. IR spectra were recorded on a PerkinElmer Spectrum One FT-IR Golden Gate. Melting points were recorded on a Stuart Scientific SMP 1 instrument and are uncorrected. X-ray crystallography was carried out on a Rigaku Oxford Diffraction SuperNova diffractometer with a duel source (Cu at zero) equipped with an AtlasS2 CCD area detector or an Xcalibur Gemini diffractometer with a Ruby CCD area detector. The procedure and full details of the kinetic ^{1}H NMR runs are given in the ESI.†

Safety and hazards

All synthetic organic chemistry has potential hazards, however azides are known to be highly reactive and required full risk assessment and care in handling throughout their preparation, use and disposal. In this study, small amounts of benzylazide (typically <10 mg) were used in NMR-scale tests of reactivity with the strained alkynes.

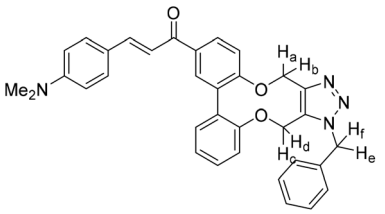
The following compounds were prepared following published methods; ditosyl-1,4-dihydroxybut-2-yne **20**,[21] *N*-tosyl-2-iodoaniline **30**,[20] dimethoxydiphenol **36**,[15] tetrahydroxybiphenyl **40**,[7] ethanedioldimesylate **39**,[22] ethylbridged tetrahydroxybiphenyl **41**[18] and the MOM derivative of 2-iodo-3-nitrophenol **46**.[19]

5-(3-(4-Dimethylaminophenyl)-1-oxo-prop-2-ene)-2,2′-biphenyldioxacyclodecyne (16)



This compound is novel. A solution of **15** (100 mg, 0.360 mmol, 1.0 eq.), 4-dimethylaminobenzaldehyde (58 mg, 0.39 mmol, 1.1 eq.) and NaOH (43 mg, 1.1 mmol, 3.0 eq.) in EtOH (2 mL) was stirred at room temperature for 24 h. H_2O (20 mL) was added and the product was extracted with EtOAc (3 × 20 mL). The combined organic extracts were dried over $MgSO_4$ and concentrated. Purification by column chromatography (eluted with 50% EtOAc/hexane) gave the pure product as a yellow solid (96 mg, 0.23 mmol, 65%). R_f = 0.38 (1 : 1 EtOAc/hexane); (found (ESI)) 432.1563 $C_{27}H_{23}NNaO_3$ requires 432.1570; ν_{max} 2916, 2865, 1641, 1565, 1526, 1180, 1170, 966, 802 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.08 (1 H, dd, J = 8.4, 2.1 Hz, Ar***H***) 7.89 (1 H, d, J = 2.1 Hz, Ar***H***) 7.80 (1 H, d, J = 15.4 Hz, COCHC***H***Ph) 7.53 (2 H, d, J = 8.9 Hz, Ar***H***) 7.42 (1 H, ddd, J = 8.0, 6.5, 2.7 Hz, Ar***H***) 7.32 (1 H, d, J = 15.4 Hz, COC***H***CHPh) 7.28 (1 H, d, J = 8.4 Hz, Ar***H***) 7.19–7.25 (3 H, m, Ar***H***) 6.67 (2 H, d, J = 8.9 Hz, Ar***H***) 4.52–4.62 (2 H, m, OC***H***$_a$H$_b$) 4.32–4.44 (2 H, m, OCH$_a$***H***$_b$) 3.03 (6 H, s, NC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 189.5, 158.0, 154.5, 152.0, 145.7, 135.8, 135.3, 135.1, 132.5, 131.9, 130.5, 129.5, 129.4, 124.3, 122.9, 122.7, 122.7, 116.7, 111.8, 87.2, 86.3, 63.7, 63.5, 40.1 ppm; m/z (ESI) 410.2 $[M + H]^+$, 432.2 $[M + Na]^+$; Fluorescence (MeCN; λ_{ex} = 420 nm); λ_{em} = 536 nm; UV-Vis (MeCN) λ_{max} (ε/M^{-1} cm^{-1}): 413 (486 976) nm.

(*E*)-1-(1-Benzyl-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazol-8-yl)-3-(4-(dimethylamino)phenyl)prop-2-en-1-one (17)

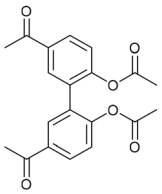


This compound is novel. A solution of **16** (10 mg, 0.024 mmol, 1 eq.) and benzyl azide (3.2 mg, 0.024 mmol, 1 eq.) in $CDCl_3$ (0.5 mL) was monitored by NMR until completion of the reaction. The solvent was removed under vacuum and the crude material was purified by column chromatography (eluted with 50% EtOAc/hexane) to give the pure product as two inseparable isomers in a 1 : 1 ratio as an orange solid (11.8 mg, 0.022 mmol, 91%).

R_f = 0.16 (1 : 1 EtOAc/hexane); (found (ESI)) 543.2379 $C_{34}H_{31}N_4O_3$ requires 543.2391; ν_{max} 2923, 1570, 1521, 1495, 1443, 1433, 1332, 1261, 1180, 1167, 1107, 810, 750 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.00 (0.5 H, dd, J = 8.5, 2.1 Hz, PhC***H***CHCO) 7.92 (1 H, dd, J = 6.6, 2.1 Hz, Ar***H***) 7.82 (0.5 H, dd, J = 8.5, 2.1 Hz, PhC***H***CHCO) 7.78 (1 H, d, J = 15.4 Hz, PhC***H***CHCO) 7.53 (2 H, d, J = 8.8 Hz, Ar***H***), 7.38–7.42 (2 H, m, Ar***H***), 7.32–7.37 (3 H, m, Ar***H***), 7.26–7.05 (7 H, m, Ar***H***), 6.78 (0.5 H, d, J = 7.9 Hz, PhCHC***H***CO) 6.68 (2 H, d, J = 8.8 Hz, Ar***H***) 6.59 (0.5 H, d, J = 8.5 Hz, PhCHC***H***CO) 5.81 (0.5 H, d, J = 16.0 Hz, C***H***$_e$H$_f$) 5.77 (0.5 H, d, J = 16.0 Hz, C***H***$_e$H$_f$) 5.63 (0.5 H, d, J = 13.7 Hz, CH$_c$***H***$_d$) 5.35–5.45 (2.5 H, m, CH$_c$***H***$_f$ + C***H***$_c$H$_d$ + CH$_c$***H***$_d$) 5.22 (1.5 H, m, 2 × OC***H***$_a$H$_b$ + OCH$_a$***H***$_b$) 5.05 (0.5 H, d, J = 13.0 Hz, OCH$_a$***H***$_b$) 3.03 (6 H, s, NC***H***$_3$); (125 MHz, $CDCl_3$) 188.8, 188.8, 159.4, 158.8, 156.9, 156.0, 152.0, 151.9, 145.5, 145.2, 144.8, 144.5, 134.6, 134.4, 134.1, 133.1, 132.3, 132.1, 131.2, 131.2, 130.7, 130.5, 130.4, 130.3, 130.1, 129.7, 129.6, 129.6, 129.4, 129.3, 129.2, 128.9, 128.9, 128.7, 127.2, 127.1, 123.6, 122.8, 122.7, 122.4, 116.5, 116.4 116.0, 114.7, 114.4, 113.5, 111.8, 63.6, 62.9, 61.0, 60.4, 52.6, 52.4, 40.1 ppm; m/z (ESI) 543.2 [M +

H]$^+$, 565.2 [M + Na]$^+$; fluorescence (MeCN; λ_{ex} = 416 nm); λ_{em} = 530 nm; UV-Vis (MeCN) λ_{max} (ε/M^{-1} cm^{-1}): 410 (70 000) nm.

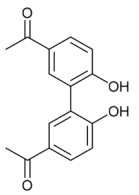
5,5′-Diacetyl-[1,1′-biphenyl]-2,2′-diyl diacetate (18)



This compound is novel. A solution of $AlCl_3$ (6.00 g, 45 mmol, 8.3 eq.) in DCM (4 mL) was cooled to 0 °C. Acetyl chloride (4.40 g, 56.0 mmol, 10.4 eq.) was added to the solution and the reaction mixture was stirred for 30 min. A solution of 2,2′-biphenol (1.00 g, 5.4 mmol, 1.0 eq.) in DCM (10 mL) was added to the reaction mixture at 0 °C and the mixture was stirred for a further 30 min. The reaction was then refluxed until completion, at which point H_2O (30 mL) was added dropwise to quench. The product was extracted with EtOAc (3 × 30 mL) and the combined organic extracts were dried over $MgSO_4$, which was removed by filtration, and concentrated under vacuum to give the crude product. Purification by column chromatography gave the pure product as a white solid (892 mg, 2.5 mmol, 47%).

R_f = 0.60 (1 : 1 EtOAc/Pet Ether); mp = 205–209 °C; (found (ESI)) 377.0982 $C_{20}H_{18}NaO_6$ requires 377.0996; ν_{max} 1740, 1683, 1600, 1355, 1191, 910, 619 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.07 (2 H, dd, J = 8.5, 2.1 Hz, Ar***H***) 7.97 (2 H, d, J = 2.1 Hz, Ar***H***) 7.33 (2 H, d, J = 8.5 Hz, Ar***H***) 2.65 (6 H, s, COC***H***$_3$) 2.09 (6 H, s, OCOC***H***$_3$) ppm; δ_C (125 MHz, $CDCl_3$) 196.5, 168.6, 151.7, 135.0, 131.6, 130.0, 129.6, 123.0, 26.7, 20.7 ppm; m/z (ESI) 377.1 [M + Na]$^+$.

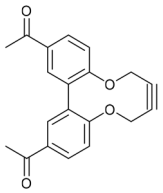
1,1′-(6,6′-Dihydroxy-[1,1′-biphenyl]-3,3′-diyl)bis(ethan-1-one) (19)



This compound is novel. A solution of compound **18** (848 mg, 2.25 mmol, 1 eq.) and LiOH (302 mg, 12.6 mmol, 5.6 eq.) in MeOH/H_2O 1 : 1 (10 mL) was refluxed for 2 h. The mixture was then cooled to room temperature before 2 M HCl (20 mL) was added. The product was then extracted with EtOAc (3 × 20 mL), the combined organic extracts were dried over $MgSO_4$ and concentrated and recrystalised in MeOH to give the pure product as a white solid (472 mg, 1.76 mmol, 70%).

R_f = 0.2 (1 : 1 EtOAc/Pet Ether); mp = 177–181 °C; (found (ESI)) 293.0782 $C_{16}H_{14}NaO_4$ requires 293.0784; ν_{max} 3222, 1651, 1579, 1383, 1354, 1255, 818, 583 cm^{-1}; δ_H (500 MHz, DMSO-d_6) 10.30 (2 H, s, O***H***) 7.84 (2 H, dd, J = 8.5, 2.3 Hz, Ar***H***) 7.78 (2 H, d, J = 2.3 Hz, Ar***H***) 7.00 (2 H, d, J = 8.5 Hz, Ar***H***) 2.50 (6 H, s, OC***H***$_3$); δ_C (125 MHz, d^6-DMSO) 196.2, 159.6, 132.3, 129.6, 128.3, 124.9, 115.4, 26.3 ppm; m/z (ESI) 293.1 [M + Na]$^+$.

