Synthetic studies on 4,5-dihydro-3*H*-1,2,4-triazole-3,5-diones bearing fluorogenic residues at N-4

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A number of fluorescent 4,5-dihydro-3*H*-1,2,4-triazole-3,5-diones have been made. Intra-inter-electrophilic substitution by the triazoledione moiety on the activated naphthalene ring of 4-[6-(5-dimethylamino-1-naphthylsulfonamido)hexyl]-4,5-dihydro-3*H*-1,2,4-triazole-3,5-dione $4a_3$ leads to rapid decomposition. The dienophilicity of the triazoledione moiety in 4-pyren-1-yl-4,5-dihydro-3*H*-1,2,4-triazole-3,5-dione 4d is dramatically lowered by steric shielding. Insertion of a three-carbon spacer unit into the latter compound, to give the 3-pyren-1-ylpropyl analogue 4e, affords a valuable fluorogenic reagent for the analysis of trace levels of 1,3-dienes. Powdered barium manganate is shown to be an excellent solid-phase oxidant for conversion of urazoles into 1,2,4-triazole-3,5-diones.

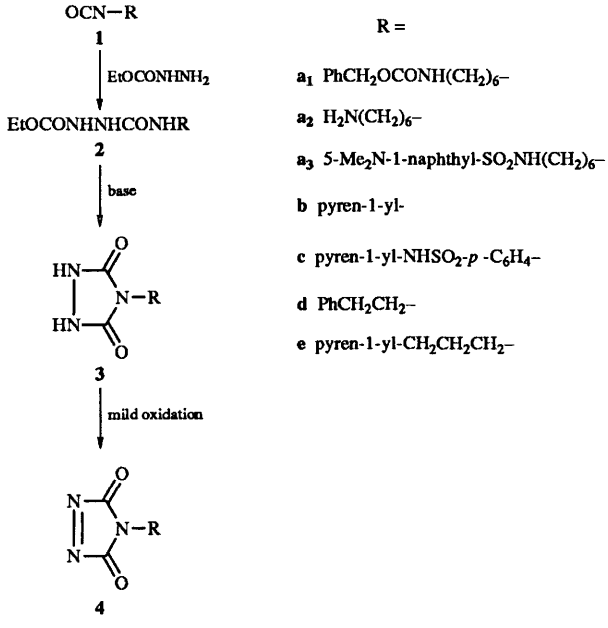
Introduction

4-Substituted 4,5-dihydro-3*H*-1,2,4-triazole-3,5-diones (TADs) are notable for their ability to participate in a wide range of concerted and stepwise additions. Considerable attention has been paid to their additions to activated alkenes[1] and highly strained systems[2] but probably it is their powerful dienophilic properties which have received the widest recognition.[3–7] In this paper we report studies aimed at the synthesis of TADs, bearing fluorogenic residues at N-4, which are suitable for the detection at femtomolar levels of biologically important dienes[8] and related species such as the D vitamins,[9] hydroperoxy dienoic acids[10] and hydroxydienoic acids.[11] The general route to N-4 substituted TADs (Scheme 1) introduced by Cookson *et al.*[4] was adopted as the basis for these studies. In addition to variation of the isocyanate precursors, this allows some synthetic elaboration to be effected at the relatively stable 1,2,4-triazolidine-3,5-dione (urazole) stage.

Results and discussion

The first synthetic target was TAD $4a_3$ in which the dansyl† fluorogen is linked by a six-carbon spacer unit to the TAD moiety. The initial strategy called for entry into the above scheme with the isocyanate $1a_1$ and the introduction of the dansyl group at the urazole $3a_2$ stage. In accord with Sayer's findings,[12] the amine precursor **5** of the isocyanate $1a_1$ could only be produced from hexane-1,6-diamine in useful yields (55%) by adopting a high-dilution and low-temperature procedure which introduced significant practical difficulties when applied on a large scale. However, in a less structured approach, satisfactory yields (20%) of the required ethyl carbazate derivative $2a_1$ were obtained from a one-pot reaction in which a 1:1 mixture of benzyl alcohol and ethyl carbazate were added directly to hexane-1,6-diyl diisocyanate. Base-catalysed cyclisation of $2a_1$ to the urazole $3a_1$ proceeded smoothly as did reductive removal of the benzyloxycarbonyl group on the spacer unit with formic acid over palladium-on-charcoal[13] to give the aminourazole $3a_2$ as the formate salt. Rigorous washing with boiling methanol was necessary to remove $3a_2$ from the support when cyclohexa-1,4-diene rather than formic acid was employed as the hydrogen source in the latter step.

Dansylation of $3a_2$ under standard conditions failed to produce the required dansylurazole $3a_3$, giving instead

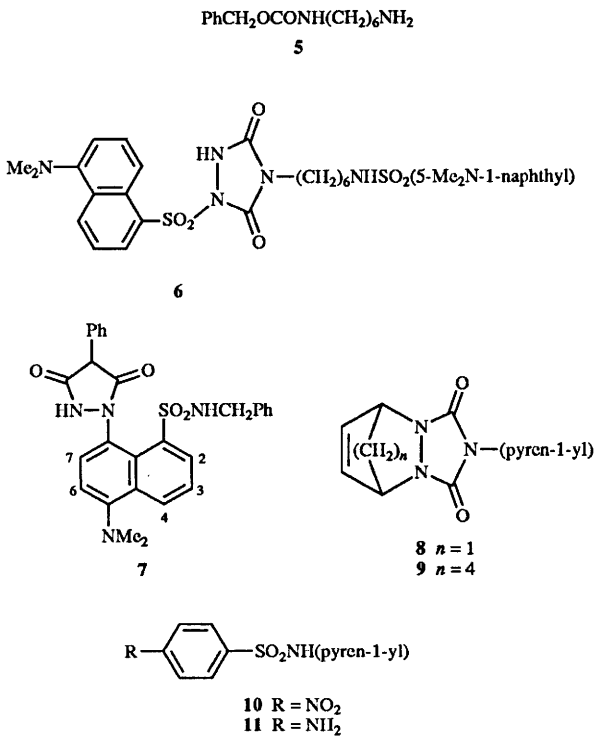


Scheme 1

appreciable amounts of the di-dansyl species **6**, even when only 1 equiv. of dansyl chloride was used. Evidently the ready deprotonation of the hydrazo residue under the weakly basic conditions gives this function a nucleophilicity comparable to that of the primary amine group.[14] Slow addition of dansyl chloride to a buffered aqueous acetonitrile solution (pH 10.3) of the amine was necessary in order to obtain the mono-dansylurazole $3a_3$. The reaction was continuously monitored and the addition stopped when a significant level of the bis species was detected. Recovered starting material was recycled.

Attempts to obtain the required TAD $4a_3$ by oxidation of $3a_3$ failed. Furthermore, no adduct of the TAD was isolated when addition of oxidants was followed directly by additions of cyclopentadiene to the reaction solutions. Suspicion that responsibility for this failure lay with rapid attack by the electrophilic TAD centre on the electron-rich ring of the dansyl group,[15] rather than an inability of the dansyl moiety to survive the weak oxidising conditions, was strengthened by the observation that 4-phenyl-TAD reacted readily with the dansyl derivative of benzylamine. Analytical data for the principal product **7** from this reaction was fully in accord with substitution by the 4-TAD at position-8 of the dansyl residue, despite this position being considerably more hindered than

† Dansyl = 5-dimethylamino-1-naphthylsulfonyl.



position-6. Highfield ^{1}H NMR spectroscopy clearly identified 8-H in the substrate by its long-range coupling (1.1 Hz) to 4-H. Both the 8-H signal and the long-range coupling at 4-H were absent in the product **7**. Furthermore, NOE studies on **7** showed a 2.0% enhancement of the 7-H signal when the one proton in the urazole ring was irradiated.

In the search for a less sensitive fluorophore attention turned to pyrene. *N*-Pyren-1-yl maleimide has been used as a fluorogenic analytical reagent for thiol groups[16] but the maleimido residue lacks the marked dienophilic characteristic of the TAD grouping. The pyrene nucleus is not attacked by TADs. With 4-pyren-1-yl-4,5-dihydro-3*H*-1,2,4-triazole-2,5-dione **4b** as the target, pyrene was converted by dinitrogen tetroxide into 1-nitropyrene which was smoothly reduced to 1-aminopyrene with hydrazine over palladium–charcoal.[17] Treatment of the amine with trichloromethyl chloroformate gave the isocyanate **1b** which, without isolation, was converted into the highly insoluble semicarbazide **2b** and subsequently into the urazole **3b** by standard methods. Oxidation of **3b** with *tert*-butyl hypochlorite produced TAD **4b** as a deep red solid.

