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Bifunctional Antitumor Agents. Derivatives of Pyrrolo[9, 10-*b*]phenanthrene- a DNA Intercalative Delivery Template

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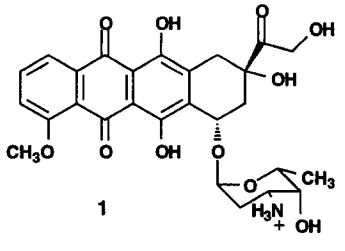
Abstract - The preparation and intercalative ability of the pyrrolo[9, 10-*b*]phenanthrene nucleus is described. Functionalization of the nucleus was achieved using standard indole chemistry. The utility of this template as a DNA intercalating drug delivery system is demonstrated by synthesis of a derived bis-chloroethylamine alkylating system and a hybrid DNA interactive enediyne system.

Introduction

Research on the development of nonpeptide based DNA interactive drugs has been intense, since such agents offer the potential to interact with their intended target without falling hostage to cellular peptidases.[1] Of the established drug:DNA associative methods, intercalation is one of the most predictable, and a variety of natural and synthetic agents which function using this binding pathway show excellent antitumor activity. Pertinent examples include the anthracyclines such as doxorubicin **1**,[2] the acridines e.g. amsacrine **2**,[3] and the



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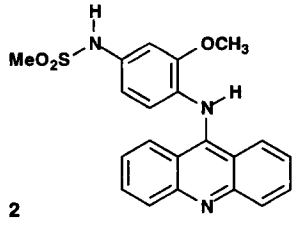

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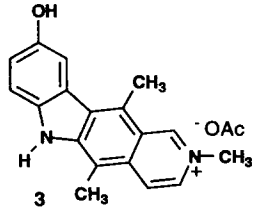

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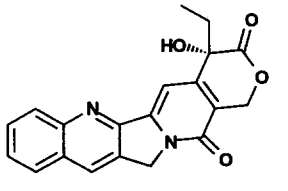

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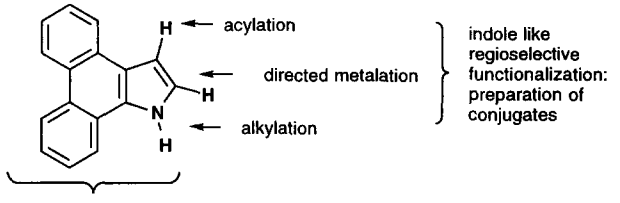
ellipticines such as elliptinium acetate **3**.[4] The driving force for intercalation, which occurs preferentially into Py-3',5'Pu, sequences rather than Pu-3',5'Py, relies on binding energy derived from removal of the agent from aqueous medium, together with Van der Waals forces between the agent and DNA .[5] Such agents may interact with DNA by a variety of additional pathways, and are often found to be targets for the regulatory enzymes topoisomerase I and II. Certain antitumor agents appear enzyme specific, notably camptothecin **4**, which is a target of mammalian topoisomerase I,[6] wheras agents **1-3** all affect topoisomerase II. The precise role such intercalative agents play in modulating the regulatory enzymes is still debated, but likely involves formation of drug:DNA:enzyme ternary complexes.[7] The unwinding of the DNA helix associated with intercalative binding of these antitumor agents may indeed facilitate binding with the topoisomerases, and assist in formation of derived ternary complexes. Since intercalation represents a reversible vehicle to deliver a DNA active agent to its target, numerous systems have been designed to harness this feature in the form of antiproliferative agents.[1] As studies herein describe, we have designed and synthesized a novel heterocyclic intercalative delivery vehicle, and report on its effectiveness in the form of bifunctional DNA interactive hybrids.

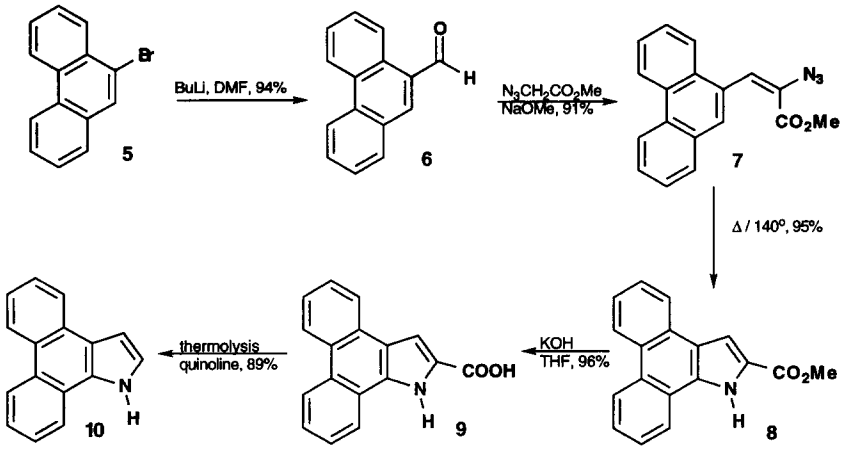
Our synthetic efforts were guided by two principle requirements: (i) to achieve effective intercalation, fused polyaromatic systems of 4-6 rings are desirable and (ii) in order to perform subsequent SAR correlations within a family of intercalative vehicles, regioselective functionalization of the polycyclic core would be advantageous. For these purposes, we envisaged that pyrrolo[9,10-*b*]phenanthrene would represent an ideal template, since classical indole type chemistry would be available for elaboration of the indolyl *N*, C-2, and C3 positions (Figure 1).[8]

Figure 1. Design features of the proposed intercalative hybrids



The original synthesis of the pyrrolophenanthrene template was conducted as shown in Scheme 1. Formylation via metal halogen exchange of bromide **5** proved an economical route to commercially available aldehyde **6**, and with this in hand, a Rees-Moody protocol was used to form azidocinnamate **7**, which in turn underwent smooth thermolysis to give tetracyclic ester **8** *via* the corresponding nitrene insertion reaction.[9] Since we eventually required unsubstituted tetracyle **10**, ester **8** was then converted to acid **9** and means to remove the carboxyl function investigated. Several procedures have been reported to effect this often problematic transformation in the indole-2-carboxylate series, with widely ranging degrees of efficiency. Problems encountered during the process usually stem from decomposition of the product under prolonged thermolysis conditions, and additional decomposition during purification of the crude product. Based on our earlier successes with microwave accelerated transformations,[10-11] including the decarboxylation of substituted indoles,[12] acid **9** was thermolyzed in a sealed vessel containing quinoline using microwave irradiation, which gave **10** cleanly and in 93% yield, after only 10 mins heating. With quantities of **10** in hand, we began to optimize conventional methods of thermolysis, and eventually found conditions which succeeded in producing **10** in 89% yield on a preparative scale (Table 1). Encouraged by this decarboxylation reaction we became interested in the possibility

Scheme 1. Synthesis of the pyrrolo[9,10]phenanthrene nucleus



of a one pot route to the parent ring system. Accordingly, ester **7** was hydrolyzed to acid **11** (Scheme 2) then subjected to both conventional thermolysis, and microwave irradiation.This indeed resulted in tandem nitrene insertion-decarboxylation to yield parent tetracycle **10** (and varying amounts of the intermediate **9**) in *one pot*. The results obtained using various conditions (Table 1) indicate the utility of this approach, which proceeds well either using conventional thermolysis conditions in quinoline (in the presence of a copper chromite catalyst) or microwave heating in the absence of catalyst. Future application in the synthesis of indoles and related benzo-fused systems would appear likely.