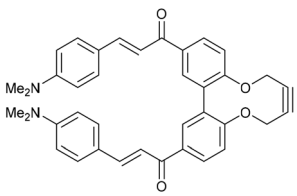
5,5′-Diacetyl-2,2′-biphenyldioxacyclodecyne (21)



This compound is novel. To a solution of Cs_2CO_3 (1.33 g, 4.1 mmol, 2.2 eq.) in MeCN (43 mL), at 60 °C, was added a solution of ditosylate **20** (733 mg, 1.86 mmol, 1 eq.) and compound **19** (500 mg, 1.86 mmol, 1 eq.) in MeCN (8.7 mL) over 6 h. The mixture was stirred for a further 12 h before being cooled to room temperature and the solvent removed under vacuum. The residue was dissolved in H_2O (30 mL) and the product was extracted with EtOAc (3 × 30 mL). The combined organic extracts were dried over $MgSO_4$ and concentrated to give the crude product, which was purified by column chromatography (eluted with 50% EtOAc/hexane) to give the crude product as a white solid (233 mg, 0.73 mmol, 39%).

R_f = 0.42 (1 : 1 EtOAc/Pet. Ether); mp = 188–189 °C; (found (ESI)) 343.0935 $C_{20}H_{16}NaO_4$ requires 343.0941; ν_{max} 3060, 2919, 1673, 1594, 1477, 1238, 1191, 956, 676 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.05 (2 H, dd, J = 8.5, 2.3 Hz, Ar***H***), 7.82 (2 H, d, J = 2.3 Hz, Ar***H***) 7.28 (2 H, d, J = 8.5 Hz, Ar***H***) 4.55–4.64 (2 H, m, OC***H***$_a$H$_b$) 4.36–4.44 (2 H, m, OCH$_a$***H***$_b$) 2.60 (6 H, s, COC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 197.1, 158.7, 135.2, 133.4, 132.6, 129.7, 123.1, 86.7, 63.7, 26.7 ppm; m/z (ESI) 343.1 [M + Na]$^+$.

5,5′-Bis(3-(4-dimethylaminophenyl)-1-oxo-prop-2-ene)-2,2′-biphenyldioxacyclodecyne (22)

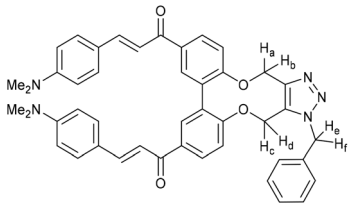


This compound is novel. A solution of compound **21** (99.2 mg, 0.310 mmol, 1.0 eq.), 4-dimethylamino benzaldehyde (101 mg, 0.680 mmol, 2.2 eq.) and NaOH (74 mg, 1.9 mmol, 6 eq.) in EtOH (2 mL) was stirred at room temperature for 12 h. H_2O (20 mL) was added, and the product was extracted with EtOAc (3 × 20 mL). The combined organic extracts were then dried over $MgSO_4$ and concentrated. Purification by column chromatography (eluted with 50% EtOAc/hexane) gave the pure product as an orange solid (59 mg, 0.10 mmol, 32%).

R_f = 0.26 (1 : 1 EtOAc/hexane); (found (ESI)) 605.2396 $C_{38}H_{34}N_2NaO_4$ requires 605.2411; ν_{max} 2906, 2854, 1647, 1569, 1518, 1331, 1163, 1109, 810, 747 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.12 (2 H, dd, J = 8.4, 2.1 Hz, Ar***H***), 7.91 (2 H, d, J = 2.1 Hz, Ar***H***), 7.81 (2 H, d, J = 15.4 Hz, COCHC***H***Ph), 7.55 (4 H, d, J = 8.9 Hz, Ar***H***), 7.34 (2 H, d, J = 15.4 Hz, COC***H***CHPh), 7.29–7.34 (2 H, m, Ar***H***), 6.69 (4 H, d, J = 8.9 Hz, Ar***H***), 4.52–4.68 (2 H, m, OC***H***$_a$H$_b$), 4.33–4.49 (2 H, m, OCH$_a$***H***$_b$), 3.09 (12 H, s, NC***H***$_3$)

ppm; δ_C (125 MHz, $CDCl_3$) 189.4, 158.0, 152.0, 145.8, 135.2, 132.4, 130.5, 129.8, 123.0, 122.7, 116.7, 111.8, 86.8, 63.7, 40.1 ppm; *m*/*z* (ESI) 583.3 $[M + Na]^+$, 605.2 $[M + Na]^+$; fluorescence (MeCN; λ_{ex} = 420 nm); λ_{em} = 536 nm; UV-Vis (MeCN) λ_{max} (ε/M^{-1} cm^{-1}): 416 (148 000) nm.

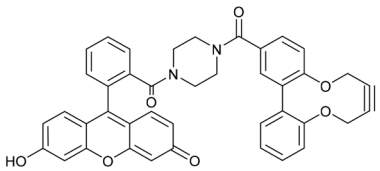
(2*E*,2′*E*)-1,1′-(1-Benzyl-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazole-8,11-diyl)bis(3-(4-(dimethylamino)phenyl)prop-2-en-1-one) (23)



This compound is novel. A solution of compound **22** (10 mg, 0.017 mmol, 1 eq.) and benzyl azide (2.3 mg, 0.017 mg, 1 eq.) in $CDCl_3$ (0.5 mL) was monitored by NMR until completion. The solvent was removed under vacuum and the crude material was purified by column chromatography (eluted with EtOAc) to give the pure product as a red solid (7.0 mg, 0.010 mmol, 58%).

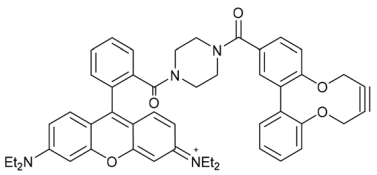
R_f = 0.20 (1 : 1 EtOAc/hexane); (found (ESI)) 716.3214 $C_{45}H_{42}N_5O_4$ requires 716.3231; ν_{max} 1575, 1521, 1334, 1167, 1117, 1026, 979, 809 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.06 (1 H, dd, *J* = 8.5, 2.1 Hz, Ar***H***), 8.02 (1 H, d, *J* = 2.1 Hz, Ar***H***) 8.00 (1 H, d, *J* = 2.1 Hz, Ar***H***), 7.94–7.99 (1 H, m, Ar***H***) 7.90–7.95 (1 H, m, Ar***H***) 7.78–7.84 (2 H, m, PhC***H***CHCO) 7.55 (4 H, m, *J* = 8.7, 3.5 Hz, Ar***H***) 7.40 (2 H, m, PhCHC***H***CO) 7.33–7.38 (3 H, m, Ar***H***) 7.20 (3 H, m, *J* = 6.1, 2.9 Hz, Ar***H***) 6.68 (4 H, m, *J* = 8.9 Hz, Ar***H***) 5.82 (1 H, d, *J* = 15.7 Hz, C***H***$_e$H$_f$) 5.58 (1 H, d, *J* = 13.4 Hz, OC***H***$_c$H$_d$) 5.39–5.47 (2 H, m, OCH$_c$***H***$_d$ + CH$_e$***H***$_f$) 5.29 (1 H, d, *J* = 13.4 Hz, OC***H***$_a$H$_b$) 5.15 (1 H, d, *J* = 13.4 Hz, OCH$_a$***H***$_b$) 3.03 (12 H, s, NC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 188.7, 159.7, 159.1, 152.0, 152.0, 145.7, 145.4, 144.5, 134.4, 134.4, 133.4, 132.0, 131.1, 131.1, 130.5, 130.4, 130.1, 130.0, 129.8, 129.4, 129.0, 128.9, 127.2, 122.7, 122.7, 116.4, 116.3, 115.6, 114.0, 111.8, 63.5, 60.8, 52.7, 40.1 ppm; *m*/*z* (ESI) 716.3 $[M + H]^+$, 738.3 $[M + Na]^+$; fluorescence (MeCN; λ_{ex} = 418 nm); λ_{em} = 532 nm; UV-Vis (MeCN) λ_{max} (ε/M^{-1} cm^{-1}): 409 (199 900) nm.

Fluorescein amide 2,2′-biphenyldioxacyclodecyne (24)



This compound is novel. 6-Hydroxy-9-(2-(piperazine-1-carbonyl)phenyl)-3*H*-xanthen-3-one **26**, synthesised as previously described,[11] (78.0 mg, 0.195 mmol) and acid alkyne **28** (54.6 mg, 0.195 mmol) were dissolved in anhydrous DMF (2 mL). DMAP (59.5 mg, 0.287 mmol) and EDCI (74.9 mg, 0.390 mmol) were added and the reaction stirred under N_2 at room temperature for 18 hours. H_2O (10 mL) was added, extracted with CH_2Cl_2/IPA (4 : 1) (3 × 10 mL) and the combined organic layers dried over $MgSO_4$. The crude mixture was purified by column chromatography (SiO_2; CH_2Cl_2/MeOH; 100 : 0 → 90 : 10) to afford the compound as an orange solid (74 mg, 0.111 mmol, 57%). R_f = 0.60 (4 : 1 DCM/MeOH); mp 187–198 (dec) °C; (found (ESI) $[M + H]^+$, 663.2125. $C_{41}H_{31}N_2O_7$ requires $[M + H]^+$, 663.2126); ν_{max} 1591, 1417, 1379, 1195, 1001, 964 and 847 cm^{-1}; δ_H (500 MHz, CD_3OD) 7.90 (2 H, s, Ar***H***), 7.80–7.61 (3 H, m, Ar***H***), 7.53–7.46 (1 H, m, Ar***H***), 7.45–7.35 (2 H, m, Ar***H***), 7.29 (1 H, d, *J* = 8.3, Ar***H***), 7.20–7.12 (4 H, m, Ar***H***), 6.76–6.68 (3 H, m, Ar***H***), 4.57–4.43 (2 H, m, OC***H***$_a$H$_b$), 4.43–4.30 (2 H, m, OCH$_a$***H***$_b$), 3.45 (8 H, br. s, NCH_2); δ_C (126 MHz, CD_3OD) 172.2, 169.8, 157.9, 156.0, 153.7, 137.7, 136.5, 136.4, 132.9, 132.8, 132.6, 132.2, 131.8, 131.6, 131.2, 131.1, 130.6, 129.4, 128.9, 125.1, 124.5, 123.8, 104.4, 88.0, 87.3, 64.4, 64.3, 64.3, 64.1; *m*/*z* (ESI) 663 (M^+ + H, 30%) and 685 (M^+ + Na, 30); UV-Vis (MeCN) lmax (ε/M^{-1} cm^{-1}): 487 (13 200), 457 (18 800), 430 (16 700), 353 (9200), 227 (59 000) nm; fluorescence (MeCN; λ_{ex} = 531 nm); λ_{em} 545 nm.

Rhodamine amide 2,2′-biphenyldioxacyclodecyne (25)

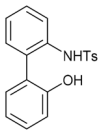


This compound is novel. *N*-(6-(Diethylamino)-9-(2-(piperazine-1-carbonyl)phenyl)-3*H*-xanthen-3-ylidene)-*N*-ethylethanaminium **27**, synthesised as previously described,[12] (100 mg, 0.195 mmol) and acid alkyne **28** (54.6 mg, 0.195 mmol) were dissolved in anhydrous DMF (2 mL). DMAP (59.5 mg, 0.287 mmol) and EDCI (74.9 mg, 0.390 mmol) were added and the reaction stirred under N_2 at room temperature for 18 hours. H_2O (10 mL) was added, extracted with CH_2Cl_2/IPA (4 : 1) (3 × 10 mL) and the combined organic layers dried over $MgSO_4$. The crude mixture was purified by column chromatography (SiO_2; CH_2Cl_2/MeOH; 100 : 0 → 90 : 10) to afford the compound as a dark purple solid (36 mg, 0.049 mmol, 24%).