Without further purification, **4b** was treated with cyclopentadiene and cycloocta-1,3-diene to give the corresponding adducts **8** and **9**, respectively. The ^{1}H NMR spectra of both of these products provided clear evidence for restricted rotation about the pyrenyl–N bond. This is apparent from the ^{1}H NMR spectrum of the cyclopentadiene adduct **8** which reveals a rotamer ratio of approximately 55:45 and is detailed in the Experimental section. The assignments of the 2-H and 10-H pyrenyl protons are based on the simpler spectrum of the urazole **3b** in which these protons can be clearly assigned by spin decoupling. Assignment of the two sets of signals from the six cyclopentene ring protons in the spectrum of **8** follows from a comparison of the spectrum from the corresponding phenyl TAD adduct.[5] The hindered rotation about an aryl to nitrogen bond accords with that observed in numerous simpler systems.[18] Less expected, however, was the finding that when a 4×10^{-3} mol dm^{-3} solution of methyl (*E*,*E*)-octadeca-9,11-dienoate in ethyl acetate was treated with a four-fold excess of **4b** under ambient conditions, unchanged ester could still be detected after 18 h. The pyrene residue clearly hinders the approach of a bulky diene to the reactive centre of the TDA, underlining the need for a dienophile to carry a spacer unit between such a fluorogen and the triazolinedione moiety.

The TAD **4c**, with a phenylsulfonyl spacer unit between the aminopyrene and the triazolinedione components, became the next target. Treatment of 1-aminopyrene with 4-nitrobenzenesulfonyl chloride gave the corresponding sulfonamide **10** in good yield. Reduction of the nitro group[17] produced the corresponding aniline **11** which, without further purification, was converted into **1c** by treatment with trichloromethyl chloroformate. Addition of ethyl carbazate to the reaction mixture afforded the carbazide **2c** in an overall yield of 30%. Cyclisation of **2c** under standard conditions gave only a low yield of the required urazole **3c**. Oxidation of the urazole with *tert*-butyl hypochlorite produced a solution showing the characteristic absorbance of a TAD but attempts to isolate this product resulted in complex mixtures. The cycloocta-1,3-diene adduct of TAD **4c** was rapidly formed when the diene was added to the oxidised solution of **3c**, in agreement with the observation by Cookson[19] that electron-withdrawing substituents positioned *para* to the triazoline dione group in phenyltriazolinediones enhance reactivity.

In view of the limited stability of **4c**, it was decided to place a trimethylene aliphatic spacer between the TAD moiety and the pyrene fluorophore using 3-pyren-1-ylbutyric acid as starting material. In a model synthesis, phenylpropanoic acid was converted into its azide with diphenylphosphoryl azide.[20] Without isolation, the azide was rearranged to the isocyanate **1d** and the product treated *in situ* with ethyl carbazate to produce the semicarbazide **2d** in an overall yield of 76%. Base cyclisation afforded the urazole **3d** in good yield. When this procedure was applied to 3-pyren-1-ylbutyric acid the corresponding semicarbazide **2e** was obtained in an overall yield of 52% and the urazole **3e** in a 75% yield.

Chlorine in tetrachloromethane, a very efficient general reagent for urazole oxidations, was used initially to oxidise urazole **3e**. However, when an excess of cycloocta-1,3-diene was added to the degassed reaction solution, to establish the effectiveness of the oxidation, the cycloocta-1,3-diene adduct of TAD **4e** was obtained in only a 50% (isolated) yield. Closer inspection of the reaction showed that the yield of dienophile from this particular urazole is lowered by some chlorine substitution in the pyrenyl ring system. An extensive survey of oxidising conditions (below) established that urazole **3e** is most satisfactorily oxidised to TAD **4e** by finely powdered barium manganate with yields reaching 95%.

Analytical reagents must fulfil two key criteria if they are to be used to quantify analytes at the femtomolar level. To avoid extended reaction times they must be highly reactive to negate the need for their use in disproportionately high concentrations. Inevitably they will be used in excess. They must, therefore, also react cleanly and be formed cleanly, if generated *in situ*, to minimise interference with the assay employed.

The dienophilicity of the TAD **4e** towards cycloocta-1,3-diene, in dry ethyl acetate at 28 °C, was compared with that of 4-phenyl-TAD and TAD **4c**. With the diene in excess, estimates of the second-order rate constants, based on the first-order decay of the characteristic triazoline dione absorption band at 530 nm,[19] were 14.5×10^{-3} and 10.2×10^{-3} s mol dm^{-3}, respectively for 4-phenyl-TAD and **4e**. The corresponding values for 4-phenyl-TAD and **4c**, based on HPLC estimates of the first-order rate of adduct formation, were 10.6×10^{-3} and 23.9×10^{-3} s^{-1} mol dm^{-3}, respectively. Subsequent application of **4e** to the determination of 25-hydroxy Vitamin D_3[21] and octadecadienoic acid[22] levels in blood serum confirmed that its dienophilicity is adequate for analyses at femtomolar levels.

The choice of barium manganate as the cleanest of the potential oxidants for the final stage of the synthesis of **4e** followed investigations into the ability of a range of mild oxidising agents to convert 4-phenylurazole into 4-phenyl-TAD. For our purposes both clean and rapid conversion were

Table 1 Phenylurazole oxidations

Oxidant [w/mg]	Amount[a]	Yield of 4-PhTAD (%) Early reading (t/min)	After 18 h
Cl_2(g)	Slow stream	100 (0.5)	98
Cl_2[b]	1.0	97 (1.2)	97
N_2O_4[b]	1.0	95 (12)	96
Bu^tOCl[b]	2.0	97 (20)	98
Tetralin OOH[b,c] + $VO(acac)_2$	1.0	8.3 (75)	14.5
DDQ[b]	1.0	12 (10), 21 (30)	63
DDQ[b]	2.0	13 (10), 21 (30)	65
NBS[b]	1.0	50 (10)	56
NBS[b]	2.0	71 (10)	78
$(PhSeO)_2O$[b]	0.5	45 (10), 47 (30)	35
$(PhSeO)_2O$[b]	1.0	59 (10), 62 (30)	—
Ag_2O[200]	ss[d]	0 (15)	78
MnO_2[100]	ss	54 (10)	53
$Ca(OCl)_2$[200]	ss	40 (20)	—
$BaMnO_4$[22]	ss	51 (2.5)	—
$BaMnO_4$[40]	ss	100 (2.5)	—
$BaMnO_4$[100]	ss	98 (2.5)	—

[a] Figures show [oxidant]/[phenylurazole], [phenylurazole] = 8.1×10^{-3} mol dm^{-3}. [b] Oxidant added in small volume of inert solvent. [c] [Tetralin OOH]:[$VO(acac)_2$] = $1:4 \times 10^{-4}$. [d] ss = stirred suspensions.

important. A gaseous or solid oxidant was favoured to facilitate ready removal of the excess from the reaction medium.

In the survey undertaken, the oxidants were added to stirred solutions of the phenylurazole at room temperature. Ethyl acetate was used as solvent. The yield of phenyl-TAD and its stability under the reaction conditions was monitored by measuring the intensity of its characteristic absorbance at 530 nm (ε 162) within 1.5 h of the start of the reaction and at 18 h. The results are summarised in Table 1. Both chlorine and dinitrogen tetraoxide[6] reacted rapidly to give excellent yields of 4-phenyl-TAD although their electrophilicity made them unsuitable for the oxidation of the pyrenylurazole **3e** (see above). No evidence for subsequent decomposition of the product was found in either case. *tert*-Butyl hypochlorite[4] also oxidised rapidly. The reaction with chlorine was particularly fast and yields in experiments where molar ratios of reagent to substrate were less than unity suggested the participation of radical-chain processes involving molecular oxygen. The photosensitive *tert*-butyl hypochlorite may also act as a radical chain initiator. Reaction with tetralin hydroperoxide in the presence of a trace of vanadium catalyst lent further support for a radical chain process although this reaction was much slower and had barely reached 15% yield in 18 h. Oxidations with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) and with *N*-bromosuccinimide (NBS) were faster. These reagents behaved similarly in that the rates of phenyl-TAD formation were little influenced by the reagent concentration, a finding suggesting the slow rate-determining breakdown of an intermediate in these cases. Neither of these reagents effected complete conversion. Oxidation with selenic anhydride[7] may be analogous but phenyl-TAD is lost over the 18 h period and the data is limited.