Scheme 2. Tandem nitrene insertion - decarboxylation route to **10**

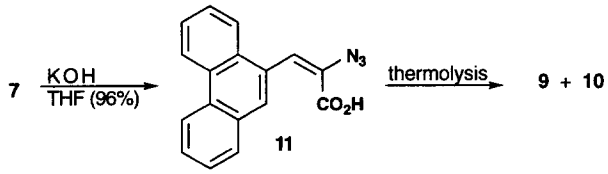


Table 1. Decarboxylative and Tandem Insertion-Decarboxylative Routes to **10**

substrate	solvent	thermolysis	method	ratio **9:10**	% yield **10**
9	quinoline	10 mins	**a**	-	**93%**
9	quinoline*	10 mins	a	-	90%
9	quinoline	1h	c	-	46%
9	quinoline*	1h	**c**	-	**89%**
11	quinoline	12 mins	**a**	0:100	**81%**
11	*o* dichlorobenzene	12 mins	a	20:80	34%
11	quinoline	12 mins	b	0:100	48%
11	quinoline*	1h	c	0:100	63%

Thermolysis methods: (a) microwave irradiation (650W), sealed tube; (b) microwave irradiation (650W), atmospheric pressure; (c) conventional heating (silicon oil bath). *denotes copper chromite catalyst added.

Biological Evaluation

Since the primary function of **10** was to serve as an intercalative delivery vehicle, a series of agent-DNA interactions were investigated to determine its effectiveness in this role. In the uv spectral analysis of a mixture of **10** and calf thymus DNA, characteristic red shifts of 10 nM at 290 nM for DNA were observed on incubation consistent with intercalation. [13] Furthermore, incubation followed by electrophoretic examination of a mixture of **10** + ΦX174 Rf I DNA showed characteristic streaking of the supercoiled species, indicative of intercalation (Figure 2, lanes 4-5). [14]

lane 1 2 3 4 5

Figure 2. DNA intercalative streaking

ΦX174 supercoiled (Type I) DNA incubated with agent for 18 h at 24oC in 10 mmol Tris-HCl, 1 mmol EDTA, pH 8.0 (total volume 6 μl) followed by gel electrophoresis (1% agarose, ethidium bromide stain). Lane 1 = DNA (50 μmol); Lane 2= DNA (50 μmol) + ethidium bromide (50 μM); Lanes 3-5 = DNA (50, 100, 150 μmol respectively) + **10** (50μmol).

Additionally, since conversion of type I to type II DNA, accelerated by the regulatory enzyme topo I can be surpressed in the presence of intercalators and poisons, we determined the effectiveness of **10** in this role. [15] Incubation of DNA with topo I in the presence of **10** did indeed result in inhibition of DNA relaxation at 250 μmol (Figure 3, lane 4), supporting the intercalative theory. Though not as efficient as ethidium bromide in this capacity (c.f. lanes 6-8) the results of this experiment, combined with the observed drug:DNA uv shifts and the unwinding experiments in Figure 2 suggest that the pyrrolo[9,10-*b*]phenanthrene nucleus interacts with DNA via an associative method.

lane 1 2 3 4 5 6 7 8

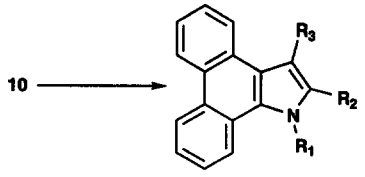
Figure 3 Topoisomerase I Inhibition assay

ΦX174 DNA (Type I) incubated (30 min, 24oC) in Tris-HCl at pH 7.5 with agents (total volume 7.3 μl) followed by gel electrophoresis (1% agarose, ethidium bromide stain). Lane 1 = DNA (50 μmol); Lane 2 = DNA (50 μmol) - topoisomerase I (1 μl: 8 units / μl); Lanes 3-5 = DNA (50 μmol), topoisomerase I (1 μl: 8 units / μl) and 50, 250 and 100 μmol of **10** respectively. Lanes 6-8 = DNA (50 μmol), topoisomerase I (1 μl: 8 units / μl) and 50, 250 and 100 μmol of ethidium bromide.

Functionalization

With an efficient synthesis of **10** secure, we wished to demonstrate its versatility as a template for the construction of derivatives which, as shown in Scheme 3, proved trivial. Formation of the N-sodium salt allowed N-alkylation to give **12** in good yield. Based on classical pyrrole and indole chemistry, electrophilic addition would be predicted to take place at the indolyl C3 position, and as expected the product of Vilsmeier formylation **13** was formed in high yield. Conversion to the N-phenylsulfonyl derivative **14** also allows regioselective (pyrrolyl C2) metalation to ensue, giving access to 2-substituted products (Scheme 3).[16-17] Alternatively, the procedures of Katritzky could be used to functionalize C2, involving intermediate N-protection, followed by regioselective metalation, giving **15-16** and carboxylates **8-9** (Scheme 3).[18] Additionally, derivatives **15** and **16** are available via reduction then re-oxidation of ester **8** (see experimental section).

Scheme 3. Regioselective functionalization of pyrrolo[9,10-*b*]phenanthrene



	R_1	R_2	R_3	reagents
12	CH_3	H	H	a
13	H	H	CHO	b
14	SO_2Ph	H	H	c
15	H	CH_2OH	H	d
16	H	CHO	H	e
8	H	CO_2CH_3	H	f
9	H	CO_2H	H	g

Reagents: (a) NaH, DMF then CH_3I (93%) (b) $POCl_3$, DMF (79%) (c) *n*BuLi then $PhSO_2Cl$ (99%) (d) *n*BuLi, CO_2, then *t*BuLi, $(CH_2O)_n$ then Δ (44%) (e) *n*BuLi, CO_2, then *t*BuLi, DMF, then Δ (55%) (f) *n*BuLi, CO_2, then *t*BuLi, CH_3OCOCl, then Δ (63%) (g) as f, then LiOH, THF (95%) or *n*BuLi, CO_2,then *t*BuLi, added to CO_2, then Δ (43%).

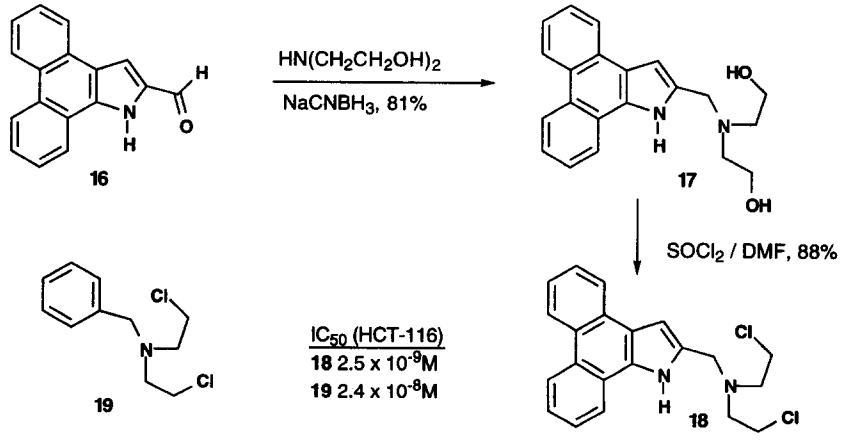
Bioactive Hybrids

Having ascertained the intercalative ability of the pyrrolophenanthrene nucleus, we sought to demonstrate practical application of such a delivery vehicle. Since three of the main classes of drug-DNA interactions are intercalation, alkylation and strand scission respectively, we wished to *combine* two such modes of interaction, and synthesize hybrid structures based on **10**, that may potentially demonstrate *synergistic enhancement in DNA interactive ability.*