R_f = 0.70 (4 : 1 DCM/MeOH); mp 169–170 (dec) °C; (found (ESI) $[M + H]^+$, 773.3692. $C_{49}H_{49}N_4O_5$ requires $[M + H]^+$, 773.3697); ν_{max} 1586, 1334, 1244, 1178, 1122, 1070, 1002 and 759 cm^{-1}; δ_H (500 MHz, CD_3OD) 7.90 (2 H, s, Ar***H***), 7.81–7.74 (2 H, m, Ar***H***), 7.70 (1 H, d, *J* = 6.8, Ar***H***), 7.54–7.49 (1 H, m, Ar***H***), 7.46–7.39 (2 H, m, Ar***H***), 7.32–7.25 (3 H, m, Ar***H***), 7.23–7.12 (5 H, m, Ar***H***), 7.05 (2 H, d, *J* = 9.7, Ar***H***), 6.95 (2 H, t, *J* = 3.1, Ar***H***), 4.55–4.43 (2 H, m, OC***H***$_a$H$_b$), 4.40–4.33 (2 H, m, OCH$_a$***H***$_b$), 3.66 (8 H, app. pent., *J* = 7.3, NC***H***$_2$CH$_3$), 3.58–3.37 (8 H, m, NC***H***$_2$), 1.29 (12 H, app. q, *J* = 7.1, NCH_2C***H***$_3$); δ_C (126 MHz, CD_3OD) 172.2, 169.6, 159.3, 157.9, 157.2, 157.1, 156.0, 137.7, 136.5, 136.4, 133.2, 132.8, 132.4, 132.2, 131.8, 131.6, 131.2, 130.7, 129.4, 128.9, 125.2, 124.6, 123.8, 114.9, 97.4, 88.0, 87.4, 64.4, 64.3, 46.9, 12.8. *m*/*z* (ESI) 773 (M^+ + H, 100%); UV-Vis (MeCN) lmax (ε/M^{-1} cm^{-1}): 558 (57 400), 522

(35 100), 352 (24 700), 522 (36 200), 250 (59 800) nm; fluorescence (MeCN; λ_{ex} = 566 nm); λ_{em} 578 nm.

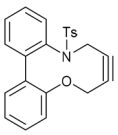
N-(2′-Hydroxy-[1,1′-biphenyl]-2-yl)-4-methylbenzenesulfonamide (31)



This compound is novel. A solution of 2-hydroxyphenyl boronic acid (175 mg, 1.27 mmol, 1.3 eq.), compound **30** (365 mg, 0.981 mmol, 1.0 eq.), K_2CO_3 (270 mg, 1.96 mmol, 2.0 eq.) and $[PdCl_2(PPh_3)_2]$ (68 mg, 0.098 mmol, 0.1 eq.) in 5 : 1 DMF-H_2O (9 mL) was stirred at 80 °C for 12 h. The reaction was cooled to room temperature and then diluted with H_2O (20 mL). The product was then extracted with EtOAc (3 × 20 mL) and the combined organic extracts were dried over $MgSO_4$ before being concentrated. The crude product was then subjected to column chromatography (graduated eluent: 9 : 1 Hex/EtOAc–7 : 3 Hex/EtOAc) to give the pure product as a white solid (177 mg, 0.523 mmol, 53%).

R_f = 0.63 (2 : 3 EtOAc/DCM); mp = 142–146 °C; (found (ESI) $[M + Na]^+$ 362.0821 $C_{16}H_{17}NNaO_3S$ requires 362.0821); ν_{max} 3422, 3321, 3231, 1596, 1484, 1163, 700 and 527 cm^{-1}; δ_H ($CDCl_3$, 500 MHz) 7.74 (1 H, d, *J* = 8.0 Hz, Ar***H***), 7.42 (1 H, t, *J* = 8.1 Hz, Ar***H***), 7.36 (2 H, d, *J* = 8.1 Hz, 2 × Ar***H***), 7.22–7.28 (1 H, m, Ar***H***), 7.15 (1 H, d, *J* = 7.6 Hz, Ar***H***), 7.09 (2 H, d, *J* = 8.1 Hz, 2 × Ar***H***), 6.94 (2 H, d, *J* = 8.0 Hz, Ar***H***), 6.85 (1 H, t, *J* = 7.5 Hz, Ar***H***) 6.57 (1 H, d, *J* = 7.5 Hz, Ar***H***) 5.07–5.12 (1 H, br. s, N***H***) 2.39 (3 H, s, ArCH_3) ppm; δ_C ($CDCl_3$, 125 MHz) 151.8, 143.6, 135.9, 134.6, 131.0, 130.0, 130.0, 129.5, 129.3, 126.9, 126.0, 123.8, 121.3, 116.0, 21.5 ppm; *m*/*z* (ESI) 362.2 $[M + Na]^+$.

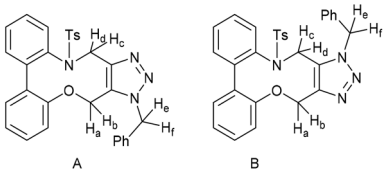
Compound 32



This compound is novel. A solution of **20** (116 mg, 0.294 mmol, 1.0 eq.), **30** (99.3 mg, 0.294 mmol, 1.0 eq.) and Cs_2CO_3 (383 mg, 1.18 mmol, 4.0 eq.) in CH_3CN (15 mL) was stirred at room temperature for 2 weeks. The solvent was then removed under vacuum and the residue taken up in water (20 mL) the product was extracted with EtOAc (3 × 20 mL). The combined organic extracts were dried over $MgSO_4$ and concentrated to give the crude product. This was then subjected to column chromatography (eluent: 1 : 1 Hex/EtOAc) to give the pure product as a white solid (46.6 mg, 0.12 mmol, 40%).

R_f = 0.82 (1 : 1 EtOAc/hexane); mp = 122–128 °C; (found (ESI) $[M + Na]^+$ 412.0981 $C_{23}H_{19}NNaO_3S$ requires 412.0978); ν_{max} 3059, 2922, 1596, 1501, 1452, 1106, 1056, 966, 688 and 575 cm^{-1}; δ_H ($CDCl_3$, 500 MHz) 7.72 (1 H, d, *J* = 7.5 Hz, Ar***H***), 7.65 (2 H, d, *J* = 8.2 Hz, 2 × Ar***H***), 7.28–7.37 (2 H, m, 2 × Ar***H***), 7.23–7.28 (3 H, m, 3 × Ar***H***), 7.17–7.23 (2 H, m, 2 × Ar***H***), 7.08 (1 H, d, *J* = 8.1 Hz, Ar***H***), 6.88 (1 H, d, *J* = 8.0 Hz, Ar***H***), 4.35 (1 H, d, *J* = 15.0 Hz, $CH_a\boldsymbol{H}_b$) 4.18–4.25 (2 H, m, C$\boldsymbol{H}_2$) 3.50 (1H, d, *J* = 15.0 Hz, C$\boldsymbol{H}_a$H$_b$), 2.39 (3 H, s, Ar–C$\boldsymbol{H}_3$) ppm; δ_C ($CDCl_3$, 125 MHz) 154.3, 144.1, 142.1, 136.9, 135.8, 135.6, 133.1, 132.4, 129.8, 129.5, 128.4, 128.2, 128.2, 127.6, 124.3, 122.3, 84.9, 84.3, 63.0, 43.5, 21.6 ppm; *m*/*z* (ESI) 362.2 $[M + Na]^+$.

(3-Benzyl-14-tosyl-3,4,14,15-tetrahydrodibenzo[*b*,*d*][1,2,3]triazolo[4,5-*h*][1,6]oxazecine) and 32B (1-benzyl-14-tosyl-1,4,14,15-tetrahydrodibenzo-[*b*,*d*][1,2,3]triazolo[4,5-*h*][1,6]oxazecine) (32A)

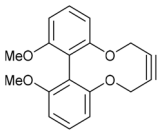


These compounds are novel. To a solution of compound **31** (20 mg, 0.051 mmol, 1.0 eq.) in $CDCl_3$ (0.5 mL) was added benzyl azide (6.8 mg, 6.4 μL, 0.051 mmol, 1 eq.). The mixture was left undisturbed at room temperature and monitored by proton NMR until full conversion was observed. The chloroform was removed by evaporation and the residue purified by column chromatography (eluted with DCM) to provide the product as two isolable isomers A (white solid) (6.8 mg, 0.013 mmol, 26%) and B (white solid) (5.4 mg, 0.010 mmol, 20%). The stereochemical assignments are arbitrary.

A; R_f = 0.28 (DCM); (found (ESI) $[M + Na]^+$ 545.1615 $C_{30}H_{26}N_4NaO_3S$ requires 545.1618); ν_{max} 3028, 2925, 2854, 1596, 1478, 1439, 1353, 1331, 1159 and 738 cm^{-1}; δ_H ($CDCl_3$, 500 MHz) 7.28–7.44 (7 H, m, Ar***H***), 7.13–7.20 (3 H, m, Ar***H***), 7.03 (2 H, d, *J* = 8.1 Hz, Ar***H***), 6.98–7.02 (1 H, m, Ar***H***), 6.91 (2 H, d, *J* = 8.1 Hz, Ar***H***), 6.88–6.90 (2 H, m, Ar***H***), 5.76 (1 H, d, *J* = 15.7 Hz, C$\boldsymbol{H}_e$H$_f$), 5.64 (1 H, d, *J* = 15.7 Hz, CH$_e\boldsymbol{H}_f$), 5.54 (1 H, d, *J* = 14.5 Hz, C$\boldsymbol{H}_c$CH$_d$), 5.14 (1H, d, *J* = 14.5 Hz, CH$_c\boldsymbol{H}_d$), 4.91 (1 H, d, *J* = 15.7 Hz C$\boldsymbol{H}_a$H$_b$), 4.47 (1 H, d, *J* = 15.7 Hz, CH$_a\boldsymbol{H}_b$), 2.39 (3 H, s, Ar–C$\boldsymbol{H}_3$) ppm; δ_C ($CDCl_3$, 125 MHz) 154.4, 144.6, 143.9, 140.2, 140.0, 135.5, 134.8, 131.3, 131.2, 131.0, 130.1, 129.3, 129.2, 129.1, 128.7, 128.6, 128.5, 128.4, 127.8, 127.1, 121.8, 112.7, 62.0, 52.4, 44.4, 21.5 ppm; *m*/*z* (ESI) 523.3 $[M + H]^+$ 545.2 $[M + Na]^+$.