Of the suspended solids examined silver carbonate on Celite, 10% palladium-on-charcoal and 10% platinum-on-charcoal were essentially inert. Silver oxide suspensions oxidised very slowly giving a 78% yield of phenyl-TAD after 18 h. Suspensions of activated manganese dioxide and calcium hypochlorite both showed limited promise. However levels of MnO_2 in excess of 150 mg per 0.1 mmol of urazole appeared to decompose the phenyl TAD produced. Neither approached the performance of powdered barium manganate which rapidly effected near quantitative conversion into the required TAD. The quality of the barium manganate is important to the success of this reaction. These studies used material supplied by Strem Chemicals Inc.

Experimental

General

Unless otherwise stated ^1H and ^{13}C NMR spectra were recorded at 250 and 62.9 MHz, respectively, on a Bruker AM 250 spectrometer using $CDCl_3$ as solvent. A Bruker AM 300 spectrometer was used for additional ^1H NMR measurements on *N*-benzyl-5-dimethylaminonaphthalene-1-sulfonamide and its adduct. UV spectra were recorded either on a Phillips PU 8720 UV–VIS scanning spectrometer or on a Pye-Unicam instrument. Fluorescence spectra were measured using a Perkin-Elmer LS-3B instrument. IR spectra were recorded as mulls or liquid films unless otherwise indicated, using a Perkin-Elmer 398 spectrometer. A VG Micromass MM1GF mass spectrometer was employed for low-resolution CI spectra. High-resolution CI spectra were carried out at the SERC Mass Spectrometry Centre, Swansea, using a VG ZAB-E instrument.

Elemental microanalyses were performed by Butterworth Laboratories Ltd., Teddington, Middlesex. Melting point measurements were made using a pre-calibrated Gallenkamp electrothermal apparatus. Flash chromatography was carried out using Merk Kieselgel 60 F_{254} (230–300 mesh). Certain preparative separations were achieved by using a Harrison 7924 T Chromatotron fitted with plates coated to a thickness of 2 mm with Merk #7749 silica gel. Kieselgel 60 F_{254} coated (0.25 mm) glass-backed plates were used for TLC. For HPLC a Walters 6000 A solvent delivery system coupled to either a Pye-Unicam PV 4020 UV detector or an LC3 UV detector was used. Gradient elution was achieved by use of a Walters 660 Solvent Programmer. The following columns and solvent systems were employed. The flow rate in each case was 1.0 cm^3 min^{-1} except for 'ODS 2, System B', where it was 1.5 cm^3 min^{-1}. Anachem, Spherisorb 5 μm S-5 nitrile, 250 × 4.6 mm, hexane–propan-2-ol–water in the ratio (i) 95:4.5:0.5 (S5 nitrile, System A), (ii) 85:14.5:0.5 (S5 nitrile, System B) and (iii) 70:29.5:0.5 (S5 nitrile, System C). Anachem, Spherisorb 5 μm ODS 2, 250 × 4.6 mm, (i) methanol–water 85:15 (ODS 2, System A) and (ii) CH_3CN–0.035 mol dm^{-3} CH_3CO_2H graded from 40:60 to 70:30 over 40 min (ODS 2, System B). Unless otherwise stated, starting materials were obtained from commercial suppliers and used without further purification. Diethyl ether and tetrahydrofuran were distilled from sodium–benzophenone ketyl immediately prior to use. Methanol and ethanol were distilled from the corresponding magnesium alkoxide. Pyridine

was distilled from and stored over potassium hydroxide pellets. Dichloromethane, light petroleum (bp 40–60 °C) and petroleum (bp 60–80 °C) were distilled from phosphorus pentaoxide. Ethyl acetate was distilled from calcium hydroxide. Benzene was distilled from and stored over sodium wire. 4-Phenyl-4,5-dihydro-*3H*-1,2,4-triazole-3,5-dione was prepared by the method reported by Cookson *et al.*[4] Palladium (10%)-on-carbon was supplied by the Engelhard Chemical Group. Barium manganate was supplied by Strem Chemical Inc.

4-[6-(Benzyloxycarbonylamino)hexyl]-1-ethoxycarbonylsemicarbazide 2a$_1$

Over a period of 45 min a mixture of benzyl alcohol (16.22 g, 0.15 mol) and ethyl carbazate (15.62 g, 0.15 mol) in dry benzene (200 cm^3) was added to a stirred solution of hexane-1,6-diyl diisocyanate (25.23 g, 0.15 mol) in benzene (50 cm^3) maintained at 0–5 °C under nitrogen. The solution was allowed to warm to room temperature after which it was refluxed for 90 min and then stored for 13 h. The waxy solid which separated was filtered off and washed with boiling ethyl acetate (2 × 50 cm^3) to give a white powder which on recrystallisation, (aqueous methanol) gave 4,4′-hexane-1,6-diylbis(1-ethoxycarbonylsemicarbazide) (19.5 g, 51 mmol, 33%), mp 197–204 °C (Found C, 44.6; H, 7.3; N, 22.6. $C_{14}H_{28}N_6O_6$ requires C, 44.67; H, 7.49; N, 22.32%); ν_{max}/cm^{-1} 3320, 3240, 3120, 1705 and 1580; δ_H([$^2H_6$]-DMSO) 1.18 (6 H, t, *J* 7.0, 2 × OCH$_2$C*H*$_3$), 1.26 (4 H, m, C*H*$_2$CH$_2$CH$_2$N), 1.41 (4 H, m, C*H*$_2$CH$_2$N), 3.18 (4 H, br q, *J* 6.5, 2 × C*H*$_2$NH), 4.08 (4 H, q, *J* 7.0, 2 × OC*H*$_2$CH$_3$), 5.74 (2 H, br t, *J* 6.0, 2 × CH$_2$N*H*CO), 7.10 (2 H, br s, 2 × NH) and 7.90 (2 H, br s, 2 × NH); *m*/*z* (20 eV) 376 (M$^+$), 273, 227 and 169.

Evaporation of the combined ethyl acetate–benzene filtrates gave a second waxy solid consisting of two components which were separated by column chromatography with ethyl acetate→ethyl acetate–methanol (6:1). The first component eluted was obtained as a white solid which crystallised from ethyl acetate to yield *N*,*N*′-bis(benzyloxycarbonyl)hexane-1,6-diamine (19.2 g, 0.049 mol, 30%), mp 126–128 °C; R_F (ethyl acetate) 0.73. The second component from the column was the title compound **2a$_1$** (10.26 g, 0.026 mol, 19%), mp 121–123 °C (propan-2-ol–hexane) (Found: C, 57.0; H, 7.5; N, 14.3. $C_{18}H_{28}N_4O_5$ requires C, 56.8; H, 7.4; N, 14.7%); HPLC t_R 11.1 min (S5 nitrile, System C); ν_{max}/cm^{-1} 3320, 3240, 1700 and 1340; λ_{max}(MeOH)/nm 251 (log ε 2.11), 257 (2.20), 263 (2.09) and 267 (1.92); δ_H 1.26 (3 H, t, *J* 7.0, OCH$_2$C*H*$_3$), 1.33 (4 H, m, C*H*$_2$CH$_2$CH$_2$NH), 1.50 (4 H, m, C*H*$_2$CH$_2$NH), 3.20 (4 H, quint, *J* 7, 2 × C*H*$_2$NH), 4.20 (2 H, q, *J* 7.0, OC*H*$_2$CH$_3$), 4.98 (1 H, br s, NH), 5.10 (2 H, s, PhCH$_2$), 5.42 (1 H, br s, NH), 6.50 (1 H, br s, NH), 6.80 (1 H, br s, NH) and 7.30 (5 H, m, Ph); *m*/*z* (20 eV) 381 (M$^+$ + 1), 380 (M$^+$), 316 and 290.