(i) Mustard Agents

We firstly investigated a potential bis alkylative hybrid containing a β–chloroethyl mustard system *viz.* **18**. A number of bisalkylative nitrogen mustard agents are currently under investigation as potential antineoplastic agents, a result of both their high reactivity and the presumed biological consequences of DNA cross linking.[19-20] Such agents allow single and double DNA alkylation via aziridinium ion formation and subsequent nucleophilic attack by a proximal base, often encompassing both inter and intrastrand alkylation. Synthesis of mustard **18** was accomplished using aldehyde **16** as a building block (Scheme 4). Reductive amination of **16**

Scheme 4. Synthesis of a hybrid pyrrolo[9,10]phenanthrene mustard agent

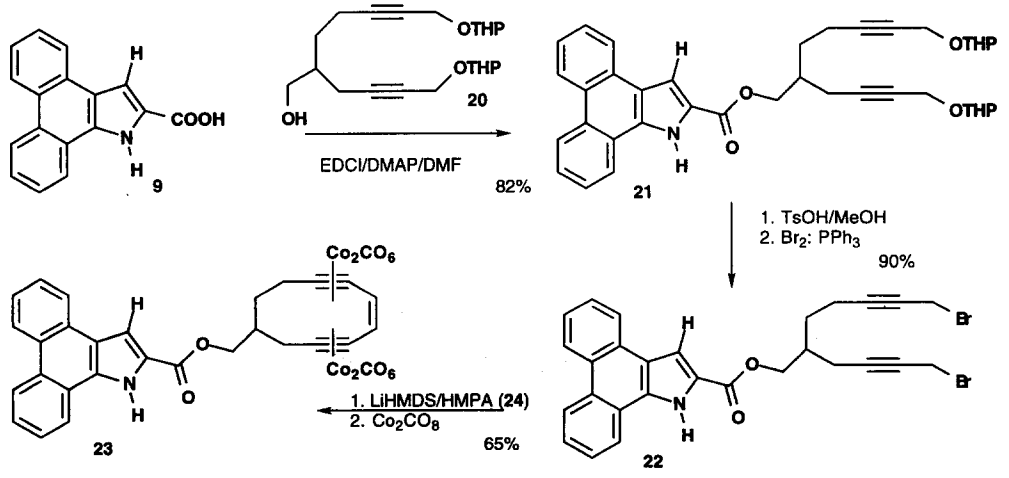


with diethanolamine, using sodium cyanoborohydride resulted in a good yield of the corresponding diol **17**, which was immediately converted to the dichloro mustard agent **18** using thionyl chloride (88%). Evaluation of the antiproliferative activity of **18** confirmed the potency of the bis chloroethylamine appendage, with good activity in the human colon tumor assay employed.[21] Comparison with the observed IC_{50} values for related mustard systems e.g **19** supported the design strategy behind the bifunctional system and the inclusion of the additional intercalative moiety.

(ii) Enediynes

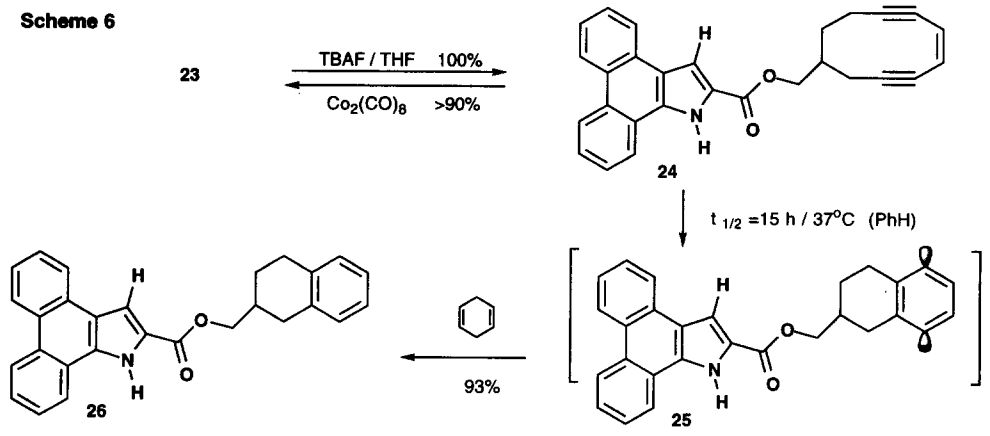
Using an entirely different approach, a hybrid intercalator-enediyne **24** was then prepared (Schemes 5, 6). As a class, the enediynes are receiving unprecedented attention due to their ability to cause diyl mediated DNA strand scission, particularly those which undergo the requisite Bergman cycloaromatization reaction under controlled conditions.[22] A number of hybrid enediyne-DNA binding systems have been reported recently, where a pre-formed ten-membered cyclic enediyne is attached covalently to a discrete DNA interactive agent.[23-29] Our interest in the synthesis of enediynes stems from a recently discovered carbenoid coupling strategy, which gives access to linear and cyclic enediynes in high yield under mild conditions.[30-32] Accordingly, ester **24** was initially identified, and prepared as shown in Scheme 5. Since the protected diyne **20** is available to us in multigram quantities,[33] esterification of this synthon with the acid **9** was conducted using the conditions of Boger,[24] giving the expected diyne ester **21**. The propargyl groups were then unmasked and brominated to give **22**, then finally converted into the desired enediyne using the carbenoid coupling-elimination method. This intramolecular route to the enediyne core is noteworthy for its mildness and tolerance of pendant functionality, and in this instance it was found unnecessary to protect the NH function. For practical purposes, the product enediyne was isolated as its bis-cobalt carbonyl complex **23**, providing a shelf-stable form of the enediyne, which could be unmasked at will to regenerate **24** using TBAF in THF (Scheme 6).

Scheme 5. Coupling of enediyne precursor to intercalative template and carbenoid cyclization



Deprotection of **23** followed by Bergman cycloaromatization of **24** was conducted at physiological temperature in the presence of the atom transfer agent 1,4-cyclohexadiene (30 equiv.), which with a half-life of approximately 15 h, gave adduct **26** in high yield, presumably *via* the 1,4 diyl intermediate **25** (Scheme 6).

