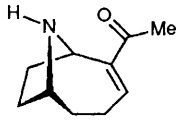
Synthetic and Stereochemical Studies Directed Towards Anatoxin-a

Nicholas J. S. Huby, Richard G. Kinsman, David Lathbury, Peter G. Vernon and Timothy Gallagher*
School of Chemistry, Bath University, Bath BA2 7AY, UK

The synthesis and stereocontrolled Ag^{I}-catalysed cyclisation of a series of allenic amino esters **8a–e** is described. For compounds **8a–d** the *cis*-2,5-disubstituted pyrrolidine **9** is formed exclusively but the primary amine **8e** undergoes cyclisation nonselectively to give a mixture of *cis*- and *trans*-pyrrolidines **9e** and **10e**. The synthetic potential of this allene-based methodology has been illustrated by the conversion of compound **8a** *via* **9a** into (±)-anatoxin-a **1** and the methoxy phosphine oxides **18** and **19** have been studied as ketone homologating agents within this context. The enzymatic resolution of (±)-**8a** using chymotrypsin is also described.

Anatoxin-1 **1**, a low molecular weight exotoxin produced by toxic blooms of *Anabaena flos-aquae*, has attracted a great deal of attention since its isolation and characterisation in the early 1970s.[1] This interest has been principally due to the profound biological effects of this and associated toxins that have periodically caused extensive loss of wildlife due to contamination of fresh water supplies.[2] The fatal effects of anatoxin-a



1 (+)-Anatoxin-a

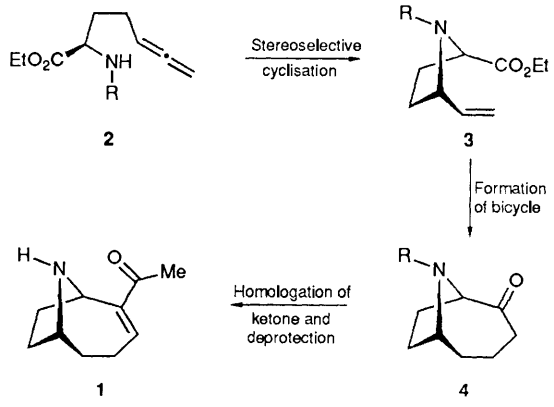
are due in part to the production of a sustained post-synaptic depolarisation at the neuromuscular junction. Further work revealed that anatoxin-a was intimately involved with acetylcholine-mediated neurotransmission pathways and this molecule is currently recognised as the most potent nicotinic agonist known.[3] This observation raises a number of important issues in terms of our understanding of the structure of the nicotinic acetylcholine receptor (nACR) and the steric and electronic requirements imposed by its agonist binding site. A good deal of progress has been made in this area using anatoxin-a and derivatives as pharmacological probes for the nACR but the limited availability of anatoxin-a is a major barrier to more widespread biological investigation. A number of synthetic studies relating to anatoxin-a have been reported[4–6] but the most significant contribution to this area has been made by Rapoport and co-workers.[4c,d,f,5b,c,f] This group have not only described the synthesis of naturally occurring (+)-anatoxin-a, but have also been the source of a large range of derivatives that have formed the basis of most of the recent biological work in the area.[5]

Our interest in anatoxin-a was drawn initially to the synthetic challenges offered by this target. This relatively simple alkaloid contains a number of reactive functional groups within its structure and was viewed as both a useful vehicle for the development of new methodology and as a demanding test of the viability of existing procedures.

In this paper we describe full details of our early synthetic studies towards both racemic and enantiomerically pure anatoxin-a and a number of more general synthetic issues are also discussed.[6]

Results and Discussion

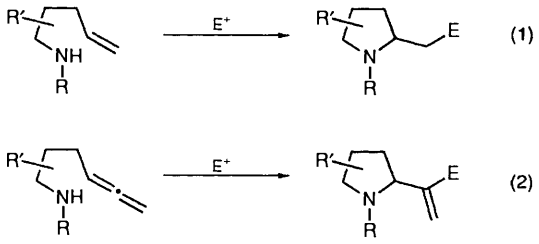
Outline of Synthetic Strategy.—The key synthetic transformations that we proposed to address in our approach to anatoxin-a are shown in Scheme 1.



Scheme 1

This is based on the cyclisation of an allenic amino ester **2** to give the *cis*-2,5-disubstituted pyrrolidine **3** but in order to be useful a high level of stereocontrol must be achieved in this cyclisation step. The use of an allene-based cyclisation incorporates two readily manipulated functional groups—the ester and alkenyl residues—into **3** that should provide the basis of a second cyclisation step to establish the 9-azabicyclo-[4.2.1]nonane skeleton of anatoxin-a. The final phase then required homologation of the ketone **4** to the α,β-unsaturated methyl ketone present in **1**. This latter problem has been addressed successfully by other groups[4a,b,h,l] but we felt that there was an opportunity available to evaluate other methods for achieving this overall transformation.

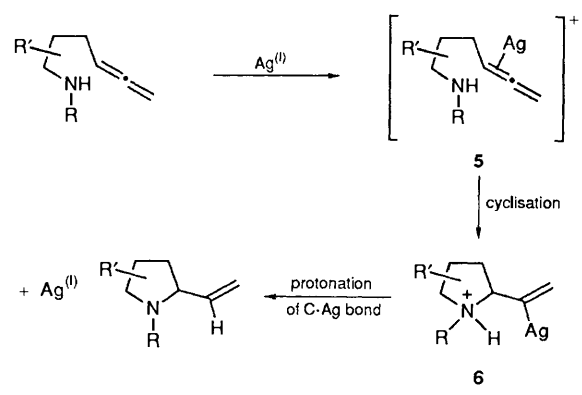
Stereocontrolled Synthesis of Functionalised cis-2,5-*Disubstituted Pyrrolidines. Stereocontrol in the* Ag^{I}-*Mediated Cyclisation of Allenic Amine Derivatives.*—Although the synthesis of nitrogen-containing heterocycles using electrophile-mediated cyclisations of alkenyl-containing substrates (eqn. 1) is well known,[7] the corresponding allenyl-based methods (eqn. 2) are less well established.[8] There are, however, a number of significant advantages attached to the incorporation of an allene moiety into this chemistry, the most obvious feature of which is the level of functionality available in the heterocyclic product shown in eqn. 2. Full use of the synthetic potential of the allene can then be made; one π-bond provides the vehicle for the cyclisation step and the second π-bond offers a versatile means of manipulating the newly formed heterocycle. In addition, the presence of the electrophilic component (E, where



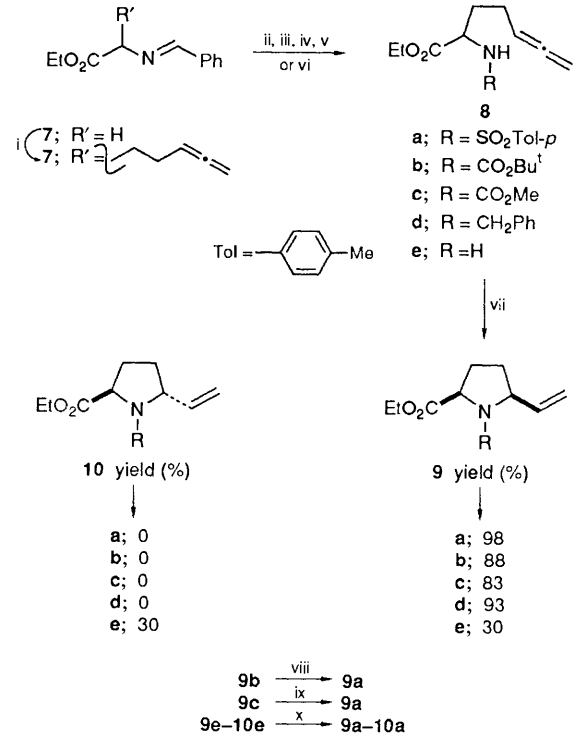
E = *e.g.*, Br, I, RS, metal) on an sp^2-centre also has implications depending on the nature of E. Less well appreciated is the level of reactivity inherent in the allene moiety. Although many electrophilic triggers operate successfully in both eqns. 1 and 2, there are some, *e.g.*, Ag^I, that are only useful in the allene series and the use of this electrophile, which is relevant to this current study, is discussed in more detail below. The increased reactivity of the allene is reflected by the broader range of nitrogen-based nucleophiles[9] that may be successfully used in eqn. 2 and, with appropriate substitution, the allene π-system also allows for the use of chiral, non-racemic substrates. Although this approach to asymmetric synthesis has been examined,[10] the lack of readily available and enantiomerically pure allenes represent a major drawback.

Clearly, in the cyclisations shown in both eqns. 1 and 2, a new stereocentre is created. Stereocontrol at this centre, with respect to other substituents on the newly formed heterocycle, is important and an understanding of the factors that determine the structure of the product is also vital if reliable predictions are going to be made as to the behaviour of new substrates. Within the area of oxygen-containing heterocycles, a great deal is already known about the stereochemical outcome of these two cyclisation processes but the situation relating to nitrogen-containing systems is less clear. Stereochemical studies have previously been carried out on substituted alkenyl amines (*cf.* eqn. 1) by a number of groups,[11] however, one of our objectives in this programme was to shed some light on the behaviour of allenyl derivatives, with an emphasis on the construction of disubstituted pyrrolidines based on the use of *catalytic* Ag^I as the electrophilic trigger. Mechanistic evidence for this unusual and highly efficient reaction, which was discovered by Claesson and co-workers,[12] is lacking but is presumed to involve activation of the allene by complexation to Ag^I to give a π-complex **5** (Scheme 2). Cyclisation *via* this complex should lead to an alkenyl silver intermediate **6** which then undergoes protonation of the C–Ag bond to give the observed product and *regenerate* Ag^I.* This reaction sequence has the advantage of being operationally simple—an aqueous wash removes the silver salt—and removal of the metal from the heterocyclic product takes place *in situ*. A disadvantage to the use of Hg^{II} salts, which are commonly used as electrophilic triggers, is that a second step—reductive cleavage or protonation—is required to decompose the initially formed heterocyclic alkenylmercury complex.†

The synthesis of the requisite allenic esters required for this stereochemical study and for the synthesis of anatoxin-a is based on Stork's method[15] for the alkylation of glycine and a series of derivatives **8a–e** were prepared that differed in terms of the nature of the substituent on nitrogen (Scheme 3). This



Scheme 2



Scheme 3 *Reagents and conditions*: i, Bu^{t}OK, THF, −78 °C, followed by 5-iodopenta-1,2-diene, −78 to 0 °C; ii, **8a**: HCl, H_2O followed by $ClSO_2Tol$-*p*, pyridine, 66%; iii, **8b**: HCl, H_2O followed by BOC_2O, CH_2Cl_2, 49%; iv, **8c**: HCl, H_2O then MeOCOCl, Et_3N, 10%; v, **8d**: $NaBH_4$, ethanol, 60%; vi, **8e**: HCl,H_2O, 59%; vii, $AgBF_4$ (0.1–1 equiv.), CH_2Cl_2, room temperature, yields (see above); viii, CF_3CO_2H, CH_2Cl_2, followed by $ClSO_2Tol$-*p*, pyridine, 50%; ix, Me_3SiCl, NaI, MeCN then $ClSO_2Tol$-*p*, pyridine, 45%; x, $ClSO_2Tol$-*p*, pyridine, 60%

is a strategy that is also amenable to the synthesis of enantiomerically pure material (see below).

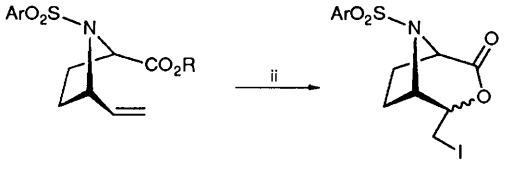
Cyclisation of compounds **8a–e** was carried out using $AgBF_4$ (0.1–1.0 equiv.) in CH_2Cl_2 at room temperature and the products were either isolated directly by crystallisation following an aqueous wash or by chromatography. For compounds **8a–d** the *only* detectable product was the *cis*-2,5-disubstituted pyrrolidine **9a–d** but in the case of primary amine **8e** a 1:1 mixture of *cis*- and *trans*-2,5-disubstituted pyrrolidines **9e** and **10e** was obtained in 60% yield. In this latter instance **9e** and **10e** were not fully characterised but converted directly into the corresponding sulphonamides **9a** and **10a**.