4-[6-(Benzyloxycarbonylamino)hexyl]-1,2,4-triazolidine-3,5-dione 3a$_1$

Freshly prepared 0.85 mol dm^{-3} KOH in ethanol–water (3:2; 16 cm^3, 13 mmol) was added to a stirred, heated (60 °C) solution of the semicarbazide **2a$_1$** (4.00 g, 1 mmol) in ethanol–water (3:2). After the reaction mixture had been heated under reflux for 2.5 h it was poured into 2 mol dm^{-3} HCl (60 cm^3) and extracted with ethyl acetate (3 × 50 cm^3). The combined extracts were dried (MgSO$_4$), filtered and evaporated to yield the title compound **3a$_1$** (2.107 g, 6.3 mmol, 60%), mp 135–136 °C (ethyl acetate–hexane) (Found C, 57.4; H, 6.8; N, 16.6. $C_{16}H_{22}N_4O_4$ requires C, 57.4; H, 6.6; N, 16.7%); HPLC t_R 7.1 min (S5 nitrile, System C); ν_{max}/cm^{-1} 3320, 3100, 1680, 1540, 1340, 1298, 790 and 700; δ_H 1.35 (4 H, m, C*H*$_2$CH$_2$CH$_2$NH), 1.49 (2 H, m, C*H*$_2$CH$_2$NH), 1.68 (2 H, br quint, *J* 7, C*H*$_2$CH$_2$urazole), 3.18 (2 H, q, *J* 6.5, C*H*$_2$NH), 3.54 (2 H, t, *J* 7.0, C*H*$_2$urazole), 4.90 (1 H, br s, NH), 5.10 (2 H, s, C*H*$_2$Ph), 7.30 (5 H, m, Ph) and 8.10 (2 H, br s, CONHNHCO); *m*/*z* (20 eV) 316, 290 and 249.

Deprotection of the dione 3a$_1$

Palladium (10%)-on-carbon (411 mg) was added to a stirred solution of the urazole **3a$_1$** (200 mg, 5.9 × 10^{-4} mol) in methanol containing 6% (v/v) formic acid (40 cm^3). The solution was heated under reflux for 3 min and then stored under nitrogen while it was sampled (10 mm^3) and monitored (HPLC) for the disappearance of the benzyloxycarbonyl moiety. After 30 min the palladium-on-carbon was filtered off and washed with boiling methanol (2 × 10 cm^3). Evaporation of the filtrate yielded the formate salt of 4-(6-aminohexyl)-1,2,4-triazolidine-3,5-dione **3a$_2$** (112 mg, 72.1%) as a glassy solid; ν_{max}/cm^{-1} 3000v br and 1680; δ_H(60 MHz) (D_2O) 1.40 [8 H, m, CH$_2$(C*H*$_2$)$_4$CH$_2$], 3.10 (2 H, br t, *J* 7.0, C*H*$_2$urazole) and 3.50 (2 H, t, *J* 7.0, C*H*$_2$NH$_3^+$). This was used in the next step without further purification.

Attempted dansylation of the preceding formate salt. A solution of dansyl chloride (90.7 mg, 3.36 × 10^{-4} mol) in acetonitrile (10 cm^3) was added in one portion to a stirred solution of the formate salt of **3a$_2$** (73.5 mg, 2.8 × 10^{-4} mol) in 1 mol dm^{-3} aq. $NaHCO_3$ (28 cm^3). Stirring was continued for 30 min at room temperature, after which time the mixture showed the presence of three fluorescent components (HPLC). After dilution of the reaction mixture with chloroform (20 cm^3) the aqueous phase was separated and extracted with chloroform (2 × 10 cm^3). The combined organic phase and extracts were dried (MgSO$_4$), filtered, and evaporated to yield a bright yellow foam (92.7 mg), column chromatography of which with chloroform→chloroform–ethanol (94:6) as eluent gave the major component **6** as a bright yellow foam (53.3 mg, 8 × 10^{-5} mol, 48%), mp 76–80 °C; HPLC t_R 11.6 min (S5 nitrile, System 3); ν_{max}/cm^{-1} 3300, 1795, 1730, 1560 and 790; δ_H 0.60 (2 H, m, C*H*$_2$CH$_2$N), 0.90 (2 H, m, C*H*$_2$CH$_2$N), 1.10 (4 H, m, CH$_2$CH$_2$CH$_2$N), 2.78 (2 H, q, *J* 6.5, C*H*$_2$NH), 2.86 (6 H, s, *Me*$_2$NAr), 2.90 (6 H, s, *Me*$_2$NAr), 3.20 (2 H, t, *J* 6.8, C*H*$_2$urazole), 4.96 (1 H, t, *J* 7.0, CH$_2$N*H*), 7.15–8.68 (12 H, complex, naphthyl) and 8.10 (1 H, v br s, NCON*H*NCO); *m*/*z* (20 eV) 433 and 377.

4-[6-(5-Dimethylamino-1-naphthylsulfonamido)hexyl]-1,2,4-triazolidine-3,5-dione 3a$_3$

A solution of the formate salt of **3a$_2$** (106 mg, 0.4 × 10^{-3} mol) in deionised water (10 cm^3) adjusted to pH 10.2 with 0.1 mol dm^{-3} aqueous NaOH was treated with aqueous sodium carbonate–sodium hydrogen carbonate buffer (pH 10.3; 25 cm^3) and acetonitrile (30 cm^3). Dansyl chloride (109 mg, 0.4 × 10^{-3} mol) in acetonitrile (55 cm^3) was added to this mixture at a rate of 1 cm^3 min^{-1} with vigorous stirring until, after the addition of 20 cm^3 (1.46 × 10^{-4} mol), HPLC indicated some formation of the bis-dansyl derivative **6**. The addition was stopped and the solution was acidified with 2 mol dm^{-3} hydrochloric acid and extracted with ethyl acetate (2 × 30 cm^3).

The aqueous phase was readjusted to pH 10.3 and then diluted with acetonitrile after which the above described dansylation procedure repeated until all the dansyl chloride solution had been added. The ethyl acetate phases were combined, dried (MgSO$_4$), filtered and evaporated to give a green foam which was purified by column chromatography using chloroform→chloroform–ethanol (86:14) as eluent to give the title compound **3a$_3$** as fluorescent green cubes (43 mg, 9.92 × 10^{-5} mol, 25%), mp 149–150 °C (CH_2Cl_2–hexane) (Found C, 55.2; H, 5.4; N, 16.1. $C_{20}H_{27}N_5O_4S$ requires C, 55.4; H, 6.3; N, 16.1%); HPLC t_R 9.2 min (S 5-nitrile, System B); ν_{max}/cm^{-1} 3360, 3200, 1690, 1465, 1160, 1145, 1330 and 640; λ_{max}(EtOH)/nm 250 (log ε 4.19) and 335 (3.67); λ_{max}^{Ex}(EtOH)/nm 340; λ_{max}^{Em}/nm 520; δ_H 1.18 (4 H, m, C*H*$_2$CH$_2$CH$_2$N), 1.32 (2 H, m, C*H*$_2$CH$_2$N), 1.53 (2 H, br quint, *J* 6.5, C*H*$_2$CH$_2$urazole), 2.86 (6 H, s, *Me*$_2$NAr), 2.89 (2 H, m, C*H*$_2$NH), 3.44 (2 H, br t, *J* 6.7, C*H*$_2$urazole), 5.60 (1 H, br t, *J* 6.0, CH$_2$N*H*), 7.1–8.55 (6 H, complex, naphthyl) and 8.30 (2 H, v br s, CON*H*NHCO); *m*/*z* (20 eV) 434 (M$^+$), 433, 375 and 250.