Scheme 6



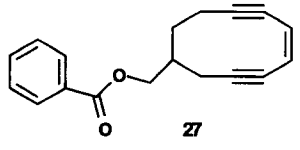
DNA Cleavage

The ability of conjugate **24** to induce DNA strand scission was evaluated using supercoiled bacteriophage (ΦX174 RfI) DNA. Lesions were evident at agent concentrations as low as 10^{-6} M, however significant strand scission did not occur until 10^{-4} M (Table 1). The predominant events involved single stranded cutting, with the onset of random single stranded cutting events at higher concentration (10^{-3} M). The additional benefit of the intercalative group was confirmed by comparison of the cleavage induced by enediyne benzoate **27**, which would not be expected to participate in intercalation to the same extent.[32]

Table 1. DNA Cleavage Induced by Enediyne Hybrid **24** and Enediyne **27**

		24				**27**			
DNA	control	10^{-6}	10^{-5}	10^{-4}	10^{-3}	10^{-6}	10^{-5}	10^{-4}	10^{-3}
RF I	95%	85%	79%	45%	7%	94%	93%	78%	15%
RF II	5%	15%	21%	55%	88%	6%	7%	22%	82%
RF III	0%	0%	0%	0%	5%	0%	0%	0%	3%

Conditions: ΦX174 RfI DNA (50 μM in base pairs) was incubated in TRIS-HCl at pH 8.5 as control or with **24/27** at indicated concentrations (M) for 12 h at 37 °C. The crude mixtures were subjected to gel electrophoresis (10% agarose), stained with ethidium bromide, then subjected to scanning densitometry.



IC_{50} (HCT-116)

24 2.4 x 10^{-5}M

27 5.2 x 10^{-4}M

Though only a minor improvement in strand scission capacity was observed for the heterocyclic conjugate, the antitumor activity of **24** was an order of magnitude greater than the control substrate **27**.[34] This difference may reflect the fact that DNA is only one of several possible cellular targets for enediynes and the diyl radicals they generate on cycloaromatization. Another potential target are the nuclear proteins.[35] Both the naturally occurring kedarcidin and the related enediyne neocarzinostatin are chromoproteins that are capable of

causing protein damage to histones.[36] The Bristol-Myers-Squibb group has also described the protein modulating activity (10^{-4} M) of a synthetic ten-membered enediyne.[29,37] Accordingly, the proteolytic activity of **24** was assessed using the (DNA protective) histones II-S, II-AS, and III-S, but evidence of agglomeration was not detectable until $>10^{-3}$ M, suggesting the primary target of the pyrrolo[9,10-*b*]phenanthrene hybrid may indeed be DNA, as originally predicted.

Conclusions

A new class of intercalative delivery vehicle - the pyrrolo[9,10-*b*]phenanthrenes have been synthesized. The parent tetracycle **10** is prepared in three steps from commercially available arene, undergoes regioselective functionalization readily, and intercalation to DNA has been demonstrated. The utility of this template in the synthesis of hybrid structures designed to capitalize on its intercalative properties has been examined. Further application of this template in antitumor agent design would seem warranted, particularly where systematic structural variations are required.

Experimental Procedures

Unless stated otherwise, all reactions described herein were performed in glassware which had been oven dried (140°C / 12h) then flame dried prior to use. Reactions were conducted under an atmosphere of nitrogen, with flasks sealed using dried septa (P_2O_5). The tips of cannulae were flame dried under a stream of dry nitrogen gas prior to use. THF and diethyl ether were distilled immediately prior to use from sodium / benzophenone ketyl. Methylene chloride was distilled from P_2O_5. All other reagents and solvents were commercial grade, and purified according to standard convention. ΦX174 RfI DNA was purchased from New England Biolabs and topoisomerase I from Sigma. Control substrates **19**[38] and **27**[32] were prepared using reported methods. Silica gel chromatography was perfomed on E. Merck 70-240 mesh gel. Analytical t.l.c. was performed on glass backed 250μ plates visualizing with anisaldehyde and phosphomolybdic acid. ^{1}H spectra were recorded at 300 MHz and ^{13}C spectra at 75 MHz, both on a Bruker AM 300 instrument. Mass spectra were recorded on either a Fisons Trio 1000, or VG-7070 instrument. Microanalyses were performed at Atlantic Microlab, Norcross, GA. Antitumor assays were conducted using reported methods.[39]

Phenanthrene-9-carboxaldehyde (**6**).

To a solution of 9-bromophenanthrene (6.00 g, 23.33 mmol) in dry THF (350 mL) at -78°C was added *n*BuLi (30.1 mL, 46.66 mmol) over a period of 30 min. The solution was stirred at -78°C for 30 min. and then maintained at 0-4°C for an additional 45 min. The solution was returned to -78°C and dry DMF (100 mL) was added dropwise, and the mixture allowed to warm to room temperature over a 12h period. After concentration *in vacuo*, the product was recrystalized from ethyl acetate and hexane to give **6** (4.54g, 94%) as an off white solid, m.p. 101-103 °C (Lit[40] 100-103 °C).

Methyl 2-azido-3-(9-phenanthryl)propenoate (**7**).

To a stirred solution of sodium methoxide (2.68 g, 116.4 mmol Na in dry methanol, 50 mL), at -7°C was added **6** (6.00 g, 29.1 mmol) and methylazidoacetate[9] (13.4 g, 116.4 mmol) in a solution of dry methanol (200 mL) and dry THF (8 mL) at -7°C over a period of 2h. The solution was allowed to stir at -7°C for an additional 2h. and then placed at +4°C for 12h. The mixture was poured over H_2O (300 mL) and extracted with ethyl

acetate (3 x 100 mL), the combined extracts were washed with H_2O (2 x 200 mL) and brine (1 x 150 mL), then dried with Na_2SO_4. After concentration *in vacuo*, the crude product was recrystalized from ethyl acetate and hexane to give **7** (8.07 g, 91%) as a pale yellow solid, m.p. 115-118 °C ; ^{1}H NMR ($CDCl_3$) δ 8.73 (d, 1H, J=1.39 Hz), 8.66 (d, 1H, J=1.39 Hz), 8.33 (d, 1H, J=1.39 Hz), 7.93 (d, 1H, J=1.35 Hz), 7.72-7.61 (m, 4H), 7.26 (s, 1H), 4.01 (s, 3H, OCH_3); ^{13}C NMR ($CDCl_3$) δ 53.0, 122.5, 122.8, 123.2, 124.2, 126.7, 126.9, 127.6, 127.9, 129.4, 129.6, 130.3, 130.4, 130.6, 130.9, 163.8; IR (neat) ν_{max} 2931, 2863, 2115, 1702, 1474, 1389, 1302, 1248, 1097, 714 cm-1; MS m/e (EI) 303 (M+); $C_{18}H_{13}N_3O_2$ requires (C, 71.28; H, 4.32; N, 13.85) found (C, 71.06; H. 4.59; N, 13.54).

Methyl 4,5 (9,10-phenanthryl)pyrrole-2-carboxylate (**8**) .

3.4 g (12.36 mmol) of **7** in dry xylene (130 mL) was added dropwise to refluxing xylene (340 mL) *via* a pressure equalizing dropping funnel. After addition was complete the solution was allowed to stir at reflux for 2h. The mixture was cooled and the solvent was removed *in vacuo*. The product was recrystalized from xylene to give **8**, (2.94 g, 95%) as a white solid, m.p. >300 °C; ^{1}H NMR ($CDCl_3$) δ 3.91 (s, 3H, -CH_3), 7.60 (m, 4H,), 7.85 (s, 1H), 8.34 (d, 1H, J=1.2,), 8.77 (m, 3H), 11.15 (s, 1H, -NH); ^{13}C NMR (D_6-DMSO) δ 51.6, 108.9, 120.3, 122.6, 123.0, 123.5, 123.6, 123.8, 124.9, 126.3, 127.0, 127.3, 128.0, 129.0, 161.4; IR (neat) ν_{max} 3266, 2920, 1947, 1678, 1496, 1443, 1420, 1297, 1144, 1004, 758, cm-1; MS m/e (EI) 275 (M+); $C_{18}H_{13}NO_2$ requires (C, 78.53; H, 4.76; N, 5.09) found (C, 78.69; H. 4.84; N, 4.87).