* Complexation of nitrogen to Ag^I is also presumed to be involved but the precise nature of this interaction and its role in the cyclisation process is not yet clear. Although no *direct* evidence has been obtained for the intermediacy of either **5** or **6**, the ionic nature of this mechanism is supported by the use of N–D- rather than N–H-containing substrates. Clean incorporation of deuterium into the alkenyl position formally occupied by Ag in **6** was observed. σ-Alkenyl silver complexes can undergo either heterolysis or homolysis of the C–Ag bond.[13]

† Mercury(II)-mediated cyclisations are useful in a more general synthetic sense since alkenylmercury intermediates undergo *e.g.*, facile halogen–metal and metal–metal exchange.[14]

In all cases, changing the concentrations of Ag^I did not have a significant influence on the product distribution and, with **9a** and **10a** available (obtained indirectly from **7e**), control experiments showed that no *cis* ⇌ *trans* equilibration took place under the reaction conditions. This does not, however, exclude the possibility that equilibration of the putative alkenylsilver intermediate (*cf.* **6**) occurs since protonation to give **9**–**10** is irreversible. Furthermore, attempts to cyclise **8a** using HBF_4 in CH_2Cl_2 failed, thus confirming the role of Ag^I in this transformation. It should also be pointed out that these reactions also proceed smoothly with a range of other Ag^I salts *e.g.*, $AgOCOCF_3$, $AgClO_4$, $AgOSO_2CF_3$ and $AgNO_3$ in a variety of solvents ($ClCH_2CH_2Cl$, $MeCO_2Et$, Me_2CO and EtOH) and using a heterogeneous catalyst such as $AgClO_4$ or $AgNO_3$ on silica gel. In this latter case—with the heterogeneous catalyst–cyclisation of **8a** was, however, nonselective but reaction does appear to take place on the solid support since catalytic activity did not diminish with continued use. With **8d**, cyclisation using this heterogeneous catalyst was also successful. However, only **9d** was observed and, as this basic amine rapidly stripped Ag^I from the support, cyclisation was assumed to have occurred in solution under homogeneous conditions. The catalytic activity of the heterogeneous catalyst was also lost under these circumstances. Sulphonamide **9a** served as a convenient focus for assignment of relative stereochemistry. The *N*-BOC and *N*-methoxycarbonyl derivatives **9b** and **9c** were readily converted into **9a** and, with **9a** and **10a** available, attempts were made to assign relative stereochemistry of these isomers using NOE experiments. All efforts in this area proved inconclusive but chemical proof of structure was readily forthcoming. Saponification of **9a** led to the corresponding carboxylic acid **11** without detectable epimerization. Treatment of **11** with KI–I_2–$NaHCO_3$ led smoothly to a single iodolactone **12**, the stereochemistry of which was not determined. The *trans* series based on **10a** failed to cyclise, under these conditions.

In the case of *N*-benzyl pyrrolidine **9d**, the *cis*-stereochemistry was most conveniently confirmed by cleavage of the alkenyl residue of the corresponding hydrochloride salt with ozone. Oxidative work-up followed by reduction using $LiAlH_4$ gave



Scheme 4 *Reagents and conditions*: i, LiOH, H_2O, 72%; ii, KI, I_2, $NaHCO_3$, H_2O. 24%

cis-*N*-benzyl-2,5-(dihydroxymethyl)pyrrolidine which was compared to an authentic sample.*

At this stage a number of comments need to be made. Firstly, high selectivity for the formation of *cis*-2,5-disubstituted pyrrolidines **9** was observed for derivatives carrying a large group on nitrogen, irrespective of whether this residue was electron withdrawing (SO_2Tol-*p*, CO_2Bu^t, CO_2Me) or electron donating (CH_2Ph). However, no stereoselectivity was apparent with the primary amine **8e**.† Secondly, these results stand in contrast to those obtained by others in alkenyl-based, electrophile-mediated cyclisations where there is generally a preference observed for the formation of *trans*-2,5-disubstituted pyrrolidines.[11] The stereochemical outcome of cyclisations of this type may be determined by a combination of factors including the nature of the electrophile, the solvent and the kinetic–thermodynamic nature of the reaction conditions used. The steric demands of the substituent on nitrogen have not, until now, been fully studied but the Bartlett model,[17] developed to explain the stereochemical outcome of the Lewis acid-mediated cyclisations of δ-alkenyl *O*-alkyl ethers to give *cis*-2,5-disubstituted tetrahydrofurans, may be of value in this area. In this model (Fig. 1), the stereochemical outcome of the

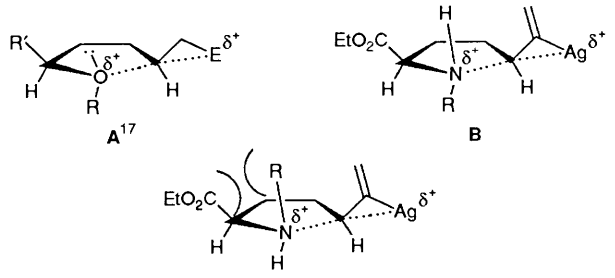


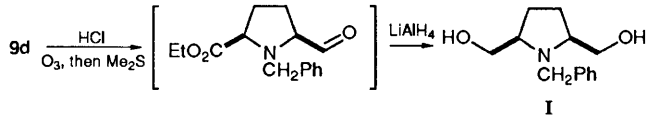
Fig. 1

cyclisation is determined by minimisation of a series of 1,2-interactions between substituents on adjacent atoms in the transition state (**A**). Extending this model to the allene-based cyclisation reactions described above would suggest a preference for **B** over **C**, where R = large group, thus leading to the *cis*-product **9**. But where R = H and the control element is lacking, cyclisation would be expected to be nonselective. This explanation differs from that suggested earlier to explain the predominance of *trans*-2,5-disubstituted pyrrolidines in alkenyl-based cyclisations [11b] but our mechanistic understanding in this area is still far from complete especially in terms of the likely interactions between Ag^I and the basic nitrogen centre. It would also be unwise to assume that parallel mechanisms operate for alkenyl *vs.* allenyl substrates in the presence of different electrophilic triggers. Direct comparisons between **8a** and the corresponding alkenyl amino ester analogue are not yet available but these studies, together with an examination of the effect of other substituents on the allenic–alkyl framework, are underway and will be reported shortly.

Synthesis of the 9-Azabicyclo[4.2.1]*nonane Skeleton of Anatoxin-a.*—With *cis*-2,5-disubstituted pyrrolidines **9a**–**d** in hand, we began to examine methods for manipulating both the alkenyl and ester functions present with the intention of carrying out a second cyclisation to establish the bicyclic skeleton of anatoxin-a, as outlined in Scheme 1. Limitations on space prevent a full description of the reactions examined in this aspect of the study but the most efficient sequence developed was based on sulphonamide **9a** and is shown in Scheme 5.

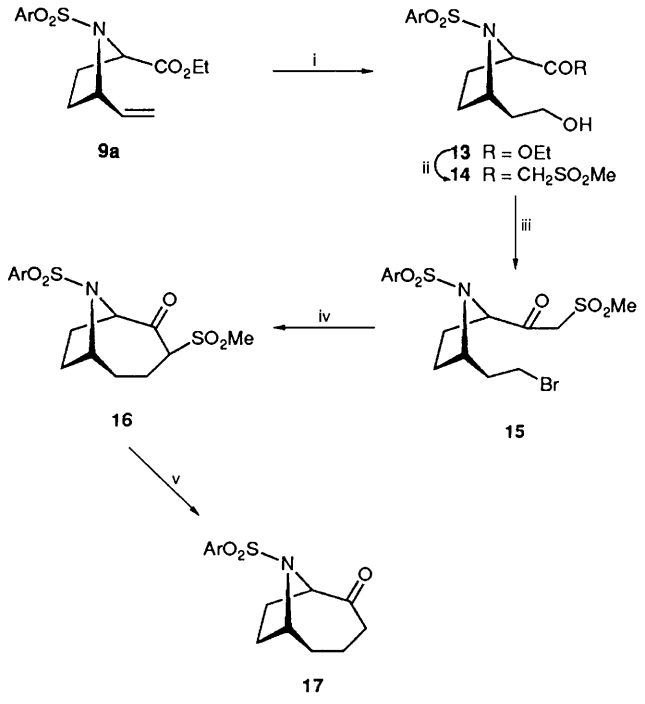
Hydroboration of **9a** (B_2H_6, THF) followed by oxidative work-up (H_2O_2, ^-OH) gave the primary alcohol **13**. Treatment of **13** with $LiCH_2SO_2Me$ (3 equiv., from Me_2SO_2, BuLi, THF) followed by reaction of the resulting β-keto sulphone **14** with

* Pyrrolidine **9d** was converted into *cis*-*N*-benzyl-2,5-dihydroxypyrrolidine I as shown below and compared to an authentic sample. The corresponding *trans* isomers of I was also prepared for purposes of comparison.[16]



I

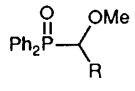
† The *N*-formyl and *N*-acetyl analogues of **i** were also prepared but failed to undergo Ag^I-catalysed cyclisation and the *N*-methyl derivative proved to be unstable and could not be obtained in sufficiently pure form for further use.



Scheme 5 *Reagents and conditions:* i, B_2H_6, THF, room temperature, then NaOH, H_2O_2, 0 °C, 90%; ii, $LiCH_2SO_2Me$ (3 equiv.), THF, −10 °C, 63%; iii, Ph_3P, Br_2. THF, 0 °C to room temperature, 78%; iv. NaH, MeSOMe, 40 °C, 83%; v, Al–Hg, THF, H_2O, 92%.

Ph_3P–Br_2 provided bromide **15**. The overall yield for this three-step sequence was 44% from pyrrolidine **9a**. Bromide **15** is set up to generate the 9-azabicyclo[4.2.1]nonane by an intramolecular alkylation of a sulphone-stabilised enolate.[18] Cyclisation took place smoothly at 40 °C using NaH in DMSO and bicycle **16** was isolated in 83% yield as a single isomer.* Finally, reductive cleavage of the sulphone residue from **16** was straightforward and the *N*-protected bicyclic ketone **17** was obtained in 34% overall yield from allenic ester **8a**.

Homologation of the Bicyclic Ketone **17** *to Anatoxin-a* **1**; *Use of Methoxy Substituted Phosphine Oxide Anions.*—As mentioned above, homologation of ketones related to **17** has been examined in other approaches to anatoxin-a but in this paper we wish to focus on the use and limitations associated



18 R = H
19 R = Me

with anions derived from the methoxy substituted phosphine oxides **18**[19,20] and **19**.[20b,21] The simple phosphine oxide reagent **18** has been used extensively for the homologation of C=O to CH_2CHO and reaction of ketone **17** with the lithio derivative of **18** proceeded smoothly to give the enol ether **20** as a mixture of *E*- and *Z*-isomers (Scheme 6). Problems were envisaged with removal of the sulphonamide residue at a later stage in the sequence so cleavage was affected at this point and the secondary amine was reprotected as its *N*-t-butoxycarbonyl (BOC) derivative **21**. The use of this deprotection–reprotection sequence proved to be more efficient than using **9b** from the outset because of the low yields that were encountered in a synthesis based on this *N*-BOC derivative.

Using the procedures developed by Nicolaou,[22] reaction of **21** with PhSeCl followed by *m*-chloroperbenzoic acid (MCPBA) gave the enal **22** in 61% yield. Conversion of **22** in o *N*-BOC anatoxin-a **23** was carried out in two steps (MeMgBr, then pyridinium chlorochromate) in 37% overall yield and removal of the BOC residue was accomplished in high yield to give (±)-anatoxin-a **1**, the spectral data for which was identical with that previously reported.[4f]

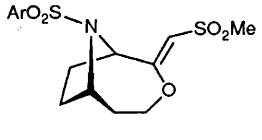
It was felt that this final stage of the synthesis could be significantly improved if both carbon atoms required in the homologation step could be introduced together. For this reason we examined the methylated phosphine oxide **19** which, in contrast to **18**, has not been as widely used as a reagent in synthesis. Deprotonation of **19** [BuLi in 1,2-dimethoxyethane (DME), −78 °C] gave the corresponding anion as a deep-red solution. Following addition of ketone **17** and fragmentation of the resulting diasteroisomeric phosphine oxides, the methyl substituted enol ethers **24** were isolated in 97% yield.