N-Benzyl-5-dimethylaminonapthalene-1-sulfonamide
Benzylamine (18.3×10^{-3} mol, 2.0 mol) was added dropwise, under an atmosphere of nitrogen, to a stirred solution of dansyl chloride (483 mg, 1.8×10^{-3} mol) in methylene dichloride (35 cm^3) held at 0 °C. After 2 min the bright yellow solution had become cloudy and showed a green fluorescence. After warming to room temperature the solution was washed with 0.4 mol dm^{-3} HCl (2×25 cm^3) and water (2×35 cm^3) and dried ($MgSO_4$). Evaporation of the solvent gave a pale green solid which yielded the title compound (489 mg, 1.4×10^{-3} mol, 80%) as fluorescent green cubes, mp 140–141.5 °C (ethyl acetate) (Found C, 6.0; H, 5.8; N, 8.2. $C_{19}H_{20}N_2O_2S$ requires C, 67.03; H, 5.92; N, 8.23%); R_F (ethyl acetate–petroleum, 1:1) 0.52; ν_{max}(Nujol)/cm^{-1} 3300, 1590, 1580, 1430, 1327, 1315, 1175, 1160, 862, 790 and 535; δ_H(300 MHz) (CD_2Cl_2) 2.90 (6 H, s, Me_2N), 4.08 (2 H, s, $PhCH_2$), 4.82 (1 H, t, SO_2NHCH_2), 7.12 (5 H, m, Ph), 7.23 (1 H, dd, *J* 7.6 and 0.8, 6-H), 7.54 (1 H, dd, *J* 7.3 and 8.5, 3-H), 7.58 (1 H, dd 7.6 and 8.7, 7-H), 8.26 (1 H, dd, *J* 7.3 and 1.3, 2-H), 8.29 (1 H, dt, *J* 8.6, 0.8 and 0.8, 8-H) and 8.57 (1 H, dt, *J* 8.5, 1.1 and 1.1, 4-H); *m/z* (EI; 70 eV) 341 ($M^+ + 1$), 339, 202 and 170.

N-Benzyl-5-dimethylamino-8-(3,5-dioxo-4-phenyl-1,2,4-triazolidin-1-yl) naphthalene-1-sulfonamide 7
4-Phenyl-1,2,4-triazolidine-3,5-dione (51 mg, 2.9×10^{-4} mol) in methylene dichloride (20 cm^3) was added in one portion, under an atmosphere of nitrogen, to a stirred solution of *N*-benzyl-5-dimethylaminonaphthalene-1-sulfonamide (100 mg, 2.9×10^{-4} mol) in dry methylene dichloride (20 cm^3) at 0 °C. The solution was allowed to warm to room temperature over 20 min, after which time the original pink colour had faded. Evaporation of the solvent yielded the title compound **7** (140 mg, 2.71×10^{-4} mol, 92%) as pale green crystals, mp 211–212 °C (ethyl acetate–hexane) (Found C, 62.6; H, 4.8; N, 13.4. $C_{27}H_{25}N_5O_4S$ requires C, 62.89; H, 4.88; N, 13.58%); R_F (ethyl acetate–light petroleum, 1:1) 0.14; ν_{max}/cm^{-1} 3301, 1783, 1719, 1580, 1503, 1458, 1424, 1367, 1317, 1266 and 1143; δ_H(300 MHz) ([$^2H_6$]-DMSO) 2.88 (6 H, s, Me_2NAr), 4.10 (2 H, m, $PhCH_2$-$NHSO_2$), 7.27 (1 H, d, *J* 8.0, 6-H), 7.30 (5 H, m, $PhCH_2NH$), 7.46 (5 H, m, *Ph* urazole), 7.64 (1 H, dd, *J* 7.5 and 8.4, 3-H), 7.81 (1 H, d, *J* 8.2, 7-H), 8.14 (1 H, dd, *J* 7.5 and 0.1, 2-H), 8.45 (1 H, dd, *J* 8.5 and 0.9, 4-H), 8.51 (1 H, br t, *J* 7.0, $NHSO_2$) and 10.80 (1 H, s, NH); *m/z* (EI; 70 eV) 515 (M^+), 408, 375 and 345.

1-Aminopyrene
Hydrazine monohydrate (0.85 cm^3, 17.5 mmol) was added to a stirred suspension of 1-nitropyrene[23] (500 mg, 2.02 mmol) in ethanol–benzene, (3:1; 35 cm^3) at room temperature followed by finely powdered 10% palladium-on-carbon (250 mg) in small portions. The resulting mixture was heated at reflux under nitrogen for 1 h before the catalyst was filtered off and washed with hot ethanol–benzene (1:3). The combined filtrate and washings were evaporated to give a pale yellow solid which was recrystallised from cyclohexane to yield 1-aminopyrene (338 mg, 1.55 mmol, 75%) as yellow plates, mp 116–117 °C (lit.,[24] 117 °C); ν_{max}/cm^{-1} 1600, 1590 and 830.

1-Ethoxycarbonyl-4-pyren-1-ylsemicarbazide 2b
Trichloromethyl chloroformate (318 mg, 1.61 mmol) was added dropwise, under a nitrogen atmosphere at room temperature, to a stirred solution of 1-aminopyrene (700 mg, 3.22 mmol) in dry benzene (250 cm^3). Throughout the remainder of the preparation the reaction solution was maintained at reflux under an atmosphere of nitrogen. After 55 min, a second aliquot of trichloromethyl chloroformate (72 mg, 0.33 mmol) was added to the cloudy solution followed, after 30 min, by an aliquot of ethyl carbazate (502 mg, 4.83 mmol) in benzene (50 cm^3) and, after a further 1 h, by a second aliquot of the carbazate (500 mg, 4.8 mmol) in benzene (20 cm^3). After a further 30 min the solution was cooled to precipitate the crude product. Evaporation of the mother liquor yielded more product. The combined crops were pre-adsorbed (ethyl acetate–acetone) onto Kieselgel and added to a Kieselgel column. Elution (ethyl acetate graded to ethyl acetate–ethanol) (9:1) gave the title compound **2b** (405 mg, 1.16 mmol, 36%), mp 270–280 °C (decomp.) (Found: C, 68.8; H, 4.9; N, 12.1. $C_{20}H_{17}N_3O_3$ requires C, 69.1; H, 4.9; N, 12.1%); HPLC t_R 9.7 min (S 5-nitrile, System B); ν_{max}/cm^{-1} 3330, 2930, 1720, 1640 and 720; λ_{max}(EtOH)/nm 243(log ε 4.85), 270 (4.36), 277 (4.60), 325 (3.36) and 340 (4.54); λ^{Em}_{max}(EtOH)/nm 340; λ^{Em}_{max}(EtOH)/nm 390; δ_H ([$^2H_6$]-DMSO) 1.28 (3 H, t, *J* 7.5, OCH_2CH_3), 4.20 (2 H, q, *J* 7.5, OCH_2CH_3), 7.80 (1 H, br, NH), 8.00–8.52 (9 H, m, pyrenyl), 8.46 (1 H, br, NH) and 8.70 (1 H, br, NH).

4-Pyren-1-yl-1,2,4-triazolidine-3,5-dione 3b
To a refluxing solution of the semicarbazide **2b** (103 mg, 0.28 mmol) in ethanol (50 cm^3) was added 1.15 mol dm^{-3} sodium ethoxide (1 cm^3, 1.15 mmol) under a nitrogen atmosphere. After 75 min, when HPLC analysis showed the reaction to be complete, the solution was poured into water (25 cm^3) containing ice (10 cm^3) and 2 mol dm^{-3} HCl (20 cm^3). The mixture was then extracted with ethyl acetate (2×25 cm^3) and the combined extracts, after warming briefly to dissolve suspended material, were dried ($MgSO_4$) and evaporated under reduced pressure to yield the title compound **3b** (40 mg, 0.13 mmol, 40%) as an off-white solid, mp 325–330 °C (decomp.) (Found: C, 69.9; H, 3.7; N, 13.9. $C_{18}H_{11}N_3O_2 \cdot 0.5H_2O$ requires C, 69.9; H, 3.9; N, 13.6%); t_R/min 13.0 (S 5-nitrile, System B); ν_{max}/cm^{-1} 3160, 1768, 1670, 1510, 1230, 850 and 825; λ_{max}(EtOH)/nm 233 (log ε 4.63), 243 (4.85), 270 (4.40), 276 (4.65), 325 (4.43) and 341 (4.58); λ^{Ex}_{max}(EtOH)/nm 340; λ^{Em}_{max}(EtOH)/nm 390; δ_H([$^2H_6$]-DMSO) 7.91 (1 H, d, *J* 9.5, 10-H), 8.07 (1 H, d, *J* 8.2, 2-H), 8.15 (1 H, t, *J* 7.5, 7-H), 8.27 (1 H, d, *J* 7.5, 4-H or 5-H), 8.30 (1 H, d, *J* 9.5, 9-H), 8.31 (1 H, d, *J* 7.5, 4-H or 5-H), 8.38 (1 H, d, *J* 9.0, 6-H or 8-H), 8.40 (1 H, d, *J* 9.0, 6-H or 8-H), 8.41 (1 H, d, *J* 8.2, 3-H) and 10.70 (2 H, br, NH-NH); *m/z* (20 eV) 302 ($M^+ + 1$), 301 (M^+), 243, 215 and 187.