4,5- (9,10-phenanthryl)pyrrole-2-carboxylic acid (**9**) .

To a solution of **8** (1.5 g, 5.45 mmol) in THF (20 mL) and H_2O (6 mL), was added KOH (2 M, 4.5 mL) and the solution was heated to 40°C and stirred for 3h. The THF was removed *in vacuo* and the residue was brought to pH2, saturated with NaCl, extracted with ethyl acetate (5 x 30 mL), and dried with Na_2SO_4. Removal of solvent gave **9** (1.37 g, 96%) as an off white solid, m.p. 205-207 °C; ^{1}H NMR ($CDCl_3$) δ 7.59 (m, 4H), 7.76 (s, 1H), 8.32 (d, 1H, J=1.1), 8.76 (m, 3H), 11.29 (s, 1H, -NH); IR (neat) ν_{max} 3419, 2948, 1704, 1611, 1501, 1468, 1371, 1198, 1120, 1076, 986 cm-1; MS m/e (EI) 261 (M+); $C_{17}H_{11}NO_2$ requires (C, 78.15; H, 4.24; N, 5.36) found (C, 78.36; H. 4.44; N, 5.28).

2,3-Pyrrolo[9,10]phenanthrene (**10**) .

(a) Conventional thermolysis of **9**

In a flask equipped with a reflux condensor was placed **9** (0.35 g, 1.34 mmol), copper chromite (BaO promoted 0.228 g), and redistilled quinoline (3.5 mL) added. The flask was lowered into a silicon oil bath preheated to 220°C and a device attached to monitor evolution of CO_2. After CO_2 liberation ceased, the mixture was poured onto cold H_2O (5 mL) and brought to pH4 with concentrared HCl. The mixture was extracted with ethyl acetate (5 x 20 mL), and the combined extracts washed with 10% HCl (5 x 50mL), saturated $NaHCO_3$ (2 x 50 mL), H_2O (3 x 50 mL) and dried with Na_2SO_4. The product was purified by flash chromatography using a mixture of (2:8) ethyl acetate:hexane as eluent to yield **10** (0.258 g, 89 %) as a dark blue solid, m.p. 165-167°C; ^{1}H NMR ($CDCl_3$) δ 7.07 (m, 1H), 7.50-7.64 (m, 3H), 7.96 (d, 1H, J=4.5 Hz), 8.23 (d, 1H, J=4.5 Hz), 8.65-8.72 (m, 2H), 9.05 (br s, 1H); ^{13}C NMR ($CDCl_3$) 120.0, 120.6, 121.0, 121.6, 123.3, 123.8, 123.9, 124.3, 126.4, 126.6, 127.7, 128.2, 128.6, 128.7, 129.2, 129.4; IR (neat) ν_{max} 3414, 11643, 1575 cm-1; MS m/e (EI) 217 (M+); $C_{16}H_{11}N$ requires (C, 88.45; H, 5.10; N, 6.45) found (C, 88.23; H. 5.24; N, 6.28).

(b) Microwave thermolysis of **9**

Acid **9** (0.040 g, 0.153 mmol) was placed in a thick walled glass tube (10 cm x 7 cm o.d.) and dissolved in redistilled quinoline (2 mL). The tube was sealed under vacuum, placed upright in the center of a 100 mL beaker and packed tight with vermiculite beads. The beaker was placed in a 650W domestic microwave oven and thermolyzed at full power for 12 min. then cooled to room temperature. The contents of the tube were extracted with ethyl acetate (2 x 5 mL) and the combined extracts washed with HCl (10%, 6 x 5 mL), NaOH (2 x 5 mL), brine (2 x 5 mL) then dried over Na_2SO_4. Removal of solvent *in vacuo* followed by recrystallization (hexane:ethyl acetate) gave **10** (0.031 g, 93%) identical to previously prepared samples.

(c) Microwave thermolysis of **11**

Using identical method to that outlined in (b) above, **11** (0.019 g, 0.066 mmol) was thermolyzed in quinoline (2 mL) for 12 mins to yield **10** (0.012 g, 81%) identical to previously prepared samples.

2-Azido-3-(9-phenanthryl)propenoic acid (**11**) .

To a stirred solution of **7** (1.0 g, 3.30 mmol) in THF (15 mL) and H_2O (6 mL) was added KOH (2M, 3.5 mL) dropwise. The mixture was allowed to stir for 4h at room temperature. The THF was then removed *in vacuo* and the residue brought to pH 4.5 - 5.0 with 10 % HCl, then saturated with solid NaCl, extracted with ethyl acetate (5 x 25 mL) and dried with Na_2SO_4. Removal of solvent gave **11**, (0.921 g, 96%) as a pale yellow solid, m.p. 117-120 °C; ^{1}H NMR ($CDCl_3$) δ 7.19 (s, 1H), 7.67 (m, 4H), 7.98 (d, 1H, J=0.78Hz), 8.06 (d, 1H, J=0.78Hz), 8.19 (s, 1H), 8.77 (d, 1H, J=0.78Hz), 8.85 (d, 1H, J=0.78Hz); ^{13}C NMR (D_6-DMSO) 122.8, 123.5, 124.3, 127.0, 127.2, 127.3, 127.8, 128.1, 128.9, 129.2, 129.7, 129.8, 130.6; IR (neat) ν_{max} 3402, 2111, 1581 cm^{-1}; MS m/e (EI) 289 (M+); $C_{17}H_{11}N_3O_2$ requires (C, 70.58; H, 3.83; N, 14.52) found (C, 70.31; H. 3.95; N, 14.26).

N-Methyl-2,3-pyrrolo[9,10]phenanthrene (**12**)

A solution of **10** (0.04 g, 0.184 mmol) in DMF (1.8 mL) was added dropwise to NaH (pentane washed, 0.005 g, 0.221 mmol) and stirred at room temperature for 1h. CH_3I (0.019 mL, 0.203 mmol) was added, and the solution of was allowed to stir at 25°C for 30 min. The mixture was poured onto ice (20 g) and extracted with ethyl acetate (3 x 20 mL). The combined extracts were washed with H_2O (4 x 35 mL), brine (1 x 25 mL) and dried with Na_2SO_4. Removal of solvent gave **12**, (0.0396 g, 93%) as a brown solid m.p. 170-173; ^{1}H NMR ($CDCl_3$) δ 7.02 (d, 1H, J=0.48 Hz), 7.04 (d, 1H, J=0.48 Hz), 7.6 (m, 4H), 8.24 (d, 1H, J=1.16 Hz), 8.46 (d, 1H, J=1.16Hz), 8.68 (d, 1H, J=1.16 Hz), (d, 1H, J=1.16 Hz) ; ^{13}C NMR ($CDCl_3$) δ 38.8, 100.5, 120.9, 122.6, 123.0, 123.2, 123.9, 124.0, 124.1, 126.2, 126.7, 126.8, 128.3, 128.5, 128.7, 128.8; IR (neat) ν_{max} 3409, 1651, 1609, 1454 1370, 904 cm^{-1}; MS (EI) m/e 231 (M+); HRMS: $C_{17}H_{13}N$ calcd: 231.1048; found: 231.1046.