Despite this excellent result, there were problems associated with the use of **19** that were not encountered with the simpler derivative **18**. The preparation of **19** that we used was based on methylation of **18**, which itself is readily available. It is important, however, to purify **19** *rigorously* by careful recrystallisation since the anion derived from **18** is kinetically much more reactive. This is of some significance since we found in model systems that our best yields were obtained with a 3- to 4-fold excess of lithiated phosphine oxide. One final point, the anion derived from **19** appears to be exceptionally sensitive to oxygen and in reactions involving this reagent all solvents were carefully dried and distilled under argon; other workers have also commented on the problems associated with the use of **19**.[21]

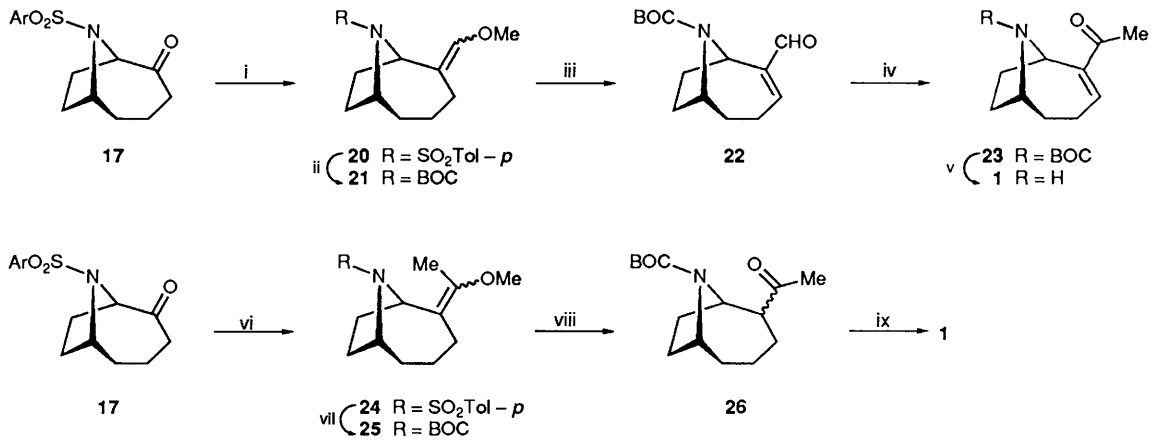
We were unable directly to convert the methylated enol ethers **25** into *N*-BOC anatoxin-a using the Nicolaou procedure and attempts to overcome this problem using an alternative photo-oxygenation[23] sequence also failed. Enol ether cleavage was accomplished using trimethylsilyl iodide (generated *in situ*) to give *N*-BOC dihydroanatoxin-a **26** in 65% yield. This intermediate has previously been converted by Rapoport into anatoxin-a.[4f,5b]

Access to Enantiomerically Pure (*R*)-**8a**.—There are currently a number of methods available for the synthesis of enantiomerically pure amino acids by the asymmetric alkylation of a glycine equivalent.[24] However, to be synthetically useful there is a requirement that the key amino ester is available, not only in high enantiomeric purity but also in multigram quantities and the major problem of using a method based on a chiral auxiliary is the very high cost of the reagents involved. For this reason we have examined an alternative means of obtaining (*R*)-**8a** by enzymatic resolution of (±)-**8a**.[25] Low selectivity was observed using pig liver esterase but chymotrypsin provided the enantiomeric discrimination required for a successful and efficient resolution. Resolution of (±)-**8a** was carried out on a 1 g scale in water–acetone (10:1) using α-chymotrypsin I (25 mg) over 8 h at constant pH 7.2. Following acid–base work-up (*R*)-**8a** was isolated in 48% yield following recrystallisation and (*S*)-**27** was obtained in 45% yield. The optical purity of (*R*)-**8a** was >95%, as judged by chiral shift experiments and the absolute

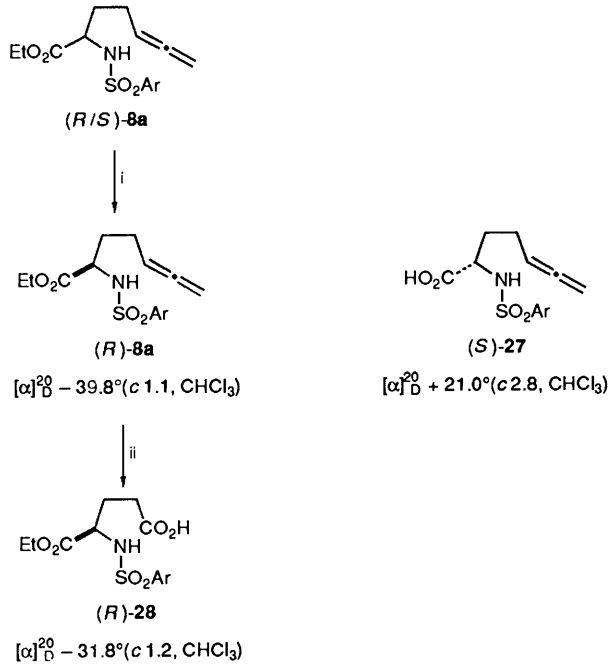
* A small amount (3% isolated yield) of II, a hydrolytically unstable product resulting from *O*-alkylation of the intermediate enolate was also obtained, again as a single isomer. The *Z*-geometry of II was assigned on the basis of a 9% NOE enhancement of the bridgehead proton at C-1 on irradiation of the alkenyl proton.



II



Scheme 6 *Reagents and conditions:* i, **18**, LDA, DME, −78 °C, then NaH, room temperature, 95%; ii, Li, NH_3, then BOC_2O, 85%; iii, PhSeCl, CH_2Cl_2, −30 °C, then MCPBA, −78 °C, 61%; iv, MeMgI, Et_2O, −78 to −40 °C, then pyridinium chlorochromate, NaOAc, CH_2Cl_2, 37%; v, conc. HCl, ethyl acetate, 98%; vi, **19**, LDA, DME, −78 °C, then NaH, room temperature, 97%; vii, Li, NH_3, then BOC_2O, 55%; viii, Me_3SiCl, NaI, MeCN, 65%; ix, ref. 4*f*, 5*b*



Scheme 7 *Reagents and conditions:* i, α-chymotrypsin, acetone, water, pH 7.2, 93% combined yield; ii, O_3, ethyl acetate, −78 °C, then H_2O_2, reflux, 57%

configuration of (*R*)-**8a** was established by ozonolysis to give the glutamic acid derivative **28** which was compared to a sample prepared from (*S*)-glutamic acid.

Summary

In summary, we have investigated the electrophile-cyclisations of substituted allenic amines and examined the stereochemical consequences in terms of the structure of the heterocyclic product, *cis*-2,5-disubstituted pyrrolidines. The size of the *N*-substituent appears to play an important role in determining the stereoselectivity observed but these results do not correlate with those involving alkenyl-based systems and further investigation is required before more concrete conclusions can be drawn. Nevertheless, this allene-based cyclisation chemistry offers considerable flexibility as illustrated by its application to the synthesis of anatoxin-a and these preliminary studies have provided the foundation of a more broadly based investigation of neuroactive cholinergic ligands.

Experimental

IR spectra were recorded using a Perkin-Elmer 1310 grating spectrophotometer and routine mass spectra from electron ionisation (EI, 70 eV), chemical ionisation (CI, isobutane) and high resolution accurate mass determination were recorded with a VG Analytical 7070E instrument with a VG2000 data system. ^1H NMR spectra were recorded at 60 MHz on Hitachi Perkin-Elmer high resolution R-23B and Varian Anaspect EM-360 spectrometers, at 270 MHz on a JEOL GX270 FT spectrometer and at 400 MHz using a Bruker 400 at the SERC facility at Warwick University. ^{13}C NMR spectra were all recorded in $CDCl_3$ on a JEOL GNM GX FT 270 spectrometer operating at 67.8 MHz and using 90 and 135 DEPT pulse sequences to aid in assignment. All *J* values are measured in Hz.

M.p.s are uncorrected and elemental microanalyses were carried out using a Carlo Erba 1106 Elemental Analyser. Optical rotations were measured using a Perkin-Elmer 141 polarimeter. Unless otherwise stated light petroleum refers to that fraction boiling in the range 60–80 °C. Tetrahydrofuran (THF) was dried over sodium benzophenone ketyl under nitrogen and all other reagents and solvents were purified before use using the methods described in D. D. Perrin, W. L. F. Armarego and D. R. Perrin, *Purification of Laboratory Chemicals*, 2nd edn., Pergamon, Oxford, 1980.

Synthesis of Allenic Amino Acid Derivatives **8a–e**.—*Ethyl 2-aminohepta-5,6-dienoate* **8e**. To a stirred suspension of freshly sublimed potassium t-butoxide (5.8 g, 0.052 mol) in THF (120 ml) at −78 °C was added a solution of ethyl *N*-benzylideneglycinate **7** (R′ = H) (9 g, 0.047 mol) in THF (20 ml). The orange solution so formed was stirred for a further 10 min and then treated with a solution of 1-iodopenta-3,4-diene[27] (10 g, 0.052 mol) in THF (25 ml); the reaction mixture was then allowed to warm to ambient temperature over 1.5 h. The solution was stirred for an additional 1.5 h, during which time the colour of the reaction mixture changed to dark green; it was then cooled to 0 °C and added to rapidly stirred ice-cold saturated aqueous ammonium chloride (100 ml). The mixture was diluted with water (40 ml), extracted with diethyl ether (5 × 75 ml), and the organic extracts were combined and concentrated to give ethyl 2-benzylideneaminohepta-5,6-dienoate **7** (R′ = $CH_2CH_2CH{=}C{=}CH_2$) as a pale yellow oil. This material was also used without purification for the preparation of the *N*-benzyl derivative **8d** (see below).

The crude alkylated imine **7** (R′ = $CH_2CH_2CH{=}C{=}CH_2$) was dissolved in diethyl ether (80 ml) and the solution was stirred rapidly with hydrochloric acid (2 mol dm^{-3}; 50 ml) at

ambient temperature for 40 min to effect imine hydrolysis. The organic layer was then separated and extracted with hydrochloric acid (2 mol dm^{-3}; 3 × 20 ml) and the combined aqueous phases were washed with diethyl ether (20 ml). The organic phases were combined, concentrated (to 20 ml), and extracted with hydrochloric acid (2 mol dm^{-3}; 2 × 20 ml). The combined acid fractions were cooled in an ice-bath and carefully treated with concentrated aqueous sodium hydroxide to pH 11. The aqueous phase was then thoroughly extracted with CH_2Cl_2 (5 × 40 ml) and the organic extracts were combined, washed with brine (50 ml), dried (Na_2SO_4) and concentrated to give the ester **8e** (5.2 g, 59%) as a yellow oil; ν_{max}(thin film)/cm^{-1} 3500, 1955 and 1725; δ_H(60 MHz; $CDCl_3$) 5.10 (1 H, m), 4.85–4.52 (2 H, m), 4.15 (2 H, q, *J* 7), 3.52 (1 H, m), 2.38 (2 H, s), 2.35–1.54 (4 H, m) and 1.25 (3 H, t, *J* 7).

The amine **8e** was used without further purification for the preparation of **8a**, **8b** and **8c** and for cyclisation studies.