2-Pyren-1-yl-2,3,5,8-tetrahydro-5,8-methano-1*H*-[1,2,4]triazolo-[1,2-*a*]pyridazine-1,3-dione 8
A solution of *tert*-butyl hypochlorite (55 mg, 0.51 mmol) in ethyl acetate (0.5 cm^3) was added to a stirred suspension of compound **3b** (53 mg, 0.17 mmol) in ethyl acetate (25 cm^3) at room temperature under a nitrogen atmosphere. After 30 min, freshly cracked cyclopentadiene (34 mg, 0.52 mmol) was added to the red solution which was then stirred for a further 30 min. Removal of the solvent gave a pale brown solid which was crystallised from aqueous ethanol and further purified by column chromatography (ethyl acetate–light petroleum, 1:1) followed by recrystallisation from methylene dichloride–hexane to give the cycloadduct **8** as pale brown crystals (15 mg, 0.041 mmol, 25%), mp 240–250 °C (decomp.) (Found: C, 72.2; H, 4.2; N, 10.7. $C_{23}H_{15}N_3O_2 \cdot H_2O$ requires C, 72.05; H, 4.5; N, 11.0%); ν_{max}/cm^{-1} 3005, 2955, 2920, 2840, 1770, 1720, 1415, 1400, 1250 and 850; **rotamer A** (55%) δ_H 2.10 (1 H, dt, *J* 8.0 and 1.5, bridging H *exo* to alkene); 2.40 (1 H, br d, *J* 8.5, bridging H *endo* to alkene), 5.26 (2 H, quint, *J* 1.8, $CHCH{=}CHCH$), 6.64 (2 H, t, *J* 1.8, $CHCH{=}CHCH$), 7.79 (1 H, d, *J* 8.2, pyrenyl 10-H), 7.95 (1 H, d, *J* 9.5, pyrenyl 2-H) and 8.00–8.30 (7 H, complex, pyrenyl); **rotamer B** (45%) δ_H 2.06 (1 H, dt, *J* 8.5 and 1.5, bridging H *exo* to alkene), 2.40 (1 H, br d, *J* 8.5, bridging H *endo* to alkene), 5.28 (2 H, quint, *J* 1.8, $CHCH{=}CHCH$), 6.79 (2 H, t, *J* 1.8, $CHCH{=}CHCH$), 7.66 (1 H, d, *J* 9.5, pyrenyl 10-H), 7.89 (1 H, d, *J* 8.2, pyrenyl 2-H) and 8.00–8.30 (7 H, complex, pyrenyl); λ^{Ex}_{max}(EtOH)/nm 273; λ^{Em}_{max}(EtOH)/nm 390 and 430; *m/z* 365 (M^+), 301 and 214.

2-Pyren-1-yl-2,3,5,8-tetrahydro-5,8-butano-1*H*-[1,2,4]triazolo-[1,2-*a*]pyridazine-1,3-dione 9
tert-Butyl hypochlorite (200 mg, 1.8 mmol) was added to a

stirred suspension of compound **3b** (54 mg, 0.17 mmol) in ethyl acetate (250 cm^3) at room temperature under nitrogen. After 10 min the homogeneous solution was evaporated to dryness. The deep red product was redissolved in ethyl acetate (10 cm^3) and cycloocta-1,3-diene (119 mg, 1.1 mmol) in ethyl acetate (5 cm^3) was added to the stirred solution under nitrogen over a 15 min interval. The solution was stirred for a further 2 h before being left at −12 °C overnight. Evaporation of the solvent gave a brown oil which, after column chromatography (ethyl acetate–light petroleum, 1:1) crystallised from methylene dichloride–ether to give the title adduct **9** (33.7 mg, 0.082 mmol, 49%) as a cream solid, mp 274–276 °C; ν_{max}/cm^{-1} 2980, 1760, 1690, 1445, 1420 and 910; δ_H 1.81 (6 H, m, butano), 2.39 (2 H, butano), 5.15 (2 H, m, C*H*CH=CHC*H*), 6.26 (1 H, dd, *J* 2 and 4, CHC*H*=C*H*CH rotamer), 6.29 (1 H, dd, *J* 2 and 4, CHC*H*=C*H*CH rotamer) and 7.90–8.30 (9 H, complex, pyrenyl); *m/z* 408 ($M^+ + 1$), 407 (M^+), 243 and 214.

4-Nitro-(*N*-pyren-1-ylsulfamoyl)benzene 10

4-Nitrobenzenesulfonyl chloride (204 mg, 0.92 mmol) in pyridine (10 cm^3) was added in one portion to a stirred solution of 1-aminopyrene (100 mg, 0.46 mmol) in pyridine (10 cm^3). The resulting solution was heated at 80 °C under nitrogen for 1 h and then poured onto crushed ice (100 cm^3). After 1 h the crude product was collected as a yellow precipitate, washed with water (50 cm^3) and crystallised from aqueous ethanol to give the title compound **10** (142 mg, 0.352 mmol, 77%) as fine yellow needles, mp 256–257 °C (Found: C, 65.1; H, 3.5; N, 6.8. $C_{22}H_{14}N_2O_4S$ required C, 65.7; H, 3.5; N, 7.0%); t_R/min 5.0 (ODS 2, System A); ν_{max}/cm^{-1} 3280, 1600, 1530, 1460, 1420, 1250, 1180, 1090, 850 and 735; $\delta_H([^2H_6]$-DMSO) 7.7–8.36 (9 H, complex, pyrenyl), 7.93 (2 H, d, *J* 9.0, 2′-H and 6′-H), 8.31 (2 H, d, *J* 9.0, 3′ and 5′-H) and 10.9 (1 H, br s, NH); *m/z* 403 ($M^+ + 1$), 402 (M^+), 372 and 218.

1-Ethoxycarbonyl-4-[*p*-(*N*-pyren-1-ylsulfamoyl)phenyl]semicarbazide 2c

Hydrazine monohydrate (0.125 cm^3, 1.5 mmol) was added to a stirred suspension of the nitro compound **10** (100 mg, 0.24 mmol) in ethanol–benzene (1:1, 10 cm^3) followed by finely powdered 10% palladium-on-carbon (25 mg). After the solution had been stirred under nitrogen for 20 min at room temperature, the catalyst was filtered off and the filtrate was evaporated and the residue dried *in vacuo* to yield compound **11** as a bright yellow foam. A solution of this in dry acetonitrile (12 cm^3) and trichloromethyl chloroformate (56 mg, 0.28 mmol) was refluxed under an atmosphere of nitrogen for 2 h after which it was treated with ethyl carbazate (50 mg, 0.46 mmol) in acetonitrile (3 cm^3). Refluxing was continued for a further 4 h after which the mixture was evaporated to give the crude semicarbazide as an oil; this was precipitated from aqueous ethanol as a solid (78 mg). The crude product **2c**, pre-adsorbed onto Kieselgel (ethanol), was eluted from a Kieselgel column with solvent graded from light petroleum–ethyl acetate 6:4 to 3:7 to give the title compound (46.3 mg, 0.092 mmol, 39%) as a powder, mp 236–238 °C (Found: C, 59.0; H, 4.2; N, 11.0. $C_{26}H_{22}N_4O_5S\cdot 1.5\ H_2O$ requires C, 59.0; H, 4.75; N, 10.6%); t_R/min 6.8 (S 5-nitrile, System C); ν_{max}/cm^{-1} 3360, 3251, 1736, 1635, 1563, 1364, 1160 and 839; λ_{max}(EtOH)/nm 243 (log ε 4.55), 267 (2.42), 277 (4.45), 330 (2.51) and 343 (4.23); $\delta_H([^2H_6]$-DMSO) 1.20 (3 H, t, *J* 7.5, OCH_2CH_3), 4.04 (2 H, q, *J* 7.5, OCH_2CH_3), 7.65 (5 H, m, 4 phenylene H + 1 pyrenyl H), 8.20 (8 H, m, pyrenyl), 8.92 (1 H, br s, NH), 9.34 (1 H, br s, NH), 10.40 (1 H, s, NH) and 10.62 (1 H, s, NH); *m/z* (20 eV) 502 (M^+), 429 and 375.

