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Synthesis of Analogues of GABA. IV* Three Unsaturated Derivatives of 3-Aminocyclopentane-1-carboxylic Acid

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Abstract

cis-4-Aminocyclopent-2-ene-1-carboxylic acid (4), 4-aminocyclopent-1-ene-1-carboxylic acid (5) and 3-aminocyclopent-1-ene-1-carboxylic acid (6) have been synthesized as conformationally restricted analogues of the inhibitory neurotransmitter GABA.

Neurotransmitter analogues of restricted conformation can provide considerable information on particular 'active conformations' of the neurotransmitter which are involved in various processes such as cellular uptake, receptor binding and enzymic transformations. For the inhibitory neurotransmitter γ-aminobutyric acid (GABA) (1),[1,2] four general classes of analogues have been used to restrict the conformation of GABA: (i) aliphatic derivatives with bulky substituents, (ii) derivatives with double or triple carbon–carbon bonds, (iii) carbocyclic analogues, and (iv) derivatives with heterocyclic ring systems.

Considerable information has been derived from the carbocyclic analogues where the C2 and C4 atoms of GABA have been linked by a carbon chain.[3] In particular, the *cis*- and *trans*-cyclopentane derivatives (2) and (3)[4,5] have a potent inhibitory action on neuronal firing in the mammalian central nervous system. However, the conformation of GABA is not rigidly fixed in these analogues because of the considerable conformational mobility of the cyclopentane ring.[6] The introduction of a double bond into the cyclopentane ring as an extra conformational constraint should reduce the flexibility of such derivatives. Cyclopentene analogues of GABA were therefore required for further investigations of active conformations that may lead

* Part III, *Aust. J. Chem.*, 1979, **32**, 2517.

[1] Johnston, G. A. R., in 'GABA and Nervous System Function' (Eds E. Roberts, T. N. Chase and D. B. Tower), p. 395 (Raven Press: New York 1975).

[2] Saclens, J. I., and Vinick, F. J., *Annu. Rep. Med. Chem.*, 1978, **13**, 31.

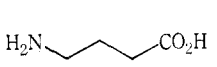
[3] Johnston, G. A. R., Allan, R. D., Kennedy, S. M. E., and Twitchin, B., in 'GABA-Neurotransmitters' (Eds P. Krogsgaard-Larsen, J. Scheel-Kruger and H. Kofod), p. 149 (Munksgaard: Copenhagen 1978).

[4] Johnston, G. A. R., Allan, R. D., Andrews, P. D., Kennedy, S. M. E., and Twitchin, B., in 'Advances in Pharmacology and Therapeutics' Vol. 12: Neurotransmitters (Ed. P. Simon), p. 11 (Pergamon Press: Oxford 1978).

[5] Allan, R. D., Johnston, G. A. R., and Twitchin, B., *Aust. J. Chem.*, 1979, **32**, 2517.

[6] Fuchs, B., in 'Topics in Stereochemistry' (Eds E. L. Eliel and N. L. Allinger), p. 1 (Interscience: New York 1978).

to agents which have selective actions on particular processes involving GABA. In this paper, we report the synthesis of the three cyclopentene GABA analogues (4), (5) and (6).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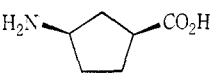
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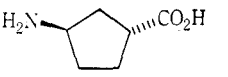
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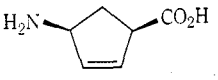
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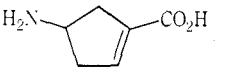
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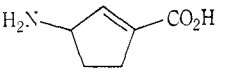
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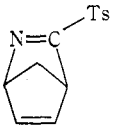
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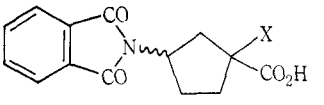
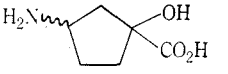

(8) X = H
(9) X = Br


(10)