Ethyl 2-(p-*tolylsulphonylamino*)*hepta*-5,6-*dienoate* **8a**. The amine **8e** was treated under standard conditions with toluene-*p*-sulphonyl chloride in pyridine at 0 °C, and, following an aqueous work-up and flash chromatography, gave the ethyl ester **8a** as colourless needles in 66% overall yield from the imine **7** (R′ = H), m.p. 45–46 °C (diethyl ether–light petroleum) (Found: C, 59.4; H, 6.6; N, 4.4. $C_{16}H_{21}NO_4S$ requires: C, 59.42; H, 6.54; N, 4.33%); ν_{max}(thin film)/cm^{-1} 3450, 1955 and 1725; δ_H(270 MHz; $CDCl_3$) 7.73 (2 H, d, part of AA′BB′, *J* 8), 7.29 (2 H, d, part of AA′BB′, *J* 8), 5.26 (1 H, br d, *J* 9.2), 5.07 (1 H, quint., *J* 6.6), 4.68 (2 H, dt, *J* 6.5, 3), 3.93 (2 H, q, *J* 7), 3.90 (1 H, m), 2.41 (3 H, s), 2.03–2.15 (2 H, m), 1.65–1.93 (2 H, m) and 1.10 (3 H, t, *J* 7); δ_C 208.4 (C=*C*=C), 171.6 (CO), 143.6 (C), 136.6 (C), 129.6 (CH), 127.2 (CH), 88.4 (CH), 75.7 (CH_2), 61.6 (CH_2), 55.2 (CH), 32.6 (CH_2), 23.5 (CH_2), 21.5 (CH_3) and 13.9 (CH_3); *m*/*z* (CO) 324 $(M + 1)^+$.

Ethyl 2-(*t-butoxycarbonylamino*)*hepta*-5,6-*dienoate* **8b**. A solution of the crude amine **8e** was treated with di-t-butyl dicarbonate in CH_2Cl_2 under standard conditions. Following an aqueous work-up and purification by flash chromatography [light petroleum–ethyl acetate (9:1)] **8b** was isolated as a colourless crystalline solid [49% yield from **7** (R′ = H)], m.p. 52–54 °C (diethyl ether–light petroleum) (Found: C, 62.6; H, 8.8; N, 5.2. $C_{14}H_{23}NO_4$ requires C, 62.43; H, 8.61; N, 5.20%); ν_{max}($CHCl_3$)/cm^{-1} 3500, 1960, 1730 and 1705; δ_H(270 MHz; $CDCl_3$) 5.12 (1 H, quint., *J* 6.5), 5.06 (1 H, br m), 4.70 (2 H, dt, *J* 6.5, 3), 4.32 (1 H, br m), 4.20 (2 H, q, *J* 7), 1.86–2.14 (3 H, m), 1.73 (1 H, m), 1.45 (9 H, s) and 1.28 (3 H, t, *J* 7); *m*/*z* (CI) 270 $(M + 1)^+$.

Ethyl 2-(*methoxycarbonylamino*)*hepta*-5,6-*dienoate* **8c**. A solution of the crude amine **8e** was treated with methyl chloroformate and triethylamine in CH_2Cl_2 under standard conditions. Following an aqueous work-up and purification by flash chromatography [light petroleum–ethyl acetate (9:1)] **8c** was isolated as a colourless oil [10% yield from **7** (R′ = H)] (Found: C, 58.15; H, 7.55; N, 6.25. $C_{11}H_{17}NO_4$ requires C, 58.13; H, 7.54; N, 6.16%); ν_{max}($CHCl_3$)/cm^{-1} 3420, 1955 and 1700; δ_H(270 MHz; $CDCl_3$) 5.30 (1 H, br d, NH), 5.12 (1 H, m), 4.74–4.68 (2 H, m), 4.38 (1 H, m), 4.20 (2 H, q, *J* 7), 3.74 (3 H, s), 2.18–1.75 (4 H, m) and 1.26 (3 H, t, *J* 7); *m*/*z* (EI) 227 (M^+), 226, 196 and 154.

The yield of this procedure was not optimised but the major by-product was the symmetrical urea which was isolated as a mixture of diastereoisomers.

Ethyl 2-(*benzylamino*)*hepta*-5,6-*dienoate* **8d**. To a solution of the crude imine **7** (R′ = CH_2CH_2CH=C=CH_2) [from ethyl *N*-(phenylmethylene)glycinate (4.0 g, 21 mmol) and isolated as described above] in ethanol (25 ml) at 0 °C was added sodium borohydride (0.5 g). The mixture was stirred at room temperature for 15 h and then hydrochloric acid (2 mol dm^{-3}; 75 ml) and water (75 ml) were added. The solution was extracted with ether (2 × 25 ml) and the aqueous phase was made basic using solid $NaHCO_3$ and then extracted with ether (5 × 25 ml). The extracts were concentrated and the residue purified by passage through a short column of silica using CH_2Cl_2 as eluant to give **8d** as a pale yellow oil [3.2 g, 60% from **7** (R = H)]; ν_{max}(thin film)/cm^{-1} 3350, 1955 and 1730; δ_H(60 MHz), 7.20 (5 H, br s), 5.01 (1 H, m), 4.75–4.55 (2 H, m), 4.13 (2 H, q, *J* 7), 3.85 (1 H, d, *J* 13), 3.56 (1 H, d, *J* 13), 3.26 (1 H, t, *J* 7), 2.40–1.50 (5 H, m, including NH) and 1.25 (3 H, t, *J* 7); *m*/*z* (CI) 260 $(M^+ + 1)$.

Although homogeneous by ^1H NMR and TLC, satisfactory elemental analysis could not be obtained for this compound.

Silver(I)*-catalysed Cyclisation of Allenic Amino Esters* **8a–d**.—cis-(p-*Tolylsulphonyl*)-5-*vinylproline ethyl ester* **9a**. A solution of the ester **8a** (5 g, 15.5 mmol) in CH_2Cl_2 (50 ml) was treated with $AgBF_4$ (300 mg, 1.54 mmol) and stirred in the dark at ambient temperature for 7 d. The reaction mixture was then washed with water (25 ml) and the aqueous washings were extracted with CH_2Cl_2 (3 × 25 ml). The organic phases were combined, washed with brine (30 ml), dried (Na_2SO_4), concentrated and purified by flash chromatography [light petroleum–ethyl acetate (5:1)] to give **9a** as a colourless crystalline solid (4.90 g, 98%), m.p. 58–59 °C (diethyl ether–light petroleum) (Found: C, 59.1; H, 6.4; N, 4.2. $C_{16}H_{21}NO_4S$ requires: C, 59.42; H, 6.54; N, 4.33%); ν_{max}($CHCl_3$)/cm^{-1} 1740 and 1600; δ_H(270 MHz; $CDCl_3$) 7.76 (2 H, d, part of AA′BB′, *J* 8), 7.30 (2 H, d, part of AA′BB′, *J* 8), 5.79 (1 H, ddd, *J* 17, 10, 6.5), 5.37 (1 H, dt, *J* 17, 1), 5.09 (1 H, dt, *J* 10, 1), 4.37 (1 H, t, *J* 7), 4.25 (1 H, m), 4.19 (2 H, qd, *J* 7, 2.5), 2.42 (3 H, s), 1.93–2.05 (2 H, m), 1.69–1.91 (2 H, m) and 1.27 (3 H, t, *J* 7); δ_C 199.0 (CO), 143.5 (C), 138.0 (CH), 135.7 (C), 129.4 (CH), 127.7 (CH), 116.4 (CH_2), 63.2 (CH), 61.8 (CH), 61.3 (CH_2), 31.9 (CH_2), 29.3 (CH_2), 21.5 (CH_3) and 14.1 (CH_3); *m*/*z* (CI) 324 $(M + I)^+$.

cis-1-(*t-Butoxycarbonyl*)-5-*vinylproline ethyl ester* **9b**. Using the same procedure as described above for **8a**, cyclisation of **8b** was carried out using $AgBF_4$ (0.25 equiv.) in CH_2Cl_2 for 72 h to give **9b** as a colourless oil in 88% yield [Found: M, 196.1299 (M − CO_2Et). $C_{11}H_{18}NO_2$ requires M, 196.1337]; ν_{max}(thin film)/cm^{-1} 1730 and 1690; δ_H(270 MHz; $CDCl_3$) 5.83 (1 H, m), 5.34 (1 H, m), 5.09 (1 H, m), 4.10–4.62 (2 H, m), 4.19 (2 H, qd, *J* 7, 3), 1.62–2.32 (4 H, m), 1.42 (9 H, br s) and 1.29 (3 H, t, *J* 7); *m*/*z* (CI) 270 $(M + 1)^+$.

The ester **9b** was correlated with **9a** by removal of the BOC residue (CF_3CO_2H, CH_2Cl_2, room temperature) followed by reaction of the resulting amine **9e** with toluene-*p*-sulphonyl chloride in pyridine at 0 °C.

cis-1-*Methoxycarbonyl*-5-*vinylproline ethyl ester* **9c**. Using the same procedure as described above for **8a**, cyclisation of **8c** was carried out using 1 equiv. of $AgBF_4$ in CH_2Cl_2 for 65 h to give **9c** as a colourless oil in 83% yield (Found: M^+, 227.117. $C_{11}H_{17}NO_4$ requires M, 227.115); ν_{max}(thin film)/cm^{-1} 1745 and 1650; δ_H(270 MHz; $CDCl_3$) 5.95 (1 H, m), 5.55 (1 H, br d, *J* 15), 5.20 (1 H, br d, *J* 10), 4.50–4.35 (2 H, m), 4.28 (2 H, q, *J* 7), 3.72 (3 H, s), 2.40–1.85 (4 H, m) and 1.31 (3 H, t, *J* 7); *m*/*z* (EI) 227 (M^+), 168 and 154.

The pyrrolidine **9c** appears to be a single isomer by ^1H NMR.

Conversion of **9c** *into* **9a**. A solution of **9c** (84 mg, 0.37 mmol) in dry acetonitrile (10 ml) was treated with NaI (290 mg, 1.93 mmol) and Me_3SiCl (214 mg, 1.97 mmol) and the resulting mixture was heated at reflux for 2 h. The solution was then cooled to room temperature diluted with methanol and stirred for 30 min. After this time, the solvents were removed and the residue was dissolved in pyridine (4 ml) and the solution cooled in an ice-bath and treated with toluene-*p*-sulphonyl chloride (78 mg, 0.14 mmol). Following standard work-up, **9a** was isolated in 54% yield and none of the corresponding *trans* isomer **10a** was detected by ^1H NMR.

cis-1-*Benzyl*-6-*vinylproline ethyl ester* **9d**. Using the same procedure as described above for **8a**, cyclisation of **8d** was carried out using $AgBF_4$ (1 equiv.) in CH_2Cl_2 for 4 h at room temperature to give, after purification by flash chromatography, **9d** in 93% yield as a colourless oil; ν_{max}(thin film)/cm^{-1} 1745 and 1640; δ_H(400 MHz; $CDCl_3$) 7.45–7.30 (5 H, m), 6.00 (1 H, m), 5.38 (1 H, br d, *J* 15), 5.28 (1 H, br d, *J* 10), 4.14 (1 H, d, *J* 14), 4.12–4.05 (2 H, m), 4.10 (2 H, q, *J* 7), 3.73 (1 H, d, *J* 14), 3.41 (1 H, m), 3.27 (1 H, m), 2.18–1.90 (2 H, m) and 1.25 (3 H, t, *J* 7).

Although homogeneous by ^1H NMR and TLC, satisfactory elemental or high resolution mass spectral data could not be obtained for this product.

The same product was isolated in 92% yield when cyclisation was carried out using $AgNO_3$ in EtOAc.