4-[*p*-(*N*-Pyren-1-ylsulfamoyl)phenyl]-1,2,4-triazolidine-3,5-dione 3c

Freshly prepared 1.72 mol dm^{-3} sodium ethoxide (17.5 cm^3, 2.9 mmol) was added to a stirred, refluxing solution of the semicarbazide **2c** (100 mg, 1.9 mmol) in dry ethanol (75 cm^3) under nitrogen. After 4 h the reaction was quenched (ethanol–12 mol dm^{-3} hydrochloric acid) and the solution and accompanying suspension were partitioned between chloroform (100 cm^3) and water (100 cm^3). The organic phase was separated, washed with water (100 cm^3) and dried ($MgSO_4$) to give the crude urazole as a brown solid (700 mg). This was pre-adsorbed onto Kieselgel (acetone) and added to a chromatographic column. Elution, with solvent graded from chloroform to chloroform–methanol (50:1), gave the urazole **3c** as an amorphous solid (193 mg, 0.42 mmol, 22%), mp 185–190 °C (Found: C, 60.5; H, 3.3; N, 11.7. $C_{24}H_{16}N_4O_4S\cdot H_2O$ requires C, 60.7; H, 3.8; N, 11.8%); t_R/min 5.4 (S 5-nitrile, System C); ν_{max}/cm^{-1} 3200, 2956, 1702, 1597, 1504, 1412, 1160, 1094 and 843; λ_{max}^{Ex}-(EtOH)/nm 280 or 350; λ_{max}^{Em}(EtOH)/nm 430; $\delta_H([^2H_6]$-DMSO) 7.76 (4 H, m, phenylene), 8.20 (9 H, m, pyrenyl) and 10.80 (3 H, br, NH); *m/z* 456 (M^+), 398, 372 and 218.

1-Ethoxycarbonyl-4-phenethylsemicarbazide 2d

Diphenylphosphoryl azide (775 mm^3, 3.6 mmol) was added to a solution of 3-phenylpropionic acid (500 mg, 3.3 mmol) in dry acetonitrile (10 cm^3), under an atmosphere of argon. After the solution had been stirred for 5 min at 0 °C, triethylamine (1.0 cm^3, 7.2 mmol) was added to it over a period of 10 min. The solution was allowed to warm to room temperature and then refluxed for 10 min and treated with ethyl carbazate (367 mg, 3.6 mmol) in acetronitrile (2 cm^3). Heating under reflux was continued for a further 12 h before the solution was concentrated (0.5 cm^3) and diluted with ethyl acetate (20 cm^3). The resulting solution was warmed with saturated aqueous $NaHCO_3$ (10 cm^3), water (10 cm^3) and 2 mol dm^{-3} hydrochloric acid (10 cm^3), dried ($MgSO_4$) and evaporated to give the crude semicarbazide (679 mg, 82%). Crystallisation of this from ethyl acetate–ethanol afforded the title compound **2d** (640 mg, 76%) as colourless prisms, mp 138 °C (Found: C, 57.3; H, 6.65; N, 16.7. $C_{12}H_{17}N_3O_3$ requires C, 57.3; H, 6.8; N, 16.7%); t_R(HPLC)/min 6.7 (S 5-nitrile, System B); ν_{max}/cm^{-1} 3348, 3232, 1686, 1605, 1552, 1496, 1345, 1062, 946 and 765; δ_H 1.20 (3 H, t, *J* 7.0, CH_3CH_2O), 2.72 (2 H, t, *J* 7.5, CH_2Ph), 3.28 (2 H, br t, *J* 7.5, CH_2NH), 4.15 (2 H, q, *J* 7.0, CH_3CH_2O), 6.32 (1 H, br t, *J* 5, CH_2NH), 7.20 (5 H, m, Ph), 7.70 (1 H, br s, NH) and 8.70 (1 H, br s, NH); *m/z* 252 ($M^+ + 1$) and 251 (M^+).

4-Phenethyl-1,2,4-triazolidine-3,5-dione 3d

A solution of potassium hydroxide (51 mg, 0.87 mmol) in ethanol–water (1:1; 5 cm^3) was added to compound **2d** (149 mg, 0.59 mmol) in ethanol–water (1:1; 4 cm^3). The solution was refluxed for 100 min before the addition of a second aliquot of potassium hydroxide (80 mg, 1.37 mmol) in the same solvent (4 cm^3). After a further 20 min under reflux the solution was neutralised with 2 mol dm^{-3} HCl and evaporated. The crude urazole recrystallised (ethanol) to give the title compound **3d** (66 mg, 0.32 mmol, 54%) as prisms, mp 201 °C (Found: C, 58.6; H, 5.4; N, 20.4. $C_{10}H_{11}N_3O_2$ requires C, 58.5; H, 5.4; N, 20.5%); t_R/min 6.4 (S 5-nitrile, System B); ν_{max}/cm^{-1} 3200br, 1672, 1211, 1105, 1069, 914 and 850; $\delta_H([^2H_6]$-DMSO) 2.88 (2 H, t, *J* 7.5, CH_2Ph), 3.60 (2 H, t, *J* 7.5, CH_2N), 7.25 (5 H, m, Ph) and 10.00 (2 H, br s, NHNH); δ_C 32.96 (CH_2), 39.02 (CH_2), 126.28 (CH), 128.30 (CH), 128.52 (CH), 138.01 (C) and 154.80 (C=O); *m/z* 203 (M^+) and 104.

1-Ethoxycarbonyl-4-(3-pyren-1-ylpropyl)semicarbazide 2e

To a stirred solution of 3-pyren-1-ylbutyric acid (961 mg, 3.3 mmol) in dry THF (50 cm^3) at 0 °C under argon, was added diphenylphosphoryl azide (1.008 g, 3.6 mmol) followed after 15 min by triethylamine (0.907 g, 8.9 mmol). After 5 min the solution was removed from the ice-bath and after a further 50 min heated under reflux under an atmosphere of nitrogen for 100 min before ethyl carbazate (570 mg, 5.58 mmol) in dry THF (5 cm^3) was added to the boiling solution. After a further 20 min the solution was cooled to room temperature

and concentrated to 20 cm^3 under reduced pressure. Ethyl acetate (100 cm^3) was added to the concentrate which was then washed successively with saturated aqueous $NaHCO_3$, water, 2 mol dm^{-3} hydrochloric acid and water, dried ($MgSO_4$) and evaporated to give the crude semicarbazide. This, after two recrystallisations (ethyl acetate), yielded the title compound **2e** (680 mg, 52%), mp 177.5–178.5 °C (Found: C, 70.95; H, 5.8; N, 10.9. $C_{23}H_{23}N_3O_3$ requires C, 70.9; H, 5.95; N, 10.8%); t_R/min 22.8 (ODS 2, System B); ν_{max}/cm^{-1} 3335, 2931, 1724, 1640, 1533, 1286, 1215, 1055, 839 and 721; λ_{max}(EtOH)/nm 234 (log ε 4.74), 243 (4.97), 265 (4.53), 276 (4.81), 312 (4.20), 327 (4.58) and 343 (4.76); λ_{max}^{Ex}(EtOH)/nm 340, 276 or 243; λ_{max}^{Em}(EtOH)/nm 376 and 390; δ_H([$^2H_6$]-DMSO) 1.20 (3 H, t, *J* 7.0, CH_3CH_2O), 1.94 (2 H, quint, *J* 7.0, CH_2CH_2NH), 3.30 (4 H, m, $CH_2CH_2CH_2$), 4.05 (2 H, q, *J* 7.0, CH_3CH_2O), 6.55 (1 H, br s, NH), 7.75 (1 H, br s, NH), 7.90–8.35 (9 H, complex, pyrenyl) and 8.80 (1 H, br s, NH); δ_C 14.47 (CH_3), 29.97 (CH_2), 32.10 (CH_2), 39.11 (CH_2), 60.30 (CH_2), 123.38 (CH), 124.12 (C), 124.20 (C), 124.66 (CH), 124.83 (CH), 124.87 (CH), 125.99 (CH), 126.35 (CH), 127.09 (CH), 127.25 (CH), 127.36 (CH), 127.98 (C), 129.17 (C), 130.36 (C), 130.84 (C), 136.59 (C), 156.89 (C=O) and 158.37 (C=O); *m*/*z* 390 (M^+ + 1), 389 (M^+) and 343.