B; R_f = 0.1 (DCM); (found (ESI) $[M + Na]^+$ 545.1615 $C_{30}H_{26}N_4NaO_3S$ requires 545.1618); ν_{max} 3028, 2925, 2854, 1596, 1478, 1439, 1353, 1331, 1159 and 738 cm^{-1}; δ_H ($CDCl_3$, 500 MHz) 7.33–7.40 (5 H, m, Ar***H***), 7.28–7.31 (2 H, m, Ar***H***),7.19–7.21 (2H, m, Ar***H***), 7.13–7.18 (4 H, m, Ar***H***), 7.03–7.08 (3 H, m, Ar***H***), 6.45 (1 H, m, Ar***H***), 5.65 (1 H, d, *J* = 15.7 Hz, C$\boldsymbol{H}_c$H$_d$), 5.31 (1 H, d, *J* = 15.7 Hz, CH$_c\boldsymbol{H}_d$) 5.05 (1 H, d, *J* = 13.7 Hz, C$\boldsymbol{H}_e$H$_f$) 5.04 (1H, d, *J* = 14.8 Hz, C$\boldsymbol{H}_a$H$_b$), 4.96 (1 H, d, *J* = 13.7 Hz CH$_e\boldsymbol{H}_f$) 4.59 (1 H, d, *J* = 14.8 Hz, CH$_a\boldsymbol{H}_b$) 2.43 (3 H, s, Ar–C$\boldsymbol{H}_3$) ppm; δ_C ($CDCl_3$, 125 MHz) 155.4, 143.7, 142.4, 140.6, 139.4, 134.4, 134.0, 133.0, 132.0, 131.7, 131.1, 129.2, 129.2, 128.8, 128.7, 128.6, 128.4, 128.3, 127.2, 126.7, 123.0, 114.8, 60.0, 52.4, 47.2, 21.6 ppm; *m*/*z* (ESI) 523.3 $[M + H]^+$ 545.2 $[M + Na]^+$.

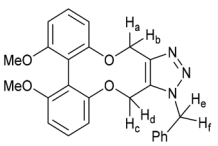
6,6-Dimethoxy-2,2′-biphenyldioxacyclodecyne (37)



This compound is novel. To a stirring solution of compound **20** (145 mg, 0.368 mmol, 1 eq.) and compound **36**[15] (90 mg, 0.37 mmol, 1 eq.) in MeCN (20 mL) was added Cs_2CO_3 (475 mg, 1.46 mmol, 4 eq.). The mixture was stirred for 10 days at 50 °C and monitored by TLC. Solvents were removed under vacuum and residue was dissolved in H_2O (30 mL). The aqueous layer was then extracted with EtOAc (3 × 30 mL) and the combined organic extracts were dried over $MgSO_4$, which was removed by filtration. The solvents were removed under vacuum to give the crude product. The crude product was then purified by column chromatography (eluted with 0–25% EtOAc/heptane) to give the pure product as a white solid (10 mg, 0.034 mmol, 10%).

Mp = 134–136 °C; (found (ESI)) 297.1180 $C_{18}H_{17}O_4$ requires 297.1127; δ_H (500 MHz, $CDCl_3$) 7.37 (2 H, t, J = 8.2 Hz, Ar***H***) 6.78–6.87 (4 H, m, Ar***H***) 4.48–4.58 (2 H, m, C***H***$_a$H$_b$) 4.35–4.44 (2 H, m, CH$_a$C***H***$_b$) 3.76 (6 H, s, OC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 158.1, 156.6, 129.3, 120.4, 114.0, 107.7, 87.2, 63.2, 56.3 ppm; m/z (ESI) 297.2 $[M + Na]^+$.

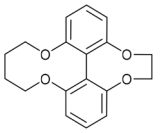
1-Benzyl-9,10-dimethoxy-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazole (38)



This compound is novel. To a solution of compound **37** (7.0 mg, 0.023 mmol, 1.0 eq.) in $CDCl_3$ (0.5 mL) was added benzyl azide (3.1 mg, 0.023 mmol, 1.0 eq.). The reaction was monitored by proton NMR. Once the reaction was complete the solvent was evaporated to give the crude product which was purified by column chromatography (eluted 0–25% EtOAc : heptane) to give the pure product as a white solid (7.5 mg, 0.017 mmol, 85%).

(Found (ESI)) 452.1580 $C_{25}H_{23}N_3NaO_4$ requires 452.1581; ν_{max} 2902, 2839, 1588, 1577, 1466, 1435, 1075 and 726 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.33–7.36 (3 H, m, Ar***H***) 7.23 (1 H, t, J = 8.3 Hz, Ar***H***) 7.18 (1 H, t, J = 8.4 Hz, Ar***H***) 7.12–7.16 (2 H, m, Ar***H***) 6.83 (1 H, d, J = 8.3 Hz, Ar***H***) 6.71 (1 H, d, J = 8.3 Hz, Ar***H***) 6.63 (1 H, d, J = 8.4 Hz, Ar***H***) 6.46 (1 H, d, J = 8.3 Hz, Ar***H***) 5.73 (1 H, d, J = 15.9 Hz, C***H***$_e$H$_f$) 5.48 (1 H, d, J = 13.7 Hz, C***H***$_c$H$_d$) 5.41 (1 H, d, J = 15.9 Hz, CH$_e$C***H***$_f$) 5.29 (1 H, d, J = 13.7 Hz, CH$_c$***H***$_d$) 5.15 (1 H, d, J = 13.2 Hz, C***H***$_a$H$_b$) 4.96 (1 H, d, J = 13.2 Hz, CH$_a$***H***$_b$) 3.75 (3 H, s, OC***H***$_3$) 3.72 (3 H, s, OC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 158.5, 158.3, 158.0, 156.2, 144.7, 134.8, 132.1, 129.0, 128.9, 128.5, 127.0, 115.4, 114.0, 113.8, 109.1, 107.7, 106.4, 105.2, 63.2, 61.3, 56.0, 55.9, 52.3 ppm; m/z (ESI) 452.2 $[M + Na]^+$.

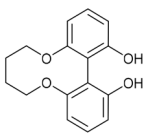
6,7,8,9-Tetrahydro-1,14-(epoxyethanooxy)dibenzo[*b*,*d*][1,6]dioxecine (42)



This compound is novel. A solution of compound **41** (357 mg, 1.47 mol, 1 eq.) and 1,4-dibromobutane (317 mg, 1.47 mmol, 1 eq.) in MeCN (15 mL) was added to a solution of C_2CO_3 (1.2 g, 3.7 mmol, 2.5 eq.) in MeCN (50 mL) at 60 °C over 5 h. The resulting solution was stirred for a further 12 h before the solvent was removed under vacuum. The residue was dissolved in H_2O (30 mL) and the product was extracted with EtOAc (3 × 30 mL). The combined organic extracts were dried over $MgSO_4$, which was removed by filtration, and then concentrated to give the crude product. Purification by column chromatography (eluted 25–100% EtOAc/hexane) gave the pure product as a white solid (352 mg, 1.18 mmol, 81%).

R_f = 0.53 (1 : 1 EtOAc/hexane); mp = 157–160 °C; (found (ESI)) 321.1094 $C_{18}H_{18}NaO_4$ requires 321.1097; ν_{max} 2928, 1590, 1563, 1257, 1221, 1065, 1023, 785 cm^{-1}; δ_H (500 MHz $CDCl_3$) 7.31 (2 H, t, J = 8.2 Hz, Ar***H***), 6.89 (2 H, d, J = 8.2 Hz, Ar***H***), 6.86 (2 H, d, J = 7.9 Hz, Ar***H***), 4.40 (2 H, d, J = 8.5 Hz, OC***H***$_2$), 4.38–4.45 (2 H, m, OC***H***$_2$), 4.25 (2 H, m, OC***H***$_2$), 4.12 (2 H, d, J = 8.5 Hz, OC***H***$_2$), 1.91–2.01 (2 H, m, C***H***$_2$), 1.79–1.88 (2 H, m, C***H***$_2$); δ_C (125 MHz, $CDCl_3$) 160.3, 157.9, 129.3, 118.9, 115.6, 111.6, 74.0, 70.7, 26.7; m/z (ESI) 321.0 $[M + Na]^+$.

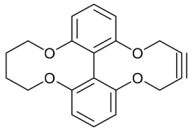
6,7,8,9-Tetrahydrodibenzo[*b*,*d*][1,6]dioxecine-1,14-diol (43)



This compound is known and fully characterised.[23] A solution of di-*tert*-butyl biphenyl (574 mg, 2.16 mmol, 8 eq.) in THF (11 mL) was cooled to 0 °C. To the solution, Li (13 mg, 1.89 mmol, 7 eq.) was added and the solution was stirred until dark blue in colour. This solution was then added to compound **42** (80 mg, 0.27 mmol, 1 eq.) and the mixture was stirred for 1 h. The mixture was then quenched with 1 M HCl (7 mL) and the product was extracted with EtOAc (3 × 10 mL). The combined organic extracts were then dried over $MgSO_4$, which was removed by filtration, and the solution concentrated to give the crude product. Purification by column chromatography (eluted with 25–50% EtOAc : hexane) gave the pure product as a white solid (48.4 mg, 0.17 mmol, 66%).

R_f = 0.46 (1 : 1 EtOAc/hexane); ν_{max} 3240, 2929, 1601, 1572, 1441, 1227, 1040, 775 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.28 (2 H, t, J = 8.3 Hz, Ar***H***), 6.73 (2 H, d, J = 8.2 Hz, Ar***H***), 6.72 (2 H, d, J = 8.2 Hz, Ar***H***), 5.15 (2 H, s, O***H***), 4.27–4.36 (2 H, m, OC***H***$_2$), 4.18–4.27 (2 H, m, OC***H***$_2$), 1.83–1.96 (2 H, m, C***H***$_2$), 1.70–1.83 (2 H, m, C***H***$_2$); δ_C (125 MHz, $CDCl_3$) 154.1, 130.1, 109.7, 108.6, 70.5, 26.0; m/z (ESI) 271.1 $[M - H]^-$, 295.1 $[M + H]^+$.

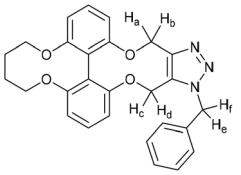
6,6′-Oxybutyloxy-linked-2,2′-biphenyldioxacyclodecyne (44)



This compound is novel. A solution of compound **43** (127 mg, 0.467 mmol, 1.0 eq.) and compound **20** (184 mg, 0.467, 1.0 eq.) in MeCN (2.2 mL) was added to a solution of Cs_2CO_3 (336 mg, 1.03 mmol, 2.2 eq.) in MeCN (11 mL) at 60 °C over 5 h. The mixture was stirred for a further 12 h before the solvent was removed. The remaining residue was dissolved in H_2O (20 mL) and extracted with EtOAc (3 × 20 mL). The combined organic extracts were dried over $MgSO_4$, which was removed by filtration, and concentrated. The crude product was then purified by column chromatography (eluted with 25% EtOAc/hexane) to give the pure product as a white solid (56 mg, 0.17 mmol, 36%).

R_f = 0.20 (1 : 3 EtOAc/hexane); mp = 166–168 °C; (found (ESI)) 345.1088 $C_{20}H_{18}NaO_4$ requires 345.1097; ν_{max} 2953, 2922, 2852, 1569, 1457, 1251, 1218, 1031, 739 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.37 (2 H, t, J = 8.2 Hz, Ar***H***), 7.00 (2 H, d, J = 8.2 Hz, Ar***H***), 6.94 (2 H, d, J = 8.2 Hz, Ar***H***), 4.54 (4 H, s, 2 × OC***H***$_2$), 4.27 (2 H, d, J = 11.9 Hz, OC***H***$_a$H$_b$), 3.80 (2 H, t, J = 11.9 Hz, CH$_a$***H***$_b$), 1.34–1.53 (4 H, m, 2 × C***H***$_2$); δ_C (125 MHz, $CDCl_3$) 156.7, 156.4, 129.2, 125.8, 115.8, 115.2, 88.4, 73.0, 62.4, 23.9; m/z (ESI) 345.1 $[M + Na]^+$.