Cyclisation of **8e**; cis *and* trans-5-*Vinylproline Ethyl Ester* **9e** *and* **10e** *and Conversion into* **9a** *and* **10a**.—Using the same procedure as described above for **8a**, cyclisation of **8e** was carried out using $AgBF_4$ (1 equiv.) in CH_2Cl_2 for 1 h at room temperature. The reaction mixture was washed with water and then dried (Na_2SO_4) and the crude product was immediately dissolved in pyridine and treated with toluene-*p*-sulphonyl chloride (1.2 equiv.) at 0 °C. After aqueous work up, purification by flash chromatography gave a 1:1 mixture of **9a** and **10a**, as judged by ^1H NMR, in 60% yield. A small quantity of **10a** was isolated following a careful separation by radial chromatography ('Chromatatron'); m.p. 104–105 °C (diethyl ether–light petroleum); ν_{max}(thin film)/cm^{-1} 1745 and 1640; δ_H(400 MHz; $CDCl_3$) 7.73 (2 H, d, part of AA'BB', *J* 8), 7.25 (2 H, d, part of AA'BB', *J* 8), 5.61 (1 H, m), 5.14 (1 H, br d, *J* 18), 5.01 (1 H, br d, *J* 10), 4.46–4.43 (2 H, m), 4.16 (2 H, m), 2.41 (3 H, s), 2.40 (1 H, m), 2.24 (1 H, m), 1.95 (1 H, ddt, *J* 13, 7, 1), 1.70 (1 H, ddt, *J* 12, 7, 2) and 1.28 (3 H, t, *J* 7).

cis-1-(p-*Tolylsulphonyl*)-5-*vinylproline* **11**.—A solution of the ester **9a** (568 mg, 1.76 mmol) in THF (8 ml) and water (5 ml) was treated with lithium hydroxide monohydrate (277 mg, 6.6 mmol). The reaction mixture was stirred at ambient temperature for 16 h and then diluted with water (20 ml) and washed with CH_2Cl_2 (20 ml). The aqueous solution was acidified to pH 2 with hydrochloric acid (2 mol dm^{-3}) and extracted with CH_2Cl_2 (4 × 25 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated to give **11** (375 mg, 72%) as a colourless solid, m.p. 150–151 °C (benzene–light petroleum) (Found: C, 57.3; H, 5.9; N, 4.8. $C_{14}H_{17}NO_4S$ requires C, 56.93; H, 5.80; N, 4.74%); ν_{max}($CHCl_3$)/cm^{-1}: 1725; δ_H(270 MHz; $CDCl_3$) 9.70 (1 H, br), 7.77 (2 H, d, part of AA'BB', *J* 8), 7.24 (2 H, d, part of AA'BB', *J* 8), 5.81 (1 H, ddd, *J* 17, 10, 6), 5.35 (1 H, dt, *J* 17, 1), 5.13 (1 H, dt, *J* 10, 1), 4.34 (1 H, dd, *J* 8.5), 4.19 (1 H, q, *J* 6), 2.44 (3 H, s), 1.89–2.21 (2 H, m) and 1.80 (2 H, qd, *J* 7, 1); δ_C 176.7 (C), 144.0 (C), 137.8 (CH), 135.0 (C), 129.7 (CH), 127.9 (CH), 116.6 (CH_2), 63.5 (CH), 61.8 (CH), 31.9 (CH_2), 29.0 (CH_2) and 21.6 (CH_3); *m/z* (CI) 296 (M + 1)$^+$.

The same procedure was applied to **10a** to give *trans*-(*p*-tolylsulphonyl)-5-vinylproline in 65% yield, m.p. 154–155 °C (from benzene); ν_{max}($CHCl_3$)/cm^{-1} 1725; δ_H(270 MHz; $CDCl_3$) 10.25 (1 H, br), 7.72 (2 H, d, part of AA'BB', *J* 8), 7.25 (2 H, d, part of AA'BB', *J* 8), 5.55 (1 H, m), 5.20–4.90 (2 H, m), 4.60–4.34 (2 H, m), 2.41 (3 H, s) and 2.38–1.54 (4 H, m); δ_C 178.0 (C), 143.5 (C), 137.7 (C), 137.3 (CH), 129.3 (CH), 127.8 (CH), 116.8 (CH_2), 62.7 (CH), 60.9 (CH), 31.5 (CH_2), 29.0 (CH_2) and 21.5 (CH_3); *m/z* (CI) 296 (M + 1).

4-(*Iodomethyl*)-(p-*tolylsulphonyl*)-3-*oxa*-8-*aza*-bicyclo-(3.2.1]*octan*-2-*one* **12**.—To a solution of acid **11** (29.5 mg, 0.1 mmol) in water (3 ml) containing sodium hydrogen carbonate (25 mg, 0.3 mmol) was added potassium iodide (100 mg, 0.6 mmol) and iodine (51 mg, 0.2 mmol) and the mixture was stirred at room temperature for 15 h. The solution was then extracted with CH_2Cl_2 (3 × 3 ml) and the extracts were washed with dilute aqueous sodium thiosulphate, dried ($NaSO_4$), and evaporated to give a waxy solid which was recrystallised from benzene to give **12** (10 mg, 24%) as colourless crystals, m.p. 185 °C (Found: C, 40.0: H, 3.9: N, 3.05. $C_{14}H_{16}INO_4S$ requires C, 39.92; H, 3.83; N, 3.33%); ν_{max}($CDCl_3$)/cm^{-1} 1770; δ_H(400 MHz; $CDCl_3$) 7.78 (2 H, d, part of AA'BB', *J* 8), 7.34 (2 H, d, part of AA'BB', *J* 8), 4.69 (2 H, t, *J* 4), 4.31 (1 H, dd, *J* 10, 4), 3.27 (1 H, dd, *J* 10, 4), 3.16 (1 H, t, *J* 10), 2.45 (3 H, s) and 2.17–1.80 (4 H, m); δ_C 166.0 (C), 145.0 (C), 135.5 (C), 130.0 (CH), 127.5 (CH), 84.9 (CH), 60.4 (CH), 56.3 (CH), 29.6 (CH_2), 27.2 (CH_2), 21.6 (CH_3) and 2.60 (CH_2); *m/z* (EI) 421 (M^+), 294, 250 and 155.

Acidification of the aqueous phase yielded 16 mg of recovered **11**, the structure of which was confirmed by ^1H NMR.

Synthetic Studies Directed Towards Anatoxin-a.—cis-5-(2-*Hydroxyethyl*)-1-(p-*tolylsulphonyl*)*proline ethyl ester* **13**. To a stirred solution of boron trifluoride–diethyl ether (250 mg, 300 μl, 2.4 mmol) in bis(2-methoxyethyl) ether (2 ml) was added a solution of sodium borohydride (1 mol dm^{-3}; 1.0 ml, 1.0 mmol) in bis(2-methoxyethyl) ether and the diborane so generated was bubbled, in a stream of nitrogen, through a solution of the alkene **9a** (200 mg, 0.62 mmol) in THF (5 ml) at 20 °C. The reaction mixture was stirred at ambient temperature for 20 min, after which it was cooled to 0 °C and carefully treated with sufficient water to destroy the excess of diborane. To the resulting solution was first added aqueous sodium hydroxide (3 mol dm^{-3}; 0.3 ml, 0.9 mmol), followed by 30% hydrogen peroxide (0.3 ml, 0.9 mmol), care being taken that the temperature of the reaction did not rise above 25 °C. The reaction mixture was then stirred at ambient temperature for 30 min after which water (10 ml) and ethyl acetate (10 ml) was added. This mixture was extracted with ethyl acetate (5 × 10 ml) and the organic extracts were combined, dried (Na_2SO_4) and concentrated and the residue was purified by flash chromatography [light petroleum–ethyl acetate (3:2)] to give the alcohol **13** (190 mg, 90%) as a colourless glass; ν_{max}($CHCl_3$)/cm^{-1} 3520 and 1735; δ_H(270 MHz; $CDCl_3$) 7.75 (2 H, d, part of AA'BB', *J* 8), 7.35 (2 H, d, part of AA'BB', *J* 8), 4.21 (2 H, q, *J* 7), 4.11 (1 H, t, *J* 8), 4.09 (1 H, m), 3.78 (1 H, dt, *J* 12, 4), 3.00 (1 H, br, OH), 2.44 (3 H, s), 1.89–2.12 (2 H, m), 1.73–1.80 (2 H, m), 1.61 (1 H, m), 1.41 (1 H, m), 1.29 (3 H, t, *J* 7) and 1.05–1.23 (1 H, m); δ_C 172.2 (CO), 144.1 (C), 134.1 (C), 129.2 (CH), 127.5 (CH), 61.7 (CH), 61.5 (CH_2), 58.8 (CH_2), 58.8 (CH), 37.6 (CH_2), 30.7 (CH_2), 29.4 (CH_2), 21.5 (CH_3) and 14.0 (CH_3); *m/z* (CI) 342 (M + 1)$^+$.

This material was homogeneous by TLC and ^1H NMR, but could not be obtained in sufficiently pure form to obtain a satisfactory elemental analysis.

cis-2-(2-*Hydroxyethyl*)-1-(p-*tolylsulphonyl*)-5-[(*methyl-sulphonyl*)*acetyl*]*pyrrolidine* **14**. A solution of dimethyl sulphone (1.36 g, 14.5 mmol) in THF (100 ml) at 0 °C was treated with a solution of butyllithium in hexane (1.6 mol dm^{-3}; 8.1 ml, 12.9 mmol) to give a colourless precipitate. This suspension was stirred at ambient temperature for 20 min and then cooled to −10 °C and treated with a solution of **13** (1.1 g, 3.2 mmol) in THF (25 ml). The reaction mixture rapidly cleared and after 10 min was quenched by addition of saturated aqueous ammonium chloride (40 ml). The mixture was acidified to pH 4 with hydrochloric acid (2 mol dm^{-3}), diluted with water (30 ml) and extracted with ethyl acetate (5 × 50 ml). The organic extracts were combined and washed with water (50 ml) and brine (70 ml), dried (Na_2SO_4) and then concentrated to give a colourless oil. This oil was purified by flash

chromatography [ethyl acetate–light petroleum (4:1)] to give **14** (0.79 g, 63%); ν_{max}(thin film)/cm^{-1} 3570 and 1740; δ_H(400 MHz; $CDCl_3$) 7.70 (2 H, d, part of AA′**BB**′, *J* 8), 7.36 (2 H, d, part of AA′**BB**′, *J* 8), 4.60 (1 H, dq, *J* 15, 1), 4.35 (1 H, dt, *J* 15, 0.5), 3.94–4.70 (4 H, m), 3.77 (1 H, br m), 3.14 (3 H, s), 2.45 (3 H, s), 2.15 (1 H, m), 1.83–1.94 (2 H, m), 1.77 (1 H, m), 1.64 (1 H, m) and 1.41 (1 H, m); δ_C 198.5 (CO), 144.8 (C), 132.6 (C), 130.1 (CH), 127.8 (CH), 69.1 (CH), 60.5 (CH_2), 59.6 (CH), 59.0 (CH_2), 42.1 (CH_2), 38.5 (CH_2), 30.5 (CH_2), 26.9 (CH_2) and 21.5 (CH_3); *m*/*z* (CI) 390 $(M + 1)^+$.

This material was homogeneous by TLC and ^{1}H NMR, but could not be obtained in sufficiently pure form to obtain a satisfactory elemental analysis.

cis 2-(2-*Bromoethyl*)-5-[(*methylsulphonyl*)*acetyl*]-1-(p-*tolylsulphonyl*)*pyrrolidine* **15**. A 2M solution of bromine in THF was added dropwise to a stirred solution of triphenylphosphine (440 mg, 1.68 mmol) in THF (15 ml) at 0 °C. A colourless precipitate immediately began to form. When just enough bromine solution had been added to give the reaction mixture a permanent yellow colour a further portion of triphenylphosphine (20 mg) was added so that the colour was removed. The reaction mixture was then stirred at ambient temperature for 10 min before being cooled to 0 °C and treated with a solution of **14** (365 mg, 0.94 mmol) in THF (5 ml). After 10 min, the reaction mixture was allowed to warm to ambient temperature when it was diluted with water (20 ml) and extracted with ethyl acetate (5 × 20 ml). The organic extracts were combined, washed with brine (20 ml), dried (Na_2SO_4) and concentrated and the residue purified by flash chromatography [ethyl acetate–light petroleum (3:2)] to give **15** as colourless needles (331 mg, 78%), m.p. 123 °C (ethyl acetate–light petroleum) (Found: C, 42.4; H, 4.8; N, 3.0. $C_{16}H_{22}BrNO_5S_2$ requires C, 42.48; H, 4.90; N, 3.09%); ν_{max}($CHCl_3$)/cm^{-1} 1720; δ_H(400 MHz; $CDCl_3$) 7.73 (2 H, d, part of AA′BB′, *J* 8), 7.38 (2 H, d, part of AA′BB′, *J* 8), 4.59 (1 H, dq, *J* 15, 1.5), 4.36 (1 H, d, *J* 15), 4.09 (1 H, t, *J* 8), 3.85 (1 H, tdd, *J* 8, 5, 3), 3.62 (1 H, dt, *J* 10, 6), 3.40 (1 H, ddd, *J* 10, 8, 5), 3.14 (3 H, s), 2.51 (1 H, ddt, *J* 14, 8, 5), 2.45 (3 H, s), 2.14 (1 H, dddd, *J* 18, 9, 8, 6), 2.02 (1 H, ddt, *J* 14, 8, 5), 1.80 (1 H, dtd, *J* 13, 7, 4) and 1.50–1.67 (2 H, m); *m*/*z* (CI) 454 and 452 $(M + 1)^+$.