4,5-Pyrrolo[9,10]phenanthryl-3-carboxaldehyde (**13**)

Freshly distilled $POCl_3$ (0.007 mL, 0.014mmol) was added dropwise to a stirred solution of DMF (0.015 mL) at 10-20°C. A solution of **10** (0.008g, 0.037 mmol) in DMF (0.011 mL, 0.16 mmol) was added dropwise to the mixture at room temperature, and stirred for 1h. NaOH (0.24 M, 0.069 mL) was added slowly so that the

pH remained <7. The solution was then heated to boiling point for 1 min. Upon cooling the product precipitated, and the mixture was filtered and rinsed carefully with ice-water to give **13** (0.007 g, 79%) as a brown solid. m.p. >300 °C ; ^{1}H NMR ($CDCl_3$) δ 7.68 (m, 4H), 7.98 (s, 1H), 8.35 (d, 4H), 8.76 (m, 3H), 9.87 (s, 1H, -CHO), 11.68 (s, 1H, -NH); ^{13}C NMR (D_6-Acetone) δ 115.1, 120.9, 122.3, 122.6, 123.4, 123.7, 123.9, 124.3, 124.5, 124.6, 124.9, 127.7, 128.4, 129.3, 133.6, 133.9, 187.1; IR (neat) ν_{max} 3192, 2818, 1721 cm^{-1}; MS (EI) 245 (M+); HRMS: $C_{17}H_{11}NO$ calcd: 245.0841; found: 245.0843.

N-Phenylsulfonyl-2,3-Pyrrolo[9,10]phenanthrene (**14**)

n-BuLi (2.66 M, 0.092 mL, 0.202 mmol) was added dropwise to a stirred solution of **10** (0.04 g, 0.184 mmol) in THF (1.3 mL) at -78°C. The mixture was allowed to warm to room temperature over the period of 1h and then returned to -78°C for the addition of $PhSO_2Cl$ (0.04 mL, 0.202 mmol). The solution was stirred at -78°C for 2h and then allowed to warm to room temperature slowly. The THF was removed *in vacuo* and $NaHCO_3$ (5%, 15 mL) was added, and the mixture extracted with ethyl acetate (3 x 15 mL). The organic extracts were washed with H_2O (2 x 15 mL), brine (1 x 10 mL), then dried with Na_2SO_4 and concentrated *in vacuo* to give **14** (0.077 g, 99%) as a brown oil. ^{1}H NMR ($CDCl_3$) δ 7.27 (d, 1H,J=1.1 Hz), 7.30 (d, 1H, J=1.1 Hz), 7.62 (m, 6H) 7.98 (d, 1H, J=1.3 Hz), 8.03 (d, 2H, J=1.3 Hz), 8.14 (d, 1H, J=1.3 Hz), 8.64 (m, 2H), 9.10 (d,1H, J=1.3 Hz); ^{13}C NMR ($CDCl_3$) 123.1, 123.3, 123.4, 123.5, 123.7, 124.0, 124.6, 125.4, 126.1, 126.4, 126.6, 126.7, 126.8, 126.9, 127.0, 127.4, 127.7, 128.6, 129.1, 129.2, 129.6, 129.7; MS (EI) 357 (M+); HRMS: $C_{22}H_{15}NO_2S$ calcd: 357.0823; found: 357.0826.

4,5-pyrrolo-(2-hydroxymethyl)[9,10]phenanthrene (**15**)

To a stirred solution of LAH (1.54 g., 40.49 mmol) in dry THF (150 mL) was added **8** (2.5 g,10.12 mmol) in dry THF (240 mL) dropwise. The mixture was stirred at room temperature for 6h. The reaction mixture was quenched with the dropwise addition of 1% HCl and the mixture was extracted with ethyl acetate (3 x 40 mL). The combined extracts were washed with H_2O (2 x 35 mL), brine (1 x 30 mL) , dried with Na_2SO_4, then concentrated *in vacuo* to give 4,5-pyrrolo-(2-hydroxymethyl)[9,10]phenanthrene **15** (2.45 g, 98%) as an off white solid m.p. >300 °C ; ^{1}H NMR (D_6-Acetone) δ 4.24 (d, 2H, J=0.29 Hz,-CH_2OH), 6.96 (s, 1H,), 7.56 (m, 4H), 8.22 (d, 1H, J=0.81 Hz), 8.40 (d, 1H, J=0.81 Hz), 8.77 (t, 2H, J=0.96 Hz), 11.18 (s, 1H, -NH); ^{13}C NMR (D_6-DMSO) δ 56.7, 100.3, 120.0, 120.9, 123.1, 123.5, 123.6, 123.7, 124.1, 126.6, 137.7; IR (neat) ν_{max} 3416, 1651, 1377 cm-1; MS m/e (EI) 247 (M+); $C_{17}H_{13}NO$ requires (C, 82.57; H, 5.30; N, 5.67) found (C, 82.73; H. 5.51; N, 5.46).

4,5-Pyrrolo[9,10]phenanthrene-2-carboxaldehyde (**16**)

Alcohol **15** (0.37 g, 1.49 mmol) was dissolved in methylene chloride (40 mL), and, with rigorous stirring at room temperature, MnO_2 (2.2 g, 25.32 mmol) was added over a period of 30 minutes. The mixture was filtered through celite, and the residue washed with hot methylene chloride (500 mL). The solvent was removed *in vacuo* to give **16** as a tan solid. The residual MnO_2 was placed in a soxhlet apparatus overnight and extracted with methylene chloride to recover additional product. The total yield of recovered solid **16** was 0.351 g (94%). m.p. 181-183 °C; ^{1}H NMR (D_6-DMSO) δ 7.68 (m, 4H), 7.96 (s, 1H), 8.37 (d, 1H, J=0.96 Hz), 9.33 (m, 3H), 9.87 (s, 1H, -CHO); ^{13}C NMR (D_6-DMSO) δ 114.2, 120.7, 122.7, 122.8, 123.3, 123.6, 123.8, 125.1, 126.7, 126.9, 127.1, 127.3, 127.8, 129.7, 133.4, 134.9, 180.9; IR (neat) ν_{max} 3406, 2822,

1719 cm-1; MS m/e (EI) 245 (M+); $C_{17}H_{11}NO$ requires (C, 83.24; H, 4.52; N, 5.71) found (C, 83.41; H. 4.66; N, 5.46).