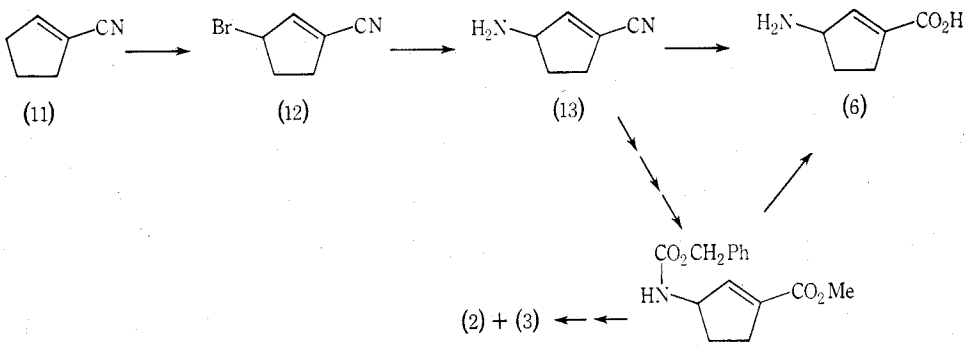
A convenient source of cyclopentene γ-amino acids follows from the reported Diels–Alder addition of tosyl nitrile to cyclopentadiene.[7] The intermediate (7), which had previously been converted into the saturated *cis* amino acid (2) by reduction of the unsaturated lactam,[7] readily underwent hydrolysis to *cis*-4-aminocyclopent-2-ene-1-carboxylic acid (4). Difficulties in obtaining a pure product were overcome by crystallization of amino acid (4) as its hydrochloride from acetic acid. An unexpected feature noted by ^{1}H n.m.r. spectroscopy during the isolation of (4) was its ready esterification if alcoholic solvents were used during attempted crystallization. Isomerization of (4) with 2 M sodium hydroxide under nitrogen gave the conjugated isomer 4-aminocyclopent-1-ene-1-carboxylic acid (5).

A possible route to a further unsaturated amino acid (6) by bromination of the nitrogen-protected cyclopentane derivative (8) and subsequent dehydrohalogenation of compound (9) gave rise to mixtures of the acids (5) and (6) which could not be readily separated. Attempted dehydrohalogenation and deprotection of (9) in refluxing 20% hydrobromic acid gave the hydroxy amino acid (10) as the major product, while refluxing 48% hydrobromic acid gave mainly (5) and (6) as a 2:1 mixture which co-crystallized.

A different synthetic route to amino acid (6) in which the double bond was formed prior to the introduction of the amino group was therefore investigated. A reaction sequence involving allylic bromination of α,β-unsaturated acids followed by bromine displacement with ammonia has previously been used successfully in this laboratory.[8,9] When applied to cyclopent-1-ene-1-carboxylic acid complex mixtures were formed. A contributing factor to the failure of this sequence was the formation of

[7] Jagt, J. C., and van Leusen, A. M., *J. Org. Chem.*, 1974, **39**, 564.
[8] Allan, R. D., and Twitchin, B., *Aust. J. Chem.*, 1978, **31**, 2283.
[9] Allan, R. D., *Aust. J. Chem.*, 1979, **32**, 2507.

the two possible products of allylic bromination (δ 7·1 and 7·3 for =CH resonances) in a ratio of 4:3. Furthermore the high reactivity of allylic cyclopentene halides[10] would be further increased by carboxylate anion formation, possibly leading to significant secondary amine by-products in the amination step.[8] The reaction scheme was therefore modified by using cyclopent-1-ene-1-carbonitrile (11) as shown in Scheme 1 rather than the corresponding acid. Reaction of (11) with *N*-bromosuccinimide was followed by ^{1}H n.m.r. spectroscopy and gave an approximately 3:1 mixture of two products (δ 6·85 and 6·95 for =CH resonances), presumably (12) and the other possible product of allylic bromination, 5-bromocyclopent-1-ene-1-carbonitrile. Amination of the crude mixture in liquid ammonia at $-60°$ to $-70°$ was complete in less than 80 min and, after removal of the excess ammonia, the crude product (13) was hydrolysed without delay to (6). The amino acid from the by-product hindered direct purification of (6) but derivatization as the *N*-benzyloxycarbonyl methyl ester gave (14) which was readily crystallized and regenerated the required amino acid (6) on hydrolysis. To preclude the possibility that the isolated product was 5-aminocyclopent-1-ene-1-carboxylic acid from the alternative allylic bromination product, (14) was reduced catalytically to a mixture of (2) and (3) from which (3) was isolated by crystallization.



Scheme 1

The three cyclopentene GABA analogues have been tested as inhibitors of the sodium-dependent uptake, the sodium-independent binding and the enzymic transamination of radioactive GABA by rat brain tissue[3] *in vitro*, as well as microelectrophoretically on bicuculline-sensitive postsynaptic receptors on dorsal horn neurones in the cat spinal cord *in vivo*. The results will be reported in detail elsewhere and are briefly summarized here. The allylic amino acid (4) was a time-dependent inhibitor of the enzymic transamination of GABA in keeping with its structural analogy with γ-vinyl GABA,[2] but was weak compared to GABA in uptake, binding, and postsynaptic receptors. Compound (5) was very much more potent than GABA at depressing neuronal firing *in vivo* and was active *in vitro* on binding and uptake with a potency similar to that of GABA. On the other hand (6) displayed some selectivity with an activity similar to that of GABA on the uptake system and a lower potency than GABA

[10] Green, M., Knox, G. R., and Paulson, P. L., in 'Rodd's Chemistry of Carbon Compounds' 2nd Edn., Vol. 2A, p. 149 (Elsevier: Amsterdam 1967).

on the GABA binding assay. The most notable feature of these results