3-(*Methylsulphonyl*)-9-(p-*tolylsulphonyl*)-9-*azabicyclo*-[4.2.1]*nonan*-2-*one* **16**. To a stirred solution of the bromide **15** (725 mg, 1.6 mmol) in dry dimethyl sulphoxide (30 ml) was added sodium hydride (154 mg, 6.4 mmol) and the reaction mixture stirred at 40 °C for 2 h. The solution was then poured into a rapidly stirred mixture of hydrochloric acid (2 mol dm^{-3}; 40 ml) and ice (20 g). After 10 min, the suspension was extracted with CH_2Cl_2 (5 × 25 ml) and the extracts were dried (Na_2SO_4) and concentrated and the residue purified by flash chromatography [ethyl acetate–light petroleum (4:1)] to give **16** (496 mg, 83%); m.p. 215 °C (methanol) (Found: C, 51.4; H, 5.6; N, 3.6. $C_{16}H_{21}NO_5S_2$ requires C, 51.73; H, 5.70; N, 3.77%); ν_{max}/cm^{-1} 1735; δ_H(400 MHz; $CDCl_3$) 7.70 (2 H, d, part of AA′BB′, *J* 8), 7.33 (2 H, d, part of AA′BB′, *J* 8), 4.56 (1 H, dd, *J* 12, 2), 4.53 (1 H, m), 4.45 (1 H, dd, *J* 10, 2), 3.05 (3 H, s), 2.66 (1 H, dtd, *J* 14, 4, 1.5), 2.44 (3 H, s), 2.28 (1 H, tt, *J* 7, 4), 1.65–2.00 (5 H, m) and 1.55 (1 H, m); *m*/*z* (CI) 372 $(M + 1)^+$.

Continued elution gave (*Z*)-2-methylsulphonylmethylene-3-oxa-9-aza-bicyclo[4.2.1]nonane as a colourless solid (15 mg, 3%), m.p. 240–242 °C (decomp.) (methanol) (Found: M^+, 371.087. $C_{16}H_{21}NO_5S_2$ requires M, 371.086); ν_{max}($CHCl_3$)/cm^{-1} 1620; δ_H(400 MHz; $CDCl_3$) 7.72 (2 H, d, part of AA′BB′, *J* 8), 7.31 (2 H, d, part of AA′BB′, *J* 8), 5.60 (1 H, s), 4.63 (1 H, dd, *J* 11, 3), 4.45–4.51 (1 H, m), 4.37 (1 H, dt, *J* 13, 4), 4.12 (1 H, td, *J* 13, 2), 3.00 (3 H, s), 2.43 (3 H, s), 2.19–2.30 (2 H, m), 1.97 (1 H, m), 1.77 (1 H, m) and 1.61–1.69 (2 H, m); *m*/*z* (EI) 371 $(M)^+$.

9-(p-*Tolylsulphonyl*)-9-*azabicyclo*[4.2.1]*nonan*-2-*one* **17**. Aluminium foil (4 cm^3) was submerged in an aqueous solution of mercuric chloride (0.074 mol dm^{-3}) for 15 s and then washed with absolute ethanol followed by diethyl ether. The freshly treated foil was added to an unstirred solution of the keto sulphone **16** (230 mg, 0.62 mmol) in THF (6 ml) and water (0.5 ml) at 60 °C. Further portions of freshly treated aluminium foil (4 cm^2) were added to the reaction every 30 min. After 3 h, the reaction mixture was filtered through a pad of Celite and the solid residue washed with ethyl acetate (5 × 5 ml). The organic filtrates were combined, dried (Na_2SO_4) and concentrated and the residue purified by flash chromatography [ethyl acetate–light petroleum (1:1)] to give **17** as a colourless solid (168 mg, 92%), m.p. 155 °C (methanol) (Found: C, 61.3; H, 6.6; N, 4.4. $C_{15}H_{19}NO_3S$ requires C, 61.41; H, 6.53; N, 4.77%); ν_{max}(Nujol)/cm^{-1} 1715; δ_H(400 MHz; $CDCl_3$) 7.71 (2 H, d, part of AA′BB′, *J* 8), 7.30 (2 H, d, part of AA′BB′, *J* 8), 4.51 (1 H, dq, *J* 12, 3), 4.32 (1 H, dd, *J* 10, 3), 3.05 (1 H, td, *J* 14, 2), 2.43 (3 H, s), 2.32 (1 H, dd, *J* 14, 6), 2.21 (1 H, tt, *J* 14, 4) and 1.90–1.50 (7 H, m); *m*/*z* (CI) 294 $(M + 1)^+$.

1-*Methoxyethyl*(*diphenyl*)*phosphine oxide* **19**. To a stirred solution of LDA [from diisopropylamine (1.62 g, 16 mmol) and butyllithium in hexane (1.6 mol dm^{-3}; 10 ml, 16 mmol)] in THF (300 ml) at at 0 °C was added a solution of phosphine oxide **18** (3.7 g, 15 mmol) in THF (50 ml) and a red colour formed. After 10 min at 0 °C, the solution was cooled to −78 °C and treated with methyl iodide (2.56 g, 18 mmol). The colour was lost immediately and after 5 min the reaction was quenched by addition of saturated aqueous ammonium chloride (100 ml). After warming to ambient temperature, the mixture was treated with water (100 ml) and extracted with CH_2Cl_2 (5 × 50 ml). The organic extracts were combined, dried (Na_2SO_4), concentrated and the residue purified twice by careful recrystallisation to give **19** as a colourless crystalline solid (2.6 g, 66%), m.p. 77–79 °C (toluene–light petroleum) (lit.,[21a] m.p. 77–78 °C); δ_H(60 MHz; $CDCl_3$) 7.4–8.3 (10 H, m), 4.1 (1 H, m), 3.3 (3 H, s) and 1.5 (3 H, dd, *J* 15, 7).

(E)- *and* (Z)-2-*Methoxymethylene*-9-(p-*tolylsulphonyl*)-9-*azabicyclo*[4.2.1]*nonane* **20**. To a stirred solution of diisopropylamine (29 mg, 0.29 mmol, 40 μl) in DME (2 ml) at −50 °C was added a solution of butyllithium in hexane (1.6 mol dm^{-3}; 24 mmol, 150 μl). The mixture was stirred for 10 min and then cooled to −78 °C and treated with a solution of phosphine oxide **18** (94 mg, 0.38 mmol) in DME (2 ml). Immediately an orange colour formed and after 20 min the reaction mixture was treated with a solution of the ketone **17** (28 mg, 0.10 mmol) in DME (2 ml). The reaction mixture was stirred for 5 min and then quenched with saturated aqueous ammonium chloride (4 ml). The mixture was warmed to ambient temperature, diluted with water (2 ml), and extracted with ethyl acetate (5 × 5 ml). The organic extracts were combined, dried (Na_2SO_4), and concentrated to give the diastereoisomeric hydroxyphosphine oxides as a colourless solid. This solid was dissolved in THF (5 ml), treated with sodium hydride (24 mg, 1.0 mmol) and stirred at ambient temperature for 14 h. The reaction mixture was then cooled to 0 °C, quenched with water (3 ml) and extracted with ethyl acetate (5 × 5 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated and the residue was purified by flash chromatography [ethyl acetate–light petroleum (2:3)] to give the colourless solid **20** as an inseparable mixture of *E*- and *Z*-isomers (29 mg, 95%) (Found: M^+, 321.139. $C_{17}H_{23}NO_3S$ requires M, 321.139); ν_{max}($CHCl_3$)/cm^{-1} 1668 and 1600; δ_H(400 MHz; $CDCl_3$) 7.73 (1.2 H, part of AA′BB′, *J* 8), 7.71 (0.8 H, d, part of AA′BB′, *J* 8), 7.26 (1.2 H, d, part of AA′BB′, *J* 8), 7.23 (0.8 H, d, part of AA′BB′, *J* 8), 5.88 (0.4 H, d, *J* 2), 5.74 (0.6 H, r, *J* 1), 4.90 (0.6 H, br d, *J* 6), 4.54 (4 H, dd, *J* 10, 2), 4.38 (0.6 H, m), 4.31 (0.4 H), 3.56 (1.2 H, s), 3.53 (1.8 H, s), 2.56 (0.4 H, dd, *J* 16, 6), 2.40 (3 H, s), 2.29 (0.6 H, t, *J* 13) and 1.25–2.00 (9 H, m); *m*/*z* (EI) 321 $(M)^+$.

(E)- *and* (Z)-*t-Butyl* 2-*methoxymethylene*-9-*azabicyclo*[4.2.1]-*nonane*-9-*carboxylate* **21**. To freshly distilled liquid ammonia (30 ml) was added a solution of the enol ether **20** (61 mg, 0.19 mmol) in THF (15 ml). The solution was cooled to −78 °C, treated with lithium metal (100 mg) and, after 15 min, when a permanent blue colour formed, quenched with solid ammonium acetate (200 mg) (colour lost). The ammonia was allowed to evaporate as the reaction mixture warmed to ambient temperature. The residue was diluted with water (15 ml), made alkaline (pH 11) with aqueous sodium hydroxide (3 mol dm^{-3}) and extracted with chloroform (5 × 15 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated to give a colourless oil which was dissolved in methanol (20 ml), treated with di-t-butyl dicarbonate (124 mg, 0.57 mmol) and stirred for 16 h. The reaction mixture was then diluted with diethyl ether (30 ml), washed with aqueous phosphoric acid (0.2 mol dm^{-3}, 10 ml) and then saturated aqueous sodium hydrogen carbonate (10 ml). The aqueous washings were combined and extracted with diethyl ether (5 × 20 ml). The organic fractions were combined, dried (Na_2SO_4) and concentrated to give a colourless oil. The crude oil was purified by flash chromatography [light petroleum–ethyl acetate (4:1)] to give **21** as a clear oil and as a mixture of isomers (43 mg, 85%) (Found: M^+, 267.183. $C_{15}H_{25}NO_3$ requires M, 267.183); $\nu_{max}(CHCl_3)/cm^{-1}$ 1670; δ_H(270 MHz; $CDCl_3$) 5.89 (1 H, m), 4.90 (0.5 H, m), 4.38 (0.5 H, m), 4.30 (1 H, m), 3.55 (3 H, m), 1.45 (9 H, m) and 1.20–2.62 (10 H, m); *m/z* (CI) 267 $(M)^+$.

t-Butyl 2-*formyl*-9-*azabicyclo*[4.2.1]*non*-2-*ene*-9-*carboxylate* **22**. A stirred solution of the enol ethers **21** (40 mg, 0.15 mmol) in CH_2Cl_2 (4 ml) at −30 °C was treated with a solution of benzeneselenenyl chloride (32 mg, 0.16 mmol) in CH_2Cl_2 (1 ml). After 30 min, the reaction mixture was cooled to −78 °C and treated with a solution of *meta*-chloroperbenzoic acid (MCPBA) (90%, 34.5 mg, 0.18 mmol) in CH_2Cl_2 (1 ml). The reaction mixture was stirred for 10 min and then quenched with water (5 ml), and allowed to warm to ambient temperature; aqueous sodium hydroxide (0.25 ml; 3 mol dm^{-3}) and saturated aqueous sodium bisulphite (0.25 ml) were then added. The mixture was extracted with CH_2Cl_2 (5 × ml) and the organic extracts were combined, washed with brine (5 ml), dried (Na_2SO_4) and concentrated. The residue was purified by flash chromatography [light petroleum–ethyl acetate (2:1)] to give **22** as a colourless oil (23 mg, 61%) (Found: M^+, 251.154. $C_{14}H_{21}NO_3$ requires M, 251.152); $\nu_{max}(CHCl_3)/cm^{-3}$ 1665; δ_H(270 MHz; $CDCl_3$) (conformational isomers present) 9.38 (0.25 H, s), 9.34 (0.75 H, s), 6.72 (0.75 H, t, *J* 7.8), 6.64 (0.25 H, t, *J* 5.3), 5.10 (0.25 H, m), 5.03 (0.75 H, d, *J* 8.6), 4.45 (0.75 H, br m), 4.33 (0.75 H, m), 2.49–2.62 (2 H, m), 2.02–2.33 (3 H, m), 1.55–1.76 (3 H, m), 1.42 (2.25 H, s) and 1.37 (6.75, s); *m/z* (EI) 251 $(M)^+$.