Preparation of 4,5- pyrrolo-(2-hydroxymethyl)[9,10]phenanthrene (15) via directed metalation

The following procedure is general: 2,3-Pyrrolo[9,10-*b*]phenanthrene (**10**) (0.05 g, 0.22 mmol) was placed in a flame dried 50 ml round bottom flask, dissolved in dry THF (25 mL) and the solution brought to -78°C. *n*-Butyllithium (2.26 M, 0.11 ml, 0.253 mmol) was added dropwise and the mixture was then allowed to warm to rt over 20 min. at which time CO_2 was pumped into the solution for 10 min. followed by N_2 for an additional 10 min. The solvent was then removed *in vacuo* and the remaining solid placed on the vacuum pump for 20 min. Dry THF (25 ml) was added and the solution was brought to -78°C; *t*-BuLi (10.0 M, 0.025 ml, 0.253 mmol) was then added dropwise. The solution was warmed to - 20°C and stirred for 1h and then brought to -78°C upon which $(CH_2O)_n$ (1.00 g) was added, and stirred at -78°C for 3h at which time the mixture was quenched with sat. NH_4Cl until a precipitate formed, brought to 0°C and H_2SO_4 added (2N)until pH4 was obtained. The solution was poured into sat. NH_4Cl (50 ml), solid NaCl was added, the mixture was extracted with EtOAc (5 X 20 ml) and then the solvent was removed *in vacuo*. The resulting brown solid was heated with a heat gun until the production of CO_2 ceased. The solid was then recrystalized from Et_2O/hexane to afford **15** (0.025 g, 44%) as a white solid, idendical to a previously prepared sample. Similarly, aldehyde **16**, ester **8** and acid **9** were prepared from **10** by the addition of DMF, methylchloroformate and (inverse addition *to*) carbon dioxide respectively.

4,5-Pyrrolo[9,10]phenanthrene diethanolobenzylamine (**17**)

To a solution of diethanolamine (0.03 g, 0.26 mmol), in methanol (2 mL) was added HCl-CH_3OH (5N, 0.16 mL), followed by **16** (0.10 g, 0.408 mmol) and $NaCNBH_3$ (0.018 g, 0.286 mmol). The solution was stirred at room temperature for 72h. The CH_3OH was removed *in vacuo*, then residue brought to pH< 2, and H_2O added (15 mL). The mixture was then extracted with CH_2Cl_2 (3 x 15 mL). The aqueous phase was brought to pH > 10 with solid KOH, then saturated with NaCl , extracted with CH_2Cl_2 (2 x 15 mL), dried with Na_2SO_4 and then the solvent removed *in vacuo* to give the corresponding diethanolamine **17** (0.193 g, 81%) as a tacky tan solid. ^1H NMR ($CDCl_3$) δ 2.74 (t,4H, J=0.5 Hz,-$N(CH_2)_2$), 3.75 (t, 4H, J=0.5 Hz,–CH_2OH), 3.85 (s, 2H,-CH_2N), 6.78 (s, 1H), 7.03 (t,1H, J=0.81 Hz), 7.09 (t,1H, J=0.81 Hz), 7.49 (t, 1H, J=0.81 Hz), 7.57 (t,1H, J=0.80 Hz), 7.69 (d, 1H, J=0.81 Hz), 8.08 (d, 1H, J=0.80 Hz), 8.24 (d, 1H, J=0.80 Hz), 8.49 (d, 1H, J=0.80 Hz); ^{13}C NMR ($CDCl_3$) δ 52.6, 56.0, 59.4, 102.1, 119.6, 123.1, 123.2, 123.3, 123.5, 123.8, 125.8, 125.3, 126.9, 128.6, 129.3; $C_{21}H_{22}N_2O_2$ requires (C, 75.43; H, 6.63; N, 8.38) found (C, 75.63; H. 6.48; N, 8.21).

4,5-Pyrrolo[9,10]phenanthrene dichloroethylbenzylamine (**18**)

Dry DMF (5 mL) was added to diethanolamine **17** (0.070 g, 0.210 mmol) while stirring at room temperature. The solution was placed in an ice bath, and freshly distilled $SOCl_2$ (0.12 mL, 1.682 mmol) was added dropwise to the solution, which was then stirred for 30 min. and partitioned between $CHCl_3$ (15 mL) and H_2O (15 mL). The organic layer was separated, washed with H_2O (3 x 15 mL), dried (Na_2SO_4) and concentrated *in vacuo*.

The residue was dissolved in HCl (1N, 10 mL) and the mixture was triturated with cold MeOH (2 mL) and the crystalline HCl salt collected by filtration, to give **18** (0.068 g, 88%) as a brown solid.

CAUTION: *This substance proved to be a potent skin irritant, and must be handled with extreme caution.* ^{1}H NMR (D_6-DMSO) δ 3.3 (br t, 4H, -CH_2N), 3.85 (br. t, 4H, -CH_2Cl), 4.13 (br. s, 2H, NCH_2-), 7.33 (br. s, 1H, C3-H), 7.62 (br. m, 4H), 7.94 (br. d, 2H), 8.40 (br. d, 1H), 8.76 (br. d, 1H).

4,5-Pyrrolo[9,10]phenanthrene-bis propargyl tetrahydropyranyl ether (21).
Acid (**9**) (1.20 g, 4.60 mmol) was added to a 25 ml round bottom flask followed by freshly distilled DMF (15 mL). EDCI (0.982 g, 5.12 mmol) was added, and the mixture was allowed to stir so that all the solid dissolved. DMAP (0.63 g, 5.12 mmol) was then added followed by **20** (2.52 g, 6.90 mmol)[33] and the resulting mixture was allowed to stir for 12h, poured into water (100 mL) and extracted with EtOAc (4 x 30 mL). The combined organic extracts were then treated with brine (1 x 50 mL), dried with Na_2SO_4 and concentrated *in vacuo*. The resulting oil was eluted through a column of silica gel (20:80 EtOAc:hexane) to give the corresponding ester **21** (2.28 g, 82 %) as a white gum. ^{1}H NMR (300 MHz, $CDCl_3$) δ 10.2 (s,1H), 8.65 (m, 3H), 8.23 (m, 1H), 7.70 (s, 1H), 7.61 (m, 4H), 4.8 (s, 2H), 4.45 (d, J = 5.7 Hz, 2H), 4.23 (m, 4H), 3.52 (m, 2H), 2.52 (m, 2H), 2.41 (m, 2H), 2.29 (m, 1H), 1.63 (m, 16H) ; ^{13}C NMR (75 MHz, $CDCl_3$) δ 161.8, 131.7, 129.8, 128.2, 127.3, 127.1, 126.7, 126.3, 124.8, 124.7, 123.8, 123.4, 123.3, 122.6, 121.2, 121.1, 109.0, 96.7, 96.6, 85.3, 83.0, 76.7, 76.5, 66.3, 61.9, 61.8, 54.5, 39.9, 33.3, 30.8, 29.2, 29.1, 25.2, 24.7, 20.6, 19.0, 16.4, 16.3 ; MS (EI) 607 (M+), 523 (100%). $C_{38}H_{41}NO_6$ requires (C, 75.10; H, 6.80; N, 2.30) found (C, 74.87; H. 6.55; N, 2.17).

4,5-Pyrrolo[9,10]phenanthrene-bis propargyl bromide (22).

The THP ether **21** (2.40 g, 3.95 mmol) was placed in a 100 ml round bottom flask and dissolved in a mixture of dry methanol (65 mL) and dry THF (10 mL). TsOH (0.1 g) was added, and the solution was allowed to stir at room temperature for 3h, at which time the solution was poured into half saturated brine (500 mL) and extracted with EtOAc (5 x 40 mL), washed with brine (1 x 40 mL) and dried with Na_2SO_4. The solvents were removed *in vacuo* and the remaining solid was eluted through a column of silica gel (40:60/EtOAc:hexane) to afford the corresponding diol (1.72 g, 99%) as a white solid. ^{1}H NMR (300 MHz, D_6-DMSO) δ 8.76 (m, 3H), 8.39 (d, J = 8.0 Hz, 1H), 7.88 (s, 1H), 7.57 (m, 4H), 4.33 (d, J = 4.8 Hz, 2H), 4.04 (t, J = 5.8 Hz, 4H), 2.35 (m, 2H), 2.14 (m, 1H), 1.68 (q, J = 7.0 Hz, 2H) ; ^{13}C NMR (75 MHz, D_6-DMSO) δ 160.96, 131.8, 129.0, 128.0, 127.2, 127.0, 126.8, 126.3, 125.1, 125.0, 124.9, 123.8, 123.6, 123.5, 122.9, 122.8, 122.6, 109.1, 83.6, 81.4, 65.4, 49.1, 49.0, 40.3, 40.0, 38.6, 29.0, 19.8, 15.6 ; IR (neat) 3318, 1693, 1491, 1282, 1188, 972, 724 ; MS (EI) m/e 439 (M+), 243 (100%), 244, 261, 215, 189.