4-(3-Pyren-1-ylpropyl)-1,2,4-triazolidine-3,5-dione 3e

To a refluxing solution of **2e** (50 mg, 1.43 mmol) in ethanol (60 cm^3) was added a solution of potassium hydroxide (403 mg, 7.1 mmol) in ethanol (10 cm^3). After the solution had been refluxed for 55 min, it was diluted with ethanol and acidified (12 mol dm^{-3} hydrochloric acid) to give a white precipitate (432 mg). This was filtered off after the liquid phase had been reduced to a quarter of its original volume. Crystallisation (ethanol) of the product gave the title compound **3e** (336 mg, 0.98 mmol, 75%), mp 262–264 °C (Found: C, 73.4; H, 5.0; N, 12.3. $C_{22}H_{12}N_3O_2$ requires C, 73.45; H, 5.0; N, 12.2%); t_R(HPLC)/min 16.4 (ODS 2, System B); ν_{max}/cm^{-1} 3200, 1735, 1690, 840 and 720; λ_{max}(EtOH)/nm 234 (log ε 4.72), 243 (4.94), 265 (4.49), 276 (4.77), 313 (4.14), 326 (4.56) and 343 (4.72); λ_{max}^{Ex}(EtOH)/nm 276 or 340; λ_{max}^{Em}(EtOH)/nm 376 and 390; δ_H([$^2H_6$]-DMSO) 2.09 (2 H, quint, *J* 7.5, $CH_2CH_2CH_2$), 3.35 (2 H, t, *J* 7.5, CH_2pyrenyl), 3.59 (2 H, t, *J* 7.5, CH_2N), 7.94–8.38 (9 H, complex, pyrenyl) and 10.20 (2 H, br s, NHNH); δ_C([$^2H_6$]-DMSO) 29.54 (CH_2), 29.72 (CH_2), 37.94 (CH_2), 123.05 (CH), 124.08 (C), 124.19 (C), 124.75 (CH), 124.87 (2 × CH), 126.04 (CH), 126.47 (CH), 127.20 (CH), 127.26 (CH), 127.35 (CH), 127.95 (C), 129.32 (C), 130.31 (C), 130.82 (C), 135.65 (C) and 153.03 (C=O); *m*/*z* 343 (M^+), 287 and 242.

4-(3-Pyren-1-ylpropyl)-4,5-dihydro-3*H*-1,2,4-triazole-3,5-dione 4e

A suspension of compound **3e** (30 mg, 0.087 mmol) and finely powdered barium manganate (150 mg, 0.58 mmol) in ethyl acetate (10 cm^3) was stirred at room temperature in the dark for 10 min and then filtered through Celite. Evaporation of the pink filtrate gave the title compound **4e** (28 mg, 0.082 mmol, 94%) as a purple solid; ν_{max}/cm^{-1} 3037, 2929, 2861, 1784, 1724, 1603, 1456, 1392, 1372, 1261, 1100, 1012, 884 and 622; λ_{max}(EtOH)/nm 526 (log ε 2.22); δ_H 2.20 (2 H, quint, *J* 7.5, $CH_2CH_2CH_2$), 3.30 (2 H, t, *J* 7.5, CH_2pyrenyl), 3.66 (2 H, t, *J* 7.5, CH_2N) and 7.74–8.20 (9 H, complex, pyrenyl); δ_C 28.57 (CH_2), 30.19 (CH_2), 41.27 (CH_2), 122.53 (C), 124.89 (C), 124.91 (CH), 125.03 (CH), 125.11 (C), 125.25 (CH), 126.03 (CH), 126.83 (CH), 127.05 (CH), 127.41 (CH), 127.84 (CH), 128.51 (C), 130.29 (C), 130.78 (C), 131.40 (C), 133.53 (C) and 159.10 (C=O); *m*/*z* 342 (M^+ + 1), 341 (M^+) and 285.

2-(3-Pyren-1-ylpropyl)-2,3,5,8-tetrahydro-5,8-methano-1*H*-[1,2,4]triazolo[1,2-*a*]pyridazine-1,3-dione

tert-Butyl hypochlorite (32 mg, 0.28 mmol) was added to a stirred solution of compound **3e** (56 mg, 0.16 mmol) in acetone (20 cm^3). After 10 min the pink solution was treated with freshly cracked cyclopentadiene (12 mg, 0.18 mmol). When the colour of the solution had discharged (30 min) the mixture was evaporated to give the crude adduct as a brown oil which, on column chromatography (ethyl acetate–hexane), yielded the title compound (35 mg, 0.086 mmol; 53%) as cream crystals, mp 158–163 °C (Found: C, 73.1; H, 5.1; N, 9.7. $C_{26}H_{21}N_3O_2 \cdot H_2O$ requires C, 73.4; H, 5.4; N, 9.9%); ν_{max}/cm^{-1} 3400, 3008, 2929, 2857, 1766, 1709, 1602, 1444, 1412, 1375, 1327, 1198, 1073, 916, 843 and 661; λ_{max}(EtOH)/nm 234 (log ε 4.44), 243 (4.63), 265 (4.19), 276 (4.42), 326 (4.18) and 342 (4.34); δ_H 1.90 (1 H, br d, *J* 9.0, *exo* H on CH_2 bridge), 2.15 (2 H, m, $CH_2CH_2CH_2$), 2.18 (1 H, br t, *J* 9.0, *endo* H on CH_2 bridge), 3.32 (2 H, t, *J* 8.0, CH_2pyrenyl), 3.62 (2 H, t, *J* 7.0, CH_2N), 5.05 (2 H, br s, CHCH=CHCH), 6.38 (2 H, br s, CH=CH) and 7.82–8.25 (9 H, complex, pyrenyl); *m*/*z* 407 (M^+), 367, 343 and 285.

2-[3-(Pyren-1-ylpropyl)]-2,3,5,8-tetrahydro-5,8-butano-1*H*-[1,2,4]triazolo[1,2-*a*]pyridazine-1,3-dione

A 0.14 mol dm^{-3} solution of chlorine in carbon tetrachloride (7.7 cm^3, 1.09 mmol) was added, in one portion, to a solution of compound **3e** (150 mg, 0.44 mmol) in ethyl acetate (500 cm^3). The solution was stirred in the dark at room temperature for 15 min after which excess chlorine was removed by a stream of dry nitrogen. After concentration of the solution under reduced pressure to 200 cm^3, cycloocta-1,3-diene (470 mg; 4.36 mmol) was added to the concentrate. The solution was stirred for 8 h in the dark after which it was evaporated to yield an oil which was purified by chromatography (Chromatotron solvent 1:1 light petroleum–ethyl acetate) to afford the cyclooctadiene adduct as an oil (117 mg). This crystallised (light petroleum) as colourless needles (97 mg, 0.22 mmol, 50%), mp 155–157 °C (Found: C, 77.8; H, 6.1; N, 9.8. $C_{29}H_{27}N_3O_2$ requires C, 77.5; H, 6.05; N, 9.35%); R_F 0.19 (1:1 ethyl acetate–light petroleum); ν_{max}/cm^{-1} 2942, 2398, 1749, 1679, 1458, 1422, 1376, 1312, 1197, 844 and 660; λ_{max}(EtOH)/nm 243 (log ε 4.81), 265 (4.30), 276 (4.57), 327 (4.35) and 343 (4.52); λ_{max}^{Ex}(EtOH)/nm 243 or 343; λ_{max}^{Em}(EtOH)/nm 380 and 395; δ_H 1.52 (2 H, m, butano), 1.68 (4 H, m, butano), 2.14 (2 H, m, butano), 2.32 (2 H, quint, *J* 7.5, $CH_2CH_2CH_2$), 3.42 (2 H, t, *J* 7.5, CH_2pyrenyl), 3.81 (2 H, t, *J* 7.5, CH_2N), 4.90 (2 H, m, CHC=CCH), 6.08 (2 H, m, CH=CH) and 7.91–8.3 (9 H, complex, pyrenyl); *m*/*z* 449 (M^+), 484, 293, 205, 149 and 118.

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