t-Butyl 2-*acetyl*-9-*azabicylo*[4.2.1]*non*-2-*ene*-9-*carboxylate* (N-*BOC anatoxin-a*) **23**. To a stirred solution of the aldehyde **22** (11 mg, 0.04 mmol) in diethyl ether (4 ml) at −78 °C was added a solution of methylmagnesium iodide (0.1 mmol) in diethyl ether (1 ml). The reaction mixture was allowed to warm to −40 °C during 10 min after which it was quenched with saturated aqueous ammonium chloride (5 ml). After warming to ambient temperature, this mixture was extracted with ethyl acetate (5 × 5 ml), the organic extracts were combined, dried (Na_2SO_4) and concentrated. The residue was dissolved in CH_2Cl_2 (2.5 ml) and treated with anhydrous sodium acetate (16.5 mg, 0.2 mmol) and pyridinium chlorochromate (43 mg, 0.2 mmol). The reaction mixture was stirred for 30 min at ambient temperature and then diluted with CH_2Cl_2 (20 ml), filtered through Florisil and concentrated. The residue was purified by flash chromatography [light petroleum–ethyl acetate (2:1)] to give **23** as a colourless oil (3.3 mg, 37%). The spectral data obtained for **23** agreed with that previously reported by Rapoport.[4f]

(±)-*Anatoxin-a* **1**. *N*-BOC anatoxin-a (3 mg, 0.012 mmol) was dissolved in a solution of concentrated hydrochloric acid in ethyl acetate (3 mol dm^{-3}; 1 ml) and allowed to stand at room temperature for 2 h. The solvents were removed under reduced pressure and the residue purified by passage through a short silica gel column, eluting with $CHCl_3$–MeOH (9:1), to give anatoxin-a hydrochloride as a colourless glass (2.3 mg, 98%). The spectral data obtained for **1**·HCl agreed with that previously reported by Rapoport.[4f]

(E)- *and* (Z)-2-(1-*Methoxyethylidene*)-9-(p-*tolylsulphonyl*)-9-*azabicyclo*[4.2.1]*nonane* **24**. To a stirred solution of diisopropylamine (56.6 mg, 78 μl, 0.56 mmol) in DME (2 ml) at −50 °C was added a solution of butyllithium in hexane (1.6 mol dm^{-3}; 330 μl, 0.53 mmol). The reaction mixture was stirred for 10 min and then cooled to −78 °C and treated with a solution of phosphine oxide **19** (146 mg, 0.56 mmol) in DME (2 ml). Immediately a deep-red colour formed, and after 20 min the reaction mixture was treated with a solution of the ketone **17** (41 mg, 0.14 mmol) in DME (2 ml). The solution was stirred for 5 min and then quenched with saturated aqueous ammonium chloride (4 ml) when the colour was lost. The mixture was warmed to ambient temperature, diluted with water (2 ml) and extracted with ethyl acetate (5 × 5 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated to give the diastereoisomeric hydroxyphosphine oxides as a colourless solid. This solid was dissolved in THF (5 ml) and the solution stirred and treated with sodium hydride (34 mg, 1.4 mmol) at ambient temperature for 14 h. After this the reaction mixture was cooled to 0 °C, carefully quenched with water (3 ml), and extracted with ethyl acetate (5 × 5 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated and the residue purified by flash chromatography [ethyl acetate–light petroleum (2:3)] to give **24** as a colourless solid (43.6 mg, 97%). The enol ether **24** was a mixture of *E*- and *Z*-isomers. A small amount of this mixture was purified by preparative TLC [light petroleum–ethyl acetate (4:1)] and the two isomers isolated in pure form but were not unambiguously assigned. Isomer A (major component of 4:3 mixture): R_f [light petroleum–ethyl acetate (2:1)] 0.57 (Found: M^+, 335.159. $C_{18}H_{25}NO_3S$ requires M, 335.155); $\nu_{max}(CHCl_3)/cm^{-1}$ 1660, 1600; δ_H(400 MHz; $CDCl_3$) 7.72 (2 H, d, part of AA′BB′, *J* 8), 7.27 (2 H, d, part of AA′BB′, *J* 8), 5.07 (1 H, d, *J* 9), 4.35 (1 H, m), 4.00 (3 H, s), 2.40 (3 H, s), 2.17–2.33 (2 H, m), 1.70–1.85 (2 H, m), 1.77 (3 H, t, *J* 1) and 1.25–1.60 (6 H, m); *m/z* (EI) 335 $(M)^+$.

Minor component: R_f [light petroleum–ethyl acetate (2:1)] 0.55 (Found: M^+, 335.160. $C_{18}H_{25}NSO_3$ requires M, 335.155); $\nu_{max}(CHCl_3)/cm^{-1}$ 1660 and 1600; δ_H(400 MHz; $CDCl_3$) 7.72 (2 H, d, part of AA′BB′, *J* 8), 7.27 (2 H, d, part of AA′BB′, *J* 8), 4.77 (1 H, d, *J* 9), 4.35–4.41 (1 H, m), 3.45 (3 H, s), 2.87 (1 H, dd, *J* 15, 8), 2.41 (3 H, s), 1.72–2.01 (3 H, m), 1.78 (3 H, d, *J* 2) and 1.46–1.68 (6 H, m); *m/z* (EI) 335 $(M)^+$.

(E)- *and* (Z)-*t-Butyl* 2-(1-*Methoxyethylidene*)-9-*azabicyclo*-[4.2.1]*nonane*-9-*carboxylate* **25**.—To freshly distilled liquid ammonia (5 ml) was added a solution of the enol ethers **24** (12 mg, 0.04 mmol) in THF (1 ml). The stirred solution was cooled at −78 °C, treated with lithium metal (20 mg) and after 3 min, when a permanent blue colour formed, quenched with solid ammonium acetate (50 mg) (colour lost). The ammonia was allowed to evaporate as the mixture warmed to ambient temperature. The residue was then diluted with water (5 ml), made alkaline (pH 10) with aqueous sodium hydroxide (3 mol dm^{-3}) and extracted with chloroform (5 × 5 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated to a colourless oil. This crude oil was dissolved in methanol (2 ml),

treated with di-t-butyl dicarbonate (25 mg, 0.11 mmol) and stirred for 16 h. The reaction mixture was then diluted with diethyl ether (4 ml), washed with aqueous phosphoric acid (0.2 mol dm^{-3}; 2 ml) and then saturated aqueous sodium hydrogen carbonate (2 ml). The aqueous washings were extracted with diethyl ether (5 × 2 ml) and the organic phases were combined, dried (Na_2SO_4) and concentrated to give a colourless oil. The crude oil was purified by flash chromatography [light petroleum–ethyl acetate (4:1)] to give **25** as a clear oil (5.5 mg, 55%); $\nu_{max}(CHCl_3)/cm^{-1}$: 1660; δ_H(270 MHz; $CDCl_3$) 4.97 (0.5 H, m), 4.69 (0.5 H, m), 4.31 (1 H, m), 3.48 (3 H, m), 2.93 (0.5 H, m), 1.81 (3 H, br s), 1.42 (9 H, m) and 1.30–2.40 (9.5 H, m). ^{1}H NMR data for **25** was extremely complex due to amide resonance and the presence of *E* and *Z* isomers. The sample was not characterised further.

(α)- + (β)-*t-Butyl 2-Acetyl-9-azabicyclo*[4.2.1]*nonane-9-carboxylate Ester* (N-*BOC Dihydroanatoxin-a*) **26**.—To a stirred solution of sodium iodide (4.1 mg, 0.03 mmol) and the enol ether **25** (5.0 mg, 0.02 mmol) in acetonitrile (0.5 ml) at ambient temperature was added trimethylsilyl chloride (2.9 mg, 0.03 mmol, 3.5 μl). After 5 min the reaction mixture was quenched with aqueous sodium thiosulphate (1 mol dm^{-3}; 2 ml) and extracted with ethyl acetate (5 × 2 ml). The organic extracts were combined, washed with brine (4 ml), dried (Na_2SO_4) and concentrated and the residue purified by flash chromatography to give **26** (3.5 mg, 65%) as a clear oil and as a 2:5 mixture of α and β isomers. The spectral data obtained for **26** agreed with that reported previously by Rapaport.[4f]

(R)-*Ethyl* 2-(p-*Tolylsulphonylamino*)*hepta*-5,6-*dienoate* (R)-**8a**.—Water (750 ml) was slowly added to a stirred solution of (±)-**8a** (1.0 g, 3.08 mmol) in acetone (85 ml) to give a very fine suspension. The suspension was treated with α-chymotrypsin I (25 mg) (purchased from Sigma) and the pH of the reaction mixture kept at 7.2 by addition of aqueous sodium hydroxide (1.05 mol dm^{-3}). After 8 h, when 0.5 mol equiv. of alkali had been added, the pH of the reaction mixture was raised to 11 in order to denature the enzyme. The mixture was then neutralised by addition of 2 mol dm^{-3} hydrochloric acid and lyophilised. The residue was suspended in aqueous sodium hydroxide (3 mol dm^{-3}; 400 ml) and extracted with diethyl ether (3 × 200 ml). The combined organic extracts were washed with brine (200 ml), dried (Na_2SO_4) and concentrated to give the resolved ester (*R*)-**8a** as a colourless crystalline solid (480 mg, 48%), m.p. 50–50.5 °C (diethyl ether–light petroleum); $[\alpha]_D^{20}$ −39.8° (*c* 1.1, $CHCl_3$). IR and ^{1}H NMR data for (*R*)-**8a** were identical with those for the racemic ester. (*R*)-**8a** was shown to be >95% pure by a chiral shift study employing tris[3- (heptafluoropropylhydroxymethylene)-(+)-camphorato] europium.

The alkaline aqueous layer obtained above was cooled to 0 °C, acidified with concentrated hydrochloric acid and extracted with diethyl ether (3 × 200 ml). The combined organic extracts were washed with brine (200 ml), dried (Na_2SO_4) and concentrated to give (*S*)-2-(*p*-tolylsulphonylamino)hepta-5,6-dienoic acid **27** as a colourless crystalline solid (410 mg, 45%), m.p. 109.9–110.2 °C (ethyl acetate–light petroleum); $[\alpha]_D^{20}$ +20.1° (*c* 2.8, $CHCl_3$). IR and ^{1}H NMR data for this material were identical with those for the racemic acid (see below); the optical purity (>95%) was established by ^{1}H NMR analysis using (*R*)-α-methylbenzylamine as a shift reagent.

2-(p-*Tolylsulphonylamino*)*hepta*-5,6-*dienoic Acid* (±)-**27**.—A solution of the allenic ester **8a** (323 mg, 1 mmol) in THF (5 ml) and water (3 ml) was treated with lithium hydroxide monohydrate (158 mg, 3.8 mmol). The reaction mixture was stirred at ambient temperature for 60 h and then diluted with water (15 ml) and washed with CH_2Cl_2 (15 ml). The aqueous solution was acidified to pH 2 with hydrochloric acid (2 mol dm^{-3}) and extracted with CH_2Cl_2 (4 × 20 ml). The organic extracts were combined, dried (Na_2SO_4) and concentrated to give the title compound (±)-**27** as a colourless solid (170 mg, 57%), m.p. 107–109 °C (benzene–light petroleum) (Found: C, 57.1; H, 5.7; N, 5.1. $C_{14}H_{17}NO_4S$ requires C, 56.93; H, 5.80; N, 4.74%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1955 and 1725; δ_H(270 MHz; $CDCl_3$) (CO_2H was not observed) 7.35 (2 H, d, part of AA′BB′, *J* 8), 7.28 (2 H, d, part of AA′BB′, *J* 8), 5.53 (1 H, br s), 5.01 (1 H, quint., *J* 6), 4.66 (2 H, dt, *J* 6, 3), 3.93 (1 H, br q, *J* 4), 2.40 (3 H, s) and 1.64–2.12 (4 H, m); *m/z* (CI) 296 $(M + 1)^+$.