The diol (1.09 g, 2.48 mmol) was dissolved in dry CH_2Cl_2 (25 mL). In a separate flask was placed dry CH_2Cl_2 (170 mL) and PPh_3 (1.36 g, 5.21 mmol), and the solution was placed in an ice bath, then bromine (0.27 ml, 0.83 g, 5.21 mmol) added dropwise and the resulting mixture allowed to sitr at 0°C for 30 min. The diol was then added dropwise to the PPh_3/Br_2 mixture at 0°C; upon completion of the addition of the diol, the solution was allowed to stir for an additional 15 min. The solvent was removed *in vacuo*, the resulting solid was preadsorbed onto silica, and the mixture columned through silica gel (20:80/EtOAc:hexane) to afford the

dibromide **22** (1.28 g, 91%) as a white solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 10.02 (s, 1H), 8.68 (m, 1H), 8.63 (d, J = 8.0 Hz, 1H), 8.23 (d, J = 8.0 Hz, 1H), 8.18 (m, 1H), 7.71 (d, J = 8.0 Hz, 2H), 7.64 (m, 3H), 4.44 (m, 2H), 3.92 (m, 4H), 2.53 (m, 2H), 2.45 (m, 2H), 2.29 (m, 1H), 1.80 (m, 2H) ; ^{13}C NMR (75 MHz, $CDCl_3$) δ 165.1, 129.9, 128.5, 127.4, 127.3, 126.9, 126.5, 125.0, 124.6, 124.0, 123.5, 123.4, 121.3, 120.9, 109.0, 86.8, 84.6, 76.5, 76.3, 66.1, 35.9, 29.0, 20.8, 16.6, 15.3, 15.1 ; MS (EI) m/e 563 (M+), 484, 81, 79; $C_{28}H_{23}Br_2NO_2$ requires (C, 59.49; H, 4.10; N, 2.48) found (C, 59.31; H. 3.93; N, 2.26).

Enediyne cobalt complex (23)

A 50 mL conical flask was charged with a solution of hexamethyldisilylazide (0.128 ml, 0.099 g) in THF (26 mL). The flask was immersed in an ice-acetone bath and 0.616 mmol of butyllithium (0.218 mL of 2.83M in hexanes) was added. In a 100 mL round bottom flask, the dibromide **22** (0.098 g, 0.176 mmol) and HMPA (0.77 mL, 0.79 g, 4.40 mmol) were dissolved in THF (20 mL) and the resulting solution was cooled to -45°C. Once the temperature had stabilized, the initially prepared solution of LiHMDS was added dropwise *via* mechanical syringe pump over the course of 5.0 h *(note - a steady dropwise addition is essential).* The reaction was monitored periodically by TLC throughout the base addition. On completion of the base addition, the reaction mixture was poured, *without warming*, onto a slurry of saturated NH_4Cl (50 mL) and crushed ice (10 g). The organic material was extracted into Et_2O (4 x 25 mL) and the combined ether extracts were further treated with cold 1% HCl (3 x 40 mL), followed by water (3 x 40 mL), saturated $NaHCO_3$, (1 x 25 mL), and brine (1 x 25 mL). The product solution was dried at 0°C over 4Å molecular sieves for 0.25 h. The resulting solution was transferred to a dry 250 mL round bottom flask. The flask was immersed in an ice bath and dicobalt octacarbonyl (0.150 g of 90% wt/wt in hexane, 0.439 mmol) was added. The resulting solution was allowed to warm slowly to room temperature and stir in the dark for 3h, after which the dark liquid was filtered through a plug of silica and concentrated *in vacuo*. Column chromatography (neutral alumina, hexane eluent) afforded the desired bis-complexed enediyne **23** (0.110 g, 65%) as dark green crystals. ^{1}H NMR (300 MHz, $CDCl_3$) δ 9.76 (s, 1H, -NH), 8.64 (m, 2H), 8.20 (d, J = 7.9 Hz, 1H), 8.08 (m, 1H), 7.61 (m, 4H), 7.42 (d, J = 7.5 Hz, 1H), 6.39 (s, 2H), 4.22 (t, J = 4.1 Hz, 2H), 3.26-3.62 (m, 4H), 2.27 (m, 1H), 2.05 (m, 2H) ; ^{13}C NMR (75 MHz, $CDCl_3$) δ 199.7, 167.5, 130.7, 130.4, 129.9, 129.0, 128.8, 128.3, 127.9, 127.6, 127.1, 126.9, 126.5 (HC=CH), 124.3, 124.0, 123.5, 122.9, 121.8, 121.4, 109.3, 96.8, 96.7, 83.7, 83.6, 67.0, 35.7, 29.2, 23.2, 14.2; MS (EI) m/e 403 (M-$[Co_2CO_6]_2$).

Deprotection of **23** *and Bergman cycloaromatization*

Bis-complexed enediyne **23** (23.0 mg, 0.024 mmol) was dissolved in THF (0.5 mL) and the resulting solution cooled to 0°C. With stirring at 0°C, was added TBAF (0.705 mmol of 1.0M in THF). The reaction mixture was allowed to stir for 15 min. then filtered through a plug of deactivated (Et_3N) silica, and the plug washed with Et_2O (10 mL). The resulting ethereal solution was shaken with water (2 x 10 mL), then dried over Na_2SO_4, filtered through a pad of celite, and concentrated *in vacuo*. The residue was taken up in benzene d_6 and quickly examined by ^{1}H NMR. After the initial ^{1}H NMR spectrum was obtained, 1,4-cyclohexadiene (0.07 mL, 0.705 mmol) was added and the tube was placed in a constant temperature bath and incubated at 37°C. The tube was removed from the bath and re-examined by ^{1}H NMR at 1.0 h. intervals. After 48 h of incubation at 37°C, the enediyne olefin signal could no longer be detected and the experiment was terminated. From this data, the $t_{1/2}$ for the enediyne was estimated to be in the range of 15 h. Concentration *in vacuo* gave

the tetrahydronaphthyl ester **26** (9.0 mg, 93%) as a brown oil. ^{1}H NMR (300 MHz, benzene d_6) δ 11.89 (s, 1H, -NH), 8.68-8.90 (m, 2H), 8.40 (d, J = 7.9 Hz, 1H), 7.87 (m, 1H), 7.49-7.77 (m, 8H), 7.30 (d, J = 7.5 Hz, 1H), 4.26-4.37 (m, 2H), 2.51-3.07 (m, 4H), 1.53-1.82 (m, 3H); MS (EI) m/e 405 (M+).

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