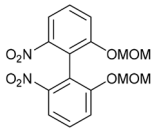
15-Benzyl-5,6,7,8,15,18-hexahydro-14*H*-4,9,13,19-tetraoxa-15,16,17-triazadibenzo[hi,qr]cyclopenta[*d*]decalene (45)



This compound is novel. To a solution of compound **44** (10 mg, 0.031 mmol, 1 eq.) in $CDCl_3$ (0.6 mL) was added benzyl azide (4.1 mg, 0.031 mmol, 1 eq.). The reaction was monitored by NMR until completion after which the solvent was removed and the residue was purified by column chromatography (eluted with 10% EtOAc/hexane) to give the pure product as a white solid (13 mg, 0.028 mmol, 95%).

R_f = 0.34; (1 : 1 EtOAc/hexane); (found (ESI)) 478.1735 $C_{27}H_{25}N_3NaO_4$ requires 478.1737; ν_{max} 2939, 1589, 1573, 1448, 1222, 1058, 716 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.33–7.39 (3 H, m, Ar***H***), 7.21 (1 H, t, J = 8.2 Hz, Ar***H***), 7.15–7.19 (2 H, m, Ar***H***), 7.12 (1 H, t, J = 8.2 Hz, Ar***H***), 6.82 (1 H, d, J = 8.2 Hz, Ar***H***), 6.78 (1 H, d, J = 8.2 Hz, Ar***H***), 6.72 (1 H, d, J = 8.2 Hz, Ar***H***), 6.39 (1 H, d, J = 8.2 Hz, Ar***H***), 5.73 (1 H, d, J = 15.7 Hz, CH$_f$***H***$_e$), 5.47 (1 H, d, J = 13.6 Hz, C***H***$_c$H$_d$), 5.38 (1 H, d, J = 15.7 Hz, C***H***$_f$H$_e$), 5.30 (1 H, d, J = 13.6 Hz, CH$_c$***H***$_d$), 5.17 (1 H, d, J = 13.3 Hz, C***H***$_a$H$_b$), 5.03 (1 H, d, J = 13.3 Hz, CH$_a$***H***$_b$), 4.16–4.35 (4 H, m, 2 × OC***H***$_2$), 1.70–1.94 (4 H, m, 2 × C***H***$_2$); δ_C (125 MHz, $CDCl_3$) 158.0, 157.8, 156.6, 144.9, 134.7, 132.3, 129.1, 128.7, 128.6, 128.5, 127.1, 116.9, 115.4, 110.9, 109.9, 108.9, 107.7, 70.8, 70.7, 62.8, 60.8, 52.3, 26.5; m/z (ESI) 478.2 $[M + Na]^+$.

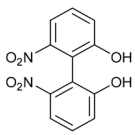
2,2′-Bis(methoxymethoxy)-6,6′-dinitro-1,1′-biphenyl (47)



This compound is novel. Precursor **46** was prepared by the published method.[19] It was purified by chromatography on silica gel using DCM as eluant. A solution of compound **46** (3.4 g, 11 mmol, 1 eq.)[19] in dry DMF (100 mL) was added to Cu powder (2.8 g, 44 mmol, 4 eq.) and the mixture heated to 100 °C overnight. The mixture was cooled to room temperature and filtered to remove solid residues. The solids in the filter paper were washed through and EtOAc (2 × 50 mL). To the combined filtrates, water (100 mL) was added and the product was extracted with EtOAc (3 × 100 mL). The combined organic extracts were washed with H_2O (3 × 100 mL) and brine (100 mL) before being dried over $MgSO_4$, which was removed by filtration, and concentrated. The crude product was then purified by column chromatography (eluted with 0–25% EtOAc/pet. Ether 40 : 60) to give the pure product as a yellow solid (1.56 g, 4.34 mmol, 79%).

R_f = 0.54 (1 : 1 EtOAc/hexane); mp = 104–106 °C; (found (ESI)) 387.0797 $C_{16}H_{16}N_2NaO_8$ requires 387.0799; ν_{max} 3075, 2959, 2831, 1580, 1531, 1456, 1253, 1205, 1083, 1001, 733 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.79–7.85 (2 H, m, Ar***H***), 7.45–7.52 (4 H, m, Ar***H***), 5.01–5.06 (4 H, m, OC***H***$_2$), 3.30 (6 H, s, OC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 154.6, 149.1, 129.4, 119.5, 119.2, 117.7, 94.9, 56.1; m/z (ESI) 387.1 $[M + Na]^+$. In another run a product was obtained in 92% yield without the need for purification by column chromatography.

6,6′-Dinitro-[1,1′-biphenyl]-2,2′-diol (48)

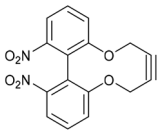


This compound is novel. To a solution of compound **47** (1.22 g, 3.35 mmol, 1 eq.) in MeOH (10 mL) was added conc. HCl (10 mL) dropwise. The mixture was stirred for 24 h and then H_2O (30 mL) was added. The product was extracted with EtOAc (3 × 30 mL) and the combined organic extracts were dried over $MgSO_4$, which was removed by filtration, and concentrated to give the crude product. Purification by column chromatography (eluted 50% EtOAc/pet.ether 40 : 60) gave the pure product (856 mg, 2.86 mmol, 81%).

R_f = 0.61 (1 : 1 EtOAc/Pet. Ether); mp = >200 °C (decomposition); (found (ESI)) 299.0269 $C_{12}H_8N_2NaO_6$ requires 299.0275; ν_{maz} 3311, 1510, 1331, 1288, 1160, 1002, 733 cm^{-1}; δ_H (500 MHz, CD_3CN) 7.64 (2 H, dd, J = 8.2, 0.8 Hz, Ar***H***), 7.62 (2 H, br. s, O***H***), 7.45 (2 H, t, J = 8.2 Hz, Ar***H***), 7.23 (2 H, dd, J =

8.2, 0.8 Hz, Ar***H***); δ_C (125 MHz, CD_3CN) 156.3, 151.1, 131.1, 121.8, 117.6, 117.3; *m*/*z* (ESI) 299.0 $[M + Na]^+$ In an alternative workup the final product was purified by recrystallisation from MeOH in 88% yield.

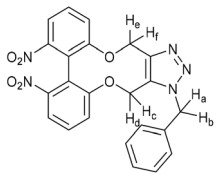
6,6′-Dinitro-2,2′-biphenyldioxacyclodecyne (49)



This compound is novel. A solution of compound **48** (856 mg, 3.10 mmol, 1.0 eq.) and compound **20** (1.22 g, 3.10 mmol, 1.0 eq.) in DMF (14 mL) was added to a solution of Cs_2CO_3 (2.50 g, 7.75 mmol, 2.5 eq.) in DMF (70 mL) at 60 °C over 5 h (using a syringe pump) and the resulting mixture was stirred for a further 12 h. The reaction mixture was then cooled to room temperature. Water (100 mL) was added and the product was extracted with EtOAc (3 × 100 mL). The combined organic extracts were then washed with H_2O (2 × 50 mL) and brine (50 mL) and dried over $MgSO_4$, which was removed by filtration, and concentrated. The water washings are required to remove DMF. The crude product was then purified by column chromatography (eluted with 50% EtOAc/pet. Ether 40 : 60) to give the pure product as a yellow solid (530 mg, 1.63 mmol, 53%).

R_f = 0.55 (1 : 1 EtOAc/hexane); mp 220–230 °C (dec); (found (ESI)) 349.0427 $C_{16}H_{10}N_2NaO_6$ requires 349.0431; ν_{max} 3088, 2926, 2852, 1519, 1349, 1337, 990, 1238, 1175, 990, 735 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 8.01 (2 H, d, *J* = 8.2 Hz, Ar***H***), 7.60 (2 H, t, *J* = 8.2 Hz, Ar***H***), 7.43 (2 H, d, *J* = 8.2 Hz, Ar***H***), 4.47–4.55 (2 H, m, OC***H***$_a$H$_b$), 4.37–4.47 (2 H, m, OCH$_a$***H***$_b$), ppm; δ_C (125 MHz, $CDCl_3$) 155.2, 149.2, 129.9, 126.9, 126.5, 121.2, 87.4, 63.6 ppm; *m*/*z* (ESI) 349 $[M + Na]^+$. In another procedure, chromatography on silica gel using DCM as eluant instead, and this gave a product in 39% yield.

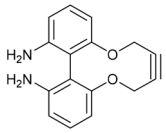
Benzyl azide cycloadduct of compound 49. 1-Benzyl-9,10-dinitro-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazole



This compound is novel. To a solution of compound **49** (9.0 mg, 0.027 mmol, 1 eq.) in $CDCl_3$ (0.5 mL) was added benzyl azide (3.6 mg, 0.027 mmol, 1 eq.). The reaction was monitored by proton NMR until completion and the solvent was removed under vacuum. The residue was purified by column chromatography (eluted with 50% EtOAc/hexane) to give the pure product as an oil (5.0 mg, 0.011 mmol, 40%).

R_f = 0.10 (1 : 1 EtOAc/hexane); (found (ESI)) 482.1067 $C_{23}H_{17}N_5NaO_6$ requires 482.1071; ν_{max} 2923, 2853, 1521, 1346, 1266, 1182, 1076, 902, 722 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.85–7.92 (1 H, m, Ar***H***), 7.76 (1 H, dd, *J* = 7.7, 1.6 Hz, Ar***H***), 7.39–7.46 (3 H, m, Ar***H***), 7.29–7.35 (3 H, m, Ar***H***), 7.09 (1 H, d, *J* = 8.1 Hz, Ar***H***), 7.01–7.07 (2 H, m, Ar***H***), 5.72 (1 H, d, *J* = 15.9 Hz, C***H***$_a$H$_b$), 5.58 (1 H, d, *J* = 13.9 Hz, OC***H***$_c$H$_d$), 5.44 (1 H, d, *J* = 15.9 Hz, CH$_a$***H***$_b$), 5.32 (1 H, d, *J* = 13.9 Hz, OCH$_c$***H***$_d$), 5.30 (1 H, d, *J* = 13.2 Hz, OC***H***$_e$H$_f$), 4.89 (1 H, d, *J* = 13.2 Hz, OCH$_e$***H***$_f$), ppm; (125 MHz, $CDCl_3$) 157.9, 155.2, 148.8, 148.6, 143.5, 134.3, 130.8, 129.8, 129.4, 129.2, 128.8, 126.8, 122.2, 122.1, 120.7, 120.5, 119.6, 118.0, 63.9, 62.9, 52.5 ppm; *m*/*z* (ESI) 482.1 $[M + Na]^+$.

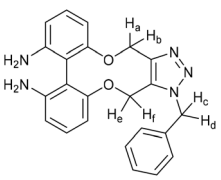
6,6′-Diamino-2,2′-biphenyldioxacyclodecyne (50)



This compound is novel. To a solution of compound **49** (238 mg, 0.730 mmol, 1 eq.) in 6 : 1 EtOH/H_2O (3.0 mL) was added NH_4Cl (39 mg, 0.73 mmol, 1 eq.) and Fe powder (204 mg, 3.65 mmol, 5 eq.). The mixture was heated to 70 °C and stirred for 1 h. The solution was allowed to cool to rt then the crude reaction was filtered through filter paper using MeOH (4 × 30 mL). Solvent was removed, then the residue was filtered through cotton wool using DCM (4 × 30 mL). Removal of the solvent gave **50** as an amorphous solid (179 mg, 0.673 mmol, 92%) without the need for column chromatography.