Correlation of Absolute Configuration of (R)-(−)-**8a**.—A solution of (−)-**8a** (206 mg, 0.637 mmol) in ethyl acetate (10 ml) was cooled to −78 °C and treated with an excess of ozone to form a blue solution. After 2.5 h, a small excess of 30% H_2O_2 was added and the mixture was heated at reflux for 2.5 h. After this time, the solution was cooled and extracted with 1 mol dm^{-3} NaOH (4 × 10 ml). The extracts were acidified with concentrated HCl, extracted with ethyl acetate (4 × 10 ml), and the combined organic extracts were dried (Na_2SO_4) and concentrated. Purification of the residue by flash chromatography gave (R)-(−)-**28** as a clear pale yellow oil; $[\alpha]_D^{20}$ −31.8° (*c* 1.2, $CHCl_3$).

A sample of ()-(+)-**28** was prepared from glutamic acid using literature procedures[26] and had $[\alpha]_D^{20}$ +35.8° (*c* 1.7, $CHCl_3$).

Acknowledgements

We thank the Science and Engineering Research Council and the University of Bath for financial support and Dr O. Howarth (University of Warwick/SERC Central NMR Facility) for his help.

References

1 C. S. Huber, *Acta Crystallogr., Sect. B*, 1972, **28**, 2577; W. W. Carmichael, D. F. Biggs and P. R. Gorham, *Science*, 1975, **187**, 542; J. P. Devlin, O. E. Edwards, P. R. Gorham, N. Hunter, R. K. Pike and B. Stavric, *Can. J. Chem.*, 1977, **55**, 1367. For recent biosynthetic studies relating to anatoxin-a, see: J. R. Gallon, K. N. Chit and E. G. Brown, *Phytochemistry*, 1990, **29**, 1107.

2 G. A. Codd and G. K. Poon, in *Biochemistry of Algae and Cyanobacteria*, ed. L. J. Rogers and J. R. Gallon, Oxford University Press, Oxford, 1988, p. 283. The impact of toxic strains of *Anabaena flos-aquae* on fresh water lakes has been recorded: E. T. Rose, *Proc. Iowa Acad. Sci.*, 1953, **60**, 738.

3 C. E. Spivak, B. Witkop and E. X. Albuquerque, *Mol. Pharmacol.*, 1980, **18**, 384; C. E. Spivak, J. Waters, B. Witkop and E. X. Albuquerque, *Mol. Pharmacol.*, 1983, **23**, 337; K. L. Swanson, C. N. Allen, R. S. Aronstam, H. Rapoport and E. X. Albuquerque, *Mol. Pharmacol.*, 1985, **29**, 250; X. Zhang, P. Stjernlöf, A. Adem and A. Nordberg, *Eur. J. Pharmacol.*, 1987, **135**, 457.

4 (*a*) H. F. Campbell, O. E. Edwards and R. Kolt, *Can. J. Chem.*, 1977, **55**, 1372; (*b*) H. F. Campbell, O. E. Edwards, J. W. Elder and R. Kolt, *Pol. J. Chem.*, 1979, **53**, 27; (*c*) H. A. Bates and H. Rapoport, *J. Am. Chem. Soc.*, 1979, **101**, 1259; (*d*) J. S. Peterson, S. Töteberg-Kaulen and H. Rapoport, *J. Org. Chem.*, 1984, **49**, 2948; (*e*) J. J. Tufariello, H. Meckler, and K. P. A. Senaratne, *J. Am. Chem. Soc.*, 1984, **106**, 7979; (*f*) J. S. Petersen, G. Fels and H. Rapoport, *J. Am. Chem. Soc.*, 1984, **106**, 4539; (*g*) R. L. Danheiser, J. M. Morin and E. J. Salaski, *J. Am. Chem. Soc.*, 1985, **107**, 8066; (*h*) J. R. Wiseman and S. Y. Lee, *J. Org. Chem.*, 1986, **51**, 2485; (*j*) K. H. Melching, H. Hiemstra, W. J. Klaver and W. N. Speckamp, *Tetrahedron Lett.*, 1986, **27**, 4799; (*k*) T. Shono, Y. Matsumura, K. Uchida and K. Tagami, *Chemistry Lett.*, 1987, 919; (*l*) B. Lindgren, P. Stjernlöf and L. Trogen, *Acta Chem. Scand., Ser. B*, 1987, **41**, 180. For the resolution of anatoxin-a, see: P. Stjernlöf, L. Trogen and A. Andersson, *Acta Chem. Scand., Ser. B*, 1989, **43**, 917.

5 For leading references to the synthesis and biological activity of derivatives of anatoxin-a, see: (*a*) D. B. Kanne and L. G. Abood, *J. Med. Chem.*, 1988, **31**, 506; (*b*) F. J. Sardina, M. H. Howard, A. M. P. Koskinen and H. Rapoport, *J. Org. Chem.*, 1989, **54**, 4654; (*c*) M. H.

Howard, F. J. Sardina and H. Rapoport, *J. Org. Chem.*, 1990, **55**, 2829; (*d*) K. L. Swanson, H. Rapoport, R. S. Aronstam and E. X. Albuquerque, *Amer. Chem. Soc. Symp. Ser.*, 1990, **418**, 107. For a discussion of the conformational and other physical properties of anatoxin-a, see: (*e*) B. Witkop and A. Brossi, in *Natural Products and Drug Development*, Alfred Benzon Symposium 20, ed. P. Krogsgaard-Larson, S. Christensen and H. Kofod, Munksgaard, Copenhagen, 1984; (*f*) A. M. P. Koskinen and H. Rapoport, *J. Org. Chem.*, *J. Med. Chem.*, 1985, **28**, 1301.

6 Preliminary communications, see: (*a*) R. Kinsman, D. Lathbury, P. Vernon and T. Gallagher, *J. Chem. Soc., Chem. Commun.*, 1987, 243; (*b*) P. Vernon and T. Gallagher, *J. Chem. Soc., Chem Commun.*, 1973, 245.

7 P. A. Bartlett, in *Asymmetric Synthesis*, ed. J. D. Morrison, Academic Press, New York, 1984, vol. 3, p. 411; L. S. Hegedus, *Tetrahedron*, 1984, **40**, 2415.

8 For an overview of the reactivity of allenes towards electrophiles, including a discussion of possible mechanistic pathways, see T. L. Jacobs, in *The Chemistry of the Allenes*, ed. S. R. Landor, Academic Press, 1982, vol. 2, p. 349; W. Smadja, *Chem. Rev.*, 1983, **83**, 263.

9 D. Lathbury, R. W. Shaw, P. A. Bates, M. B. Hursthouse and T. Gallagher, *J. Chem. Soc., Perkin Trans. 1*, 1989, 2415.

10 D. Lathbury and T. Gallagher, *J. Chem. Soc., Chem. Commun.*, 1986, 114.

11 (*a*) D. L. J. Clive, V. Farina, A. Singh, C. K. Wong, W. A. Kiel and S. M. Menchen, *J. Org. Chem.*, 1980, **45**, 2120; (*b*) K. E. Harding and S. R. Burks, *J. Org. Chem.*, 1981, **46**, 3920; (*c*) J. Barluenga, C. Jiménez, N. Nájera and M. Yus, *J. Chem. Soc., Chem. Commun.*, 1981, 1178; (*d*) J. Barluenga, C. Nájera and M. Yus, *J. Heterocycl. Chem.*, 1981, **18**, 1297; (*e*) S. Danishefsky, E. Taniyama and R. R. Webb, *Tetrahedron Lett.*, 1983, **24**, 11; (*f*) M. B. Gasc, A. Lattes and J. J. Perie, *Tetrahedron*, 1983, **39**, 703; (*g*) J. Barluenga, C. Jiménez, C. Nájera and M. Yus, *J. Heterocycl. Chem.*, 1984, **21**, 1733; (*h*) J. Barluenga, C. Jiménez, C. Nájera and M. Yus, *J. Chem. Soc., Perkin Trans. 1*, 1984, 721; (*i*) K. E. Harding and S. R. Burks, *J. Org. Chem.*, 1984, **49**, 40; (*j*) K. E. Harding and T. H. Marman, *J. Org. Chem.*, 1984, **49**, 2838; (*k*) A. Toshimitsu, K. Terao and S. Uemura, *J. Org. Chem.*, 1986, **51**, 1724; (*l*) M. Tokuda, Y. Yamada and H. Suginome, *Chemistry Lett.*, 1988, 1289.

12 A. Claesson, C. Sahlberg and K. Luthman, *Acta Chem. Scand. B*, 1979, **33**, 309; L.-I. Olsson and A. Claesson, *Synthesis*, 1979, 743.

13 G. M. Whitesides, C. P. Casey and J. K. Krieger, *J. Am. Chem. Soc.*, 1971, **93**, 1379.

14 R. C. Larock, *Tetrahedron*, 1982, **28**, 1713.

15 G. Stork, A. Y. W. Leong and A. M. Touzin, *J. Org. Chem.*, 1976, **41**, 3491.

16 G. Gignarella and G. Nathansohn, *J. Org. Chem.*, 1961, **26**, 1500; G. Gignarella and G. Nathansohn, *Gazz. Chim. Ital.*, 1960, **190**, 1495; Y. Kawanami, Y. Ito, T. Kitagawa, Y. Taniguchi, T. Katsuki and M. Yamaguchi, *Tetrahedron Lett.*, 1984, **25**, 857 and references therein.

17 S. C. Rychnovsky and P. A. Bartlett, *J. Am. Chem. Soc.*, 1981, **103**, 3963.

18 F. Cooke and P. Magnus, *J. Chem. Soc., Chem. Commun.*, 1976, 519.

19 S. Trippett, *J. Chem. Soc.*, 1961, 2813.

20 (*a*) M. Schlosser and H. B. Thuong, *Chimia*, 1976, **30**, 197; (*b*) C. Earnshaw, C. J. Wallis and S. Warren, *J. Chem. Soc., Perkin Trans. 1*, 1979, 3099.

21 (*a*) B. M. Trost, C. G. Caldwell, E. Muayama, and D. Heissler, *J. Org. Chem.*, 1983, **48**, 3252; (*b*) M. Maleki, A. Miller and O. W. Lever, *Tetrahedron Lett.*, 1981, **22**, 365—these authors report the melting point of **19** as 60–62 °C.

22 K. C. Nicolaou, R. L. Magolda and W. J. Sipio, *Synthesis*, 1979, 982.

23 G. Rousseau, P. Le Perchec, J. M. Conia, *Synthesis*, 1976, 67; G. Neef, U. Eder, A. Seeger and R. Wiechert *Chem. Ber.*, 1980, **113**, 1184; E. W. H. Asveld and R. M. Kellogg, *J. Am. Chem. Soc.*, 1980, **102**, 3644.

24 R. M. Williams, in *Synthesis of Optically Active α-Amino Acids*, Pergamon, Oxford, 1989. For an alternative synthesis of enantiomerically pure allenic amino acids, see: J. E. Baldwin, R. M. Adlington and A. Basak, *J. Chem. Soc., Chem. Commun.*, 1984, 1284. For another route to enantiomerically pure derivatives of **9** and **10**, see: T. Ohta, A. Hosoi, T. Kimura and S. Nozoe, *Chemistry Lett.*, 1987, 2091.

25 J. B. Jones and J. F. Beck, in *Techniques of Chemistry: Applications of Biochemical Systems in Organic Synthesis*, eds. J. B. Jones, C. J. Sih and D. Perlman, Wiley-Insterscience, New York, 1976, vol. 10, pt. 1, p. 107.

26 C. R. Harrington and R. C. G. Moggridge, *J. Chem. Soc.*, 1940, 706; G. H. L. Nefkens and R. J. F. Nivard, *Recl. Trav. Chim. Pays-Bas*, 1964, **83**, 199.

27 W. G. Dauben and G. Shapiro, *J. Org. Chem.*, 1984. **49**, 4252.

Paper 0/03267K
Received 20*th July* 1990
Accepted 21*st August* 1990