R_f = 0.38 (1 : 1 EtOAc/hexane); (found (ESI)) 289.0946 $C_{16}H_{14}N_2NaO_2$ requires 289.0947; ν_{max} 3465, 3360, 2960, 2914, 2864, 1611, 1565, 1461, 1302, 1248, 1116, 1020, 920, 729 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.21 (2 H, t, *J* = 8.1 Hz, Ar***H***), 6.65 (2 H, d, *J* = 8.1 Hz, Ar***H***), 6.61 (2 H, d, *J* = 8.1 Hz, Ar***H***), 4.54–4.61 (2 H, m, C***H***$_a$H$_b$), 4.40–4.47 (2 H, m, CH$_a$***H***$_b$), 3.63 (4 H, br. s., N***H***$_2$) ppm; δ_C (125 MHz, $CDCl_3$) 156.2, 146.2, 129.9, 116.5, 112.3, 111.8, 87.2, 63.3 ppm; *m*/*z* (ESI) 267.1 $[M + H]^+$, 289.1 $[M + Na]^+$.

Benzyl azide cycloadduct of compound 50. 1-Benzyl-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazole-9,10-diamine

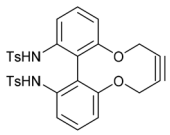


This compound is novel. To a solution of compound **50** (3.0 mg, 0.011 mmol, 1 eq.) in $CDCl_3$ (0.5 mL) was added benzyl azide (1.5 mg, 0.011 mmol, 1 eq.). The reaction was monitored by proton NMR until completion, at which point the solvent was removed under vacuum. The crude material was purified by column chromatography (eluted with 25%–

100% EtOAc/pet. Ether 40 : 60) to give the pure product as a white waxy solid (4 mg, 0.01 mmol, 91%).

R_f = 0.30 (1 : 1 EtOAc/hexane); (found (ESI)) 422.1590 $C_{23}H_{21}N_5NaO_2$ requires 422.1587; ν_{max} 3450, 3356, 2926, 2854, 1615, 1574, 1456, 1231, 1072, 909, 724 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.32–7.38 (3 H, m. Ar***H***), 7.11–7.16 (2 H, m, Ar***H***), 7.08 (1 H, t, *J* = 8.0 Hz, Ar***H***), 7.01 (1 H, t, *J* = 8.0 Hz, Ar***H***), 6.60 (1 H, d, *J* = 8.1 Hz, Ar***H***), 6.50 (1 H, d, *J* = 8.0 Hz, Ar***H***), 6.42 (1 H, d, *J* = 8.1 Hz, Ar***H***), 6.22 (1 H, d, *J* = 8.0 Hz, Ar***H***), 5.73 (1 H, d, *J* = 15.8 Hz, C***H***$_c$H$_d$), 5.43 (1 H, d, *J* = 13.6 Hz, C***H***$_a$H$_b$), 5.42 (1 H, d, *J* = 15.8 Hz, CH$_c$***H***$_d$), 5.28 (1 H, d, *J* = 13.6 Hz, CH$_a$***H***$_b$), 5.12 (1 H, d, *J* = 13.3 Hz, C***H***$_e$H$_f$), 4.94 (1 H, d, *J* = 13.3 Hz, CH$_e$***H***$_f$), 3.74 (4 H, br. s., N***H***$_2$); δ_C (125 MHz, $CDCl_3$) 158.7, 157.0, 145.6, 145.4, 144.8, 134.8, 132.1, 129.5, 129.1, 128.5, 127.0, 111.7, 110.6, 110.3, 109.6, 106.5, 105.4, 63.3, 61.2, 52.3 ppm; *m*/*z* (ESI) 398.1 $[M - H]^-$.

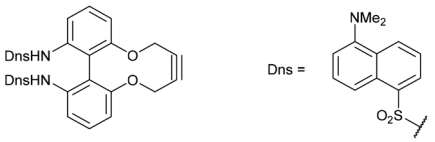
6,6′-Bis(tosylamido)-2,2′-biphenyldioxacyclodecyne (51)



This compound is novel. To a solution of compound **50** (61 mg, 0.23 mmol, 1 eq.) and toluenesulfonyl chloride (109 mg, 0.58 mmol, 2.5 eq.) in DCM (1.7 mL) was added pyridine (45 mg, 0.58 mmol, 2.5 eq.). The reaction was stirred for 4 h at room temperature. H_2O was added (10 mL) and the product was extracted with EtOAc (3 × 10 mL) and the combined organic extracts were dried over $MgSO_4$, which was removed by filtration, and concentrated. The product was a white solid (96 mg, 0.16 mmol, 73%) which was taken forward without further purification.

R_f = 0.51 (1 : 1 EtOAc/Pet. Ether); (found (ESI)) 597.1114 $C_{30}H_{26}N_2NaO_6S_2$ requires 597.1124; ν_{max} 3365, 2917, 1596, 1576, 1454, 1378, 1321, 1212, 1157, 1029, 1004, 935, 658, 536 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.51 (4 H, d, *J* = 8.1 Hz, Ar***H***), 7.51 (2 H, d, *J* = 7.9 Hz, Ar***H***), 7.38 (2 H, t, *J* = 8.1 Hz, Ar***H***), 7.19 (4 H, d, *J* = 8.1 Hz, Ar***H***), 6.90 (2 H, d, *J* = 7.9 Hz, Ar***H***), 5.95 (2 H, br. s, N***H***), 4.18–4.26 (2 H, mC***H***$_a$H$_b$), 4.08–4.18 (2 H, m, CH$_a$***H***$_b$); δ_C (125 MHz, $CDCl_3$) 154.4, 144.3, 136.4, 135.8, 131.0, 129.6, 127.3, 120.7, 118.6, 117.6, 86.7, 63.3, 21.6 ppm; *m*/*z* (ESI) 597.1 $[M + Na]^+$.

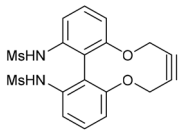
6,6′-Bis(dansylamido)-2,2′-biphenyldioxacyclodecyne (52)



This compound is novel. To a solution of compound **50** (50 mg, 0.19 mmol, 1 eq.) and dansyl chloride (128 mg, 0.48 mmol, 2.5 eq.) in DCM (1.4 mL) was added pyridine (37.5 mg, 0.48 mmol, 2.5 eq.). The reaction was stirred for 4 h before H_2O (10 mL) was added and the product was extracted with EtOAc (3 × 10 mL). The combined organic extracts were then dried over $MgSO_4$ and concentrated to give the crude product. Purification by column chromatography (eluted with 25–50% EtOAc/pet.ether 40 : 60) gave the pure product as a yellow solid (22.6 mg, 0.0310 mmol, 16%).

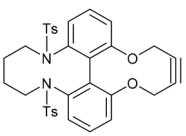
R_f = 0.37 (1 : 1 EtOAc/hexane); (found (ESI)) 733.2153 $C_{40}H_{37}N_4O_6S_2$ requires 733.2149; ν_{max} 3356, 2939, 1572, 1453, 1321, 1144, 1032, 786, 622, 566 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 8.47 (2 H, d, *J* = 8.5 Hz, Ar***H***), 8.03–8.10 (2 H, m, Ar***H***), 7.96 (2 H, d, *J* = 8.7 Hz, Ar***H***), 7.35–7.43 (4 H, m, Ar***H***), 7.20–7.24 (2 H, m, Ar***H***), 7.16 (4 H, t, *J* = 7.9 Hz, Ar***H***), 6.64 (2 H, dd, *J* = 8.0, 1.0 Hz, Ar***H***), 6.20 (2 H, br. s, N***H***), 3.98–4.09 (4 H, m, OC***H***$_2$), 2.85–2.92 (12 H, s, NC***H***$_3$); δ_C (125 MHz, $CDCl_3$) 154.4, 151.7, 136.5, 135.2, 130.6, 130.4, 129.8, 129.5, 128.9, 128.21, 123.1, 121.7, 118.9, 118.7, 118.7, 115.1, 86.7, 63.1, 45.4 ppm; *m*/*z* (ESI) 733.2 $[M + H]^+$, 755.2 $[M + Na]^+$.

6,6-Bis(mesylamido)-2,2′-biphenyldioxacyclodecyne (53)



This compound is novel. To a stirred solution of compound **50** (69 mg, 0.259 mmol, 1.0 eq.) in anhydrous DCM (1.9 mL) was added mesyl chloride (0.1 mL, 148 mg, 1.29 mmol, >5 eq.) and pyridine (0.1 mL, 98 mg, 1.83 mmol, >5 eq.). The solution was degassed, then stirred at room temperature for six hours. Water (10 mL) was added, then the product was extracted with ethyl acetate (3 × 10 mL). The combined organic extracts were dried over $MgSO_4$, filtered and concentrated under reduced pressure. The crude product was purified by column chromatography (eluent: DCM – 5 : 1 DCM/MeOH) to yield the pure product as a white solid (73 mg, 0.173 mmol, 70%). R_f 0.55 (99 : 1 DCM/MeOH); m.p. > 200 °C; found (ESI-TOF) 445.0494, $[M + Na]^+$ calcd for $C_{18}H_{18}N_2O_6S_2Na$ 445.0498; ν_{max} 3271, 2355, 2330, 1578, 1454, 1356, 1323, 1216, 1154, 1014, 965, 745, 524 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.61 (2H, d, *J* = 8.0, Ar***H***), 7.52 (2H, t, *J* = 8.0, Ar***H***), 7.08 (2H, d, *J* = 8.0, Ar***H***), 6.14 (2H, br. s, –N***H***–), 4.55–4.42 (4H, m, –OC***H***$_2$–), 2.89 (6H, s, –SO$_2$C***H***$_3$); δ_C (125 MHz, $CDCl_3$) 154.9, 136.8, 131.7, 121.8, 119.5, 118.4, 86.9, 63.8, 39.7; *m*/*z* (ESI) 445.0 $[M + Na]^+$.

6,6′-Bis(tosylamide)-2,2′-biphenyldioxacyclodecyne butyl bridged derivative (54)

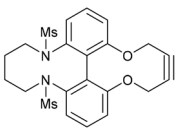


This compound is novel. A solution of 1,4-dibromobutane (34.5 mg, 0.160 mmol, 1 eq.) and compound **51** (96 mg, 0.160 mmol, 1 eq.) in MeCN (1.6 mL) was added to a mixture of Ca_2CO_3 (130 mg, 0.400 mmol, 2.5 eq.) in MeCN (6 mL) at 75 °C over 5 h. The reaction was stirred for a further 12 h

before the solvent was removed under vacuum and H_2O (20 mL) was added. The product was extracted with EtOAc (3 × 20 mL) and the combined organic extracts were dried over $MgSO_4$, which was removed by filtration, and concentrated. The crude material was then purified by column chromatography (eluted with 50% EtOAc/pet. ether 40 : 60) to give the pure product as an oil (43 mg, 0.068 mmol, 44%).

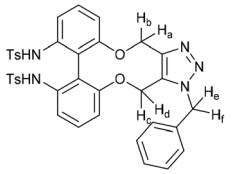
R_f = 0.48 (1 : 1 EtOAc/hexane); (found (ESI)) 651.1586 $C_{34}H_{32}N_2NaO_6S_2$ requires 651.1594; ν_{max} 3026, 2924, 2872, 1592, 1453, 1371, 1125, 1041, 813, 684, 571, 555 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.62 (4 H, d, J = 8.2 Hz, Ar***H***), 7.49 (2 H, t, J = 7.6 Hz, Ar***H***), 7.31 (2 H, d, J = 7.6 Hz, Ar***H***), 7.21 (2 H, d, J = 7.6 Hz, Ar***H***), 7.16 (4 H, d, J = 8.2 Hz, Ar***H***), 4.58–4.69 (4 H, m, 2 × OC***H***$_2$), 3.06–3.13 (2 H, m, NC***H***$_2$), 2.95–3.02 (2 H, m, NC***H***$_2$), 2.38 (6 H, s, 2 × C***H***$_3$), 1.00–1.12 (2 H, m, C***H***$_2$), 0.84–0.98 (2 H, m, C***H***$_2$); δ_C (125 MHz, $CDCl_3$) 158.0, 143.5, 141.1, 135.9, 132.8, 123.0, 129.2, 128.5, 124.5, 121.1, 89.8, 62.1, 52.8, 22.1, 21.5 ppm; m/z (ESI) 629.2 $[M + H]^+$, 651.2 $[M + Na]^+$.

6,6′-Bis(mesylamide)-2,2′-biphenyldioxacyclodecyne butyl bridged derivative (56)



This compound is novel. To a solution of caesium carbonate (147 mg, 0.45 mmol, 2.5 eq.) in dry acetonitrile (6.8 mL), stirred and heated at 75 °C was added dropwise a solution of compound **53** (76 mg, 0.18 mmol, 1.0 eq.) and 1,4-dibromobutane (65 μL, 0.54 mmol, 3.0 eq.) in acetonitrile (20 mL) over 24 hours. The solution was then stirred for a further 24 hours, before being cooled to room temperature. Solvents were removed *in vacuo*. Water (20 mL) was added, and the product was extracted with ethyl acetate (3 × 20 mL). The combined organic layers were dried over $MgSO_4$, filtered and concentrated under reduced pressure. The resulting crude product was purified by column chromatography (eluent: DCM/MeOH gradient) to afford the pure product as a brown solid (53 mg, 0.11 mmol, 62%). R_f: 0.28 (29 : 1 DCM/MeOH); m.p. 131–132 °C (dec.); found 499.0959 (ESI-TOF) m/z: $[M + Na]^+$ calcd for $C_{22}H_{24}N_2O_6S_2Na$ 499.0968; ν_{max} 3034, 2932, 2871, 2331, 1723, 1573, 1449, 1322, 1150, 1047, 969, 935, 894, 748, 664, 529; δ_H (500 MHz, $CDCl_3$) 7.49 (2H, t, J = 8.0, Ar***H***), 7.29–7.27 (2H, m, Ar***H***) 7.19 (2H, d, J = 8.0, Ar***H***), 4.57–4.45 (4H, m, –OC***H***$_2$–), 3.33–3.26 (4H, m, MsNC***H***$_2$–), 2.89 (6H, s, –SO_2C***H***$_3$) 1.33–1.09 (4H, m, MsNCH_2C***H***$_2$–); δ_C (125 MHz, $CDCl_3$) 156.4, 141.0, 133.0, 130.4, 126.9, 121.5, 88.3, 62.5, 52.3, 40.3, 23.7; m/z (ESI) $[M + Na]^+$ 499.0.

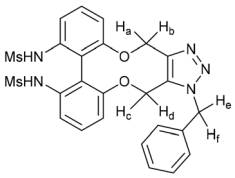
N,N′-(1-Benzyl-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazole-9,10-diyl)bis(4-methylbenzenesulfonamide) (57)



This compound is novel. To a solution of compound **51** (28.8 mg, 0.050 mmol, 1 eq.) in $CDCl_3$ (0.5 mL) was added benzyl azide (6.7 mg, 0.050 mmol, 1 eq.). The reaction was monitored by proton NMR until completion. The solvent was removed under vacuum and the crude material was purified by column chromatography (eluted with 50–100% EtOAc/hexane) to give the pure product (12.1 mg, 0.017 mmol, 34%).

R_f = 0.27 (1 : 1 EtOAc/hexane); (found (ESI)) 730.1770 $C_{37}H_{33}N_5NaO_6S_2$ requires 730.1764; ν_{max} 3337, 3059, 2925, 2854, 1596, 1455, 1288, 1043, 728, 553 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.70 (2 H, d, J = 8.1 Hz, Ar***H***), 7.54 (2 H, d, J = 8.1 Hz, Ar***H***), 7.31–7.35 (3 H, m, Ar***H***), 7.23–7.29 (4 H, m, Ar***H***), 7.14–7.22 (3 H, m, Ar***H***), 7.10 (1 H, d, J = 8.2 Hz, Ar***H***), 7.03 (2 H, m, J = 5.0 Hz, Ar***H***), 6.77 (1 H, d, J = 8.2 Hz, Ar***H***), 6.50 (1 H, d, J = 8.1 Hz, Ar***H***), 6.18 (2 H, br. s, N***H***), 5.62 (1 H, d, J = 15.8 Hz, C***H***$_e$H$_f$), 5.41 (1 H, d, J = 15.8 Hz, CH$_e$***H***$_f$), 5.17 (1 H, d, J = 13.6 Hz, C***H***$_a$H$_b$), 5.01 (1 H, d, J = 13.6 Hz, CH$_a$***H***$_b$), 4.93 (1 H, d, J = 13.4 Hz, C***H***$_c$H$_d$), 4.68 (1 H, d, J = 13.4 Hz, CH$_c$***H***$_d$); δ_C (125 MHz, $CDCl_3$) 158.1, 156.7, 144.3, 144.1, 144.0, 136.2, 136.1, 136.1, 136.0, 134.4, 131.3, 130.8, 130.8, 129.9, 129.7, 129.2, 128.7, 127.5, 127.4, 126.9, 115.6, 114.9, 113.7, 113.7, 112.6, 112.1, 63.9, 61.5, 52.3, 21.7, 21.6 ppm; m/z (ESI)706.2 $[M - H]^-$.

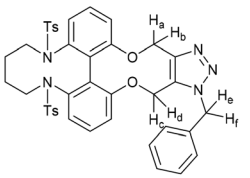
N,N′-(1-Benzyl-4,15-dihydro-1*H*-dibenzo[7,8:9,10][1,6]dioxecino[3,4-*d*][1,2,3]triazole-9,10-diyl)dimethanesulfonamide (59)



This compound is novel. Compound **53** (10 mg, 23.7 μmol, 1.0 eq.) was dissolved in $CDCl_3$ (0.6 mL) to which was added benzyl azide (3.1 μL, 24.8 μmol, 1.0 eq.). After 9 days, the reaction was deemed complete; solvents were removed *in vacuo* and the residue was purified by column chromatography (eluent: DCM/MeOH gradient) to yield the pure product as a yellow-brown solid (20 mg, since the yield is >100%, an NMR-silent impurity is likely present). R_f: 0.20 (99 : 1 DCM/MeOH); found 578.1136 (ESI-TOF) m/z: $[M + Na]^+$ calcd for $C_{25}H_{25}N_5O_6S_2Na$ 578.1138; δ_H (500 MHz, $CDCl_3$) 7.42–7.30 (9H, m, Ar***H***), 7.11–7.09 (2H, m, Ar***H***), 6.15 (1H, s, –N***H***–), 6.04 (1H, s, –N***H***–), 5.71 (1H, d, J = 15.5, PhC***H***$_e$H$_f$–), 5.49 (1H, d, J = 16.0, PhH$_e$***H***$_f$–), 5.46 (1H, d, J = 14.0, –OC***H***$_a$H$_b$–), 5.32 (1H, d, J

= 14.0, –OCH$_a$***H***$_b$–), 5.18 (1H, d, *J* = 13.5, –OC***H***$_c$H$_d$–), 4.94 (1H, d, *J* = 13.0, –OCH$_c$***H***$_d$–); δ_C (125 MHz, $CDCl_3$); 158.4, 157.0, 144.0, 136.4, 136.1, 134.4, 131.7, 131.4, 129.5, 129.3, 128.8, 119.5, 115.7, 114.7, 114.4, 113.8, 113.7, 112.7, 64.2, 61.8, 52.5, 40.3, 40.2; *m*/*z* (ESI) $[M + Na]^+$ 578.1.

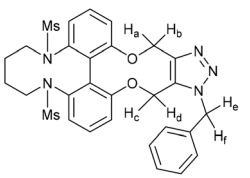
15-Benzyl-4,9-ditosyl-4,5,6,7,8,9,15,18-octahydro-14*H*-13,19-dioxa-4,9,15,16,17-pentaazadibenzo[hi,qr]cyclopenta-[d] decalene (60)



This compound is novel. To a solution of compound **54** (12.5 mg, 0.020 mmol, 1 eq.) in $CDCl_3$ (0.5 mL) was added benzyl azide (2.7 mg, 0.020 mmol, 1 eq.). The reaction was monitored by proton NMR until completion. The solvents were removed under vacuum and the crude material was purified by column chromatography (eluted with 50% EtOAc/pet. ether 40 : 60) to give the pure product as a colourless oil (9.2 mg, 0.12 mmol, 60%).

R_f = 0.15 (1 : 19 MeOH/DCM); (found (ESI)) 784.2222 $C_{41}H_{39}N_5NaO_6S_2$ requires 784.2234; ν_{max} 2950, 1452, 1348, 1163, 1068, 728, 694 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.30–7.40 (5 H, m, Ar***H***), 7.22–7.30 (5 H, m, Ar***H***), 7.10–7.20 (6 H, m, Ar***H***), 6.88 (1 H, d, *J* = 7.9 Hz, Ar***H***), 6.50 (1 H, d, *J* = 8.1 Hz, Ar***H***), 6.20 (1 H, d, *J* = 7.8 Hz, Ar***H***), 5.76 (1 H, d, *J* = 15.4 Hz, C***H***$_e$H$_f$), 5.65 (1 H, d, *J* = 13.4 Hz, OC***H***$_c$H$_d$), 5.52 (1 H, d, *J* = 15.4 Hz, CH$_e$***H***$_f$), 5.42 (1 H, d, *J* = 13.4 Hz, OCH$_c$***H***$_d$), 5.37 (1 H, d, *J* = 12.5 Hz, OC***H***$_a$H$_b$), 5.12 (1 H, d, *J* = 12.5 Hz, OCH$_a$***H***$_b$), 3.24–3.56 (4 H, m, 2 × NC***H***$_2$), 2.41 (6 H, s, 2 × C***H***$_3$), 1.77–1.97 (2 H, m, C***H***$_2$), 1.55–1.77 (2 H, m, C***H***$_2$); δ_C (125 MHz, $CDCl_3$) 159.7, 157.4, 145.0, 143.5, 143.5, 140.4, 140.1, 134.8, 133.9, 133.3, 132.3, 130.1, 129.3, 129.1, 129.1, 128.7, 128.6, 128.6, 128.5, 128.3, 127.2, 120.5, 118.6, 115.9, 113.6, 62.8, 61.6, 51.1, 51.0, 24.7, 24.3, 21.6 ppm; *m*/*z* (ESI) 784.2 $[M + Na]^+$.

15-Benzyl-4,9-bis(methylsulfonyl)-4,5,6,7,8,9,15,18-octahydro-14*H*-13,19-dioxa-4,9,15,16,17-pentaazadibenzo-[hi,qr] cyclopenta[*d*]decalene (61)



This compound is novel. Compound **56** (10.4 mg, 21.8 μmol, 1.0 eq.) was dissolved in $CDCl_3$ (0.6 mL), to which was added benzyl azide (2.72 μL, 21.8 μmol, 1.0 eq.). After 28 hours, the reaction was deemed complete, and solvents were removed *in vacuo*. The reaction was performed again with Compound **11** (5.2 mg, 10.9 μmol, 1.0 eq.) and benzyl azide (1.35 μL, 10.8 μmol, 1.0 eq.). After 26 hours, the reaction was deemed complete, and solvents were removed *in vacuo*. The residues from each attempt at this reaction were purified together by column chromatography (eluent: DCM/MeOH gradient) to afford the pure product as a yellow-brown solid (8 mg, 13.1 μmol, 40%). R_f: 0.19 (99 : 1 DCM/MeOH); HRMS (ESI-TOF) *m*/*z*: found 632.1603 $[M + Na]^+$ calcd for $C_{25}H_{31}N_5O_6S_2Na$ 632.1608; ν_{max} 2928, 2253, 1714, 1573, 1452, 1336, 1149, 1066, 971, 904, 723, 647, 513 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 7.36–7.32 (4H, m, Ar***H***), 7.23 (1H, t, *J* = 8.0, Ar***H***), 7.17–7.15 (3H, m, Ar***H***), 7.08 (1H, d, *J* = 8.0, Ar***H***), 6.99 (1H, d, *J* = 8.0, Ar***H***), 6.68 (1H, d, *J* = 7.5, Ar***H***), 5.73 (1H, d, *J* = 16.0, PhC***H***$_e$H$_f$–), 5.52 (1H, d, *J* = 14.0, –OCH$_a$***H***$_b$–), 5.41 (1H, d, *J* = 16.0, PhC***H***$_e$H$_f$–), 5.37 (1H, d, *J* = 14.0, –OCH$_a$***H***$_b$–), 5.25 (1H, d, *J* = 13.0, –OC***H***$_c$H$_d$–), 5.05 (1H, d, *J* = 13.0, –OCH$_c$***H***$_d$–), 4.17–3.64 (4H, m, 2× –NC***H***$_2$–), 2.63 (3H, s, –SO_2CH_3), 2.57 (3H, s, –SO_2CH_3), 2.10–1.69 (4H, m, –NCH$_2$C***H***$_2$–); δ_C (125 MHz, $CDCl_3$) 159.0, 157.5, 144.5, 140.6, 140.0, 134.6, 132.0, 129.4, 129.3, 129.0, 128.7, 128.3 128.1, 127.1, 121.1, 118.9, 115.5, 114.2, 63.4, 61.2, 52.4, 51.3, 51.2, 36.5, 35.3, 25.4, 24.1; *m*/*z* (ESI) 632.1 $[M + Na]^+$

Data availability

The research data (and/or materials) supporting this publication can be accessed at **https://wrap.warwick.ac.uk/**.

Author contributions

Sam Forshaw, Richard C. Knighton, Neelam Tiwari, Samson M. Oladeji and Andrew C. Stevens carried out synthetic chemistry, contributed to the design of the project and contributed to the writing up. Jeremy S. Parker contributed to the project design. William T. Scott carried out synthetic chemistry and molecular biology, contributed to the design of the project and contributed to the writing up. Yean Ming Chew and Jami Reber carried out molecular biology, contributed to the design of the project and contributed to the writing up. Guy J. Clarkson completed the X-ray crystal structure analyses and provided data for the project. Mohan K. Balasubramanian and Martin Wills conceived and directed the project.

Conflicts of interest

There are no conflicts to declare.

Acknowledgements

We thank the EPSRC and AstraZeneca for support of SF through a National Productivity Investment Fund (NPIF) studentship and the EPSRC for support of RCK through research grant EP/M006670/1. Lijiang Song is thanked for the Mass Spectrometry analyses. The X-ray diffraction instrument was obtained through the Science City Project with support from Advantage West Midlands (AWM) and part funded by the

European Regional Development Fund (ERDF). YMC and WS were supported by PhD studentships from the MRC funded Doctoral Training Partnership in Interdisciplinary Biomedical Research (grant numbers MR/R502212/1 and MR/N014294/1, respectively). SMO was supported by a Scholarship from the Commonwealth Scholarships Commission. SMO and JR were students supported by the Warwick University Analytical Sciences and Instrumentation MSc course, MKB thanks the Welcome Trust (grant 101885/C/13/Z). The authors acknowledge Dr Cleidi Zampronio and the Warwick Proteomics Research Technology Platform for Mass Spectrometry analyses. The authors thank Dr Saravanan Palani for assistance and advice with the molecular biology.

References

1 (*a*) N. Stephanopoulos and M. B. Francis, *Nat. Chem. Biol.*, 2011, **7**, 876–884; (*b*) S. M. Ryan, G. Mantovani, X. Wang, D. M. Haddleton and D. J. Brayden, *Expert Opin. Drug Delivery*, 2008, **5**, 371–383; (*c*) A. Beck, L. Goetsch, C. Dumontet and N. Corvaia, *Nat. Rev. Drug Discovery*, 2017, **16**, 315–337; (*d*) J. A. Prescher and C. R. Bertozzi, *Nat. Chem. Biol.*, 2005, **1**, 13–21.

2 (*a*) G. Wittig and A. Krebs, *Chem. Ber.*, 1961, **94**, 3260; (*b*) T. Harris and I. V. Alabugin, *Mendeleev Commun.*, 2019, **29**, 237–248; (*c*) D. H. Ess, G. O. Jones and K. N. Houk, *Org. Lett.*, 2008, **10**, 1633–1636; (*d*) J. Dommerholt, F. P. J. T. Rutjes and F. L. van Delft, *Top. Curr. Chem.*, 2016, **374**, 16.

3 (*a*) R. Huisgen, *Angew. Chem., Int. Ed. Engl.*, 1963, **2**, 565–598; (*b*) V. V. Rostovtsev, L. G. Green, V. V. Fokin and B. K. Sharpless, *Angew. Chem., Int. Ed.*, 2002, **41**, 2596–2599; (*c*) C. W. Tornoe, C. Christensen and M. Meldel, *J. Org. Chem.*, 2002, **67**, 3057–3064.

4 (*a*) N. J. Agard, J. A. Prescher and C. R. Bertozzi, *J. Am. Chem. Soc.*, 2004, **126**, 15046–15047; (*b*) N. J. Agard, J. M. Baskin, J. A. Prescher, A. Lo and C. R. Bertozzi, *ACS Chem. Biol.*, 2006, **1**, 644–648; (*c*) J. M. Baskin, J. A. Prescher, S. T. Laughlin, N. J. Agard, P. V. Chang, I. A. Miller, A. LoJ, A. Codelli and C. R. Bertozzi, *Proc. Natl. Acad. Sci. U. S. A.*, 2007, **104**, 16793–11697; (*d*) X. Ning, J. Guo, M. A. Wolfert and G. J. Boons, *Angew. Chem., Int. Ed.*, 2008, **47**, 2253–2255; (*e*) N. E. Mbua, J. Guo, M. A. Wolfert, R. Steet and G. J. Boons, *Chem. Biochem.*, 2011, **12**, 1912–1921; (*f*) J. Dammerholt, S. Schmidt, R. Temming, L. J. A. Hendricks, F. P. J. T. Rutjes, J. C. M. van Hest, D. J. Lefeber, P. Friedl and F. L. van Delft, *Angew. Chem., Int. Ed.*, 2010, **49**, 9422–9425; (*g*) M. F. Debets, S. S. van Berkel, S. Schoffelen, F. P. J. T. Rutjes, J. C. M. van Hest and F. L. van Delft, *Chem. Commun.*, 2010, **46**, 97–99; (*h*) J. Jewett, E. Sletten and C. R. Bertozzi, *J. Am. Chem. Soc.*, 2010, **132**, 3688–3690.

5 A. Del Grosso, L. D. Galanapoulos, C. K. C. Chiu, G. J. Clarkson, P. B. O'Conner and M. Wills, *Org. Biomol. Chem.*, 2017, **15**, 4517–4521.

6 A. Mistry, R. C. Knighton, S. Forshaw, Z. Dualeh, J. S. Parker and M. Wills, *Org. Biomol. Chem.*, 2018, **16**, 8965–8975.

7 R. C. Knighton, K. Sharma, N. S. Robertson, D. R. Spring and M. Wills, *ACS Omega*, 2019, **4**, 2160–2167.

8 T. Harris, G. dos Passos Gomes, S. Ayad, R. J. Clark, V. V. Lobodin, M. Tuscan, K. Hanson and I. V. Alabugin, *Chem*, 2017, **3**, 629–640.

9 U. Koch-Pomeranz, H. J. Hansen and H. Schmidt, *Helv. Chim. Acta*, 1973, **56**, 2981–3004.

10 B. Zhou, P. Jiang, J. Lu and C. Xing, *Arch. Pharma. Chem. Life Sci.*, 2016, **349**, 539–552.

11 C. Huang, Q. Yin, J. Meng, W. Zhu, Y. Yang, X. Qian and Y. Xu, *Chem. – Eur. J.*, 2013, **19**, 7739–7747.

12 T. Nguyen and M. B. Francis, *Org. Lett.*, 2003, **5**, 3245–3248.

13 G. de Almeida, E. M. Sletten, H. Nakamura, K. K. Palaniappan and C. R. Bertozzi, *Angew. Chem., Int. Ed.*, 2012, **51**, 2443–2447.

14 R. Ni, N. Mitsuda, T. Kashiwagi, K. Igawa and K. Tomooka, *Angew. Chem., Int. Ed.*, 2015, **54**, 1190–1194.

15 J. M. Yoon, C. Y. Lee, Y. I. Jo and C. H. Cheon, *J. Org. Chem.*, 2016, **81**, 8464–8469.

16 T. Harada, S. Ueda, T. Yoshida, A. Inoue, M. Takeuchi, N. Ogawa, A. Oku and M. Shiro, *Org. Lett.*, 2000, **2**, 1319–1322.

17 Y. Zhuang, Y. He, Z. Zhou, W. Xia, C. Cheng, M. Wang, B. Chen, Z. Zhou, J. Pang and L. Qiu, *J. Org. Chem.*, 2015, **80**, 6968–6975.

18 G. Delogu, D. Fabbri, S. Menichetti and C. Nativi, *Tetrahedron*, 2003, **59**, 2131–2136.

19 T. He, L. Peng, S. Li, F. Hu, C. Xie, S. Huang, S. Jia, W. Qin and H. Yan, *Org. Lett.*, 2020, **22**, 6966–6971.

20 C. Bressy, D. Alberico and M. Lautens, *J. Am. Chem. Soc.*, 2005, **127**, 13148–13149.

21 T. Geiger, A. Haupt, C. Maichle-Mössmer, C. Schrenk, A. Schnepf and H. F. Bettinger, *J. Org. Chem.*, 2019, **84**, 10120–10135.

22 C. Tahtaoui, I. Parrot, P. Klotz, F. Guillier, J. L. Galzi, M. Hibert and B. Ilien, *J. Med. Chem.*, 2004, **47**, 4300–4315.

23 Y. Liu, W. M. Ren, C. Liu, S. Fu, M. Wang, K. K. He, R. R. Li, R. Zhang and X. B. Lu, *Macromolecules*, 2014, **47**, 7775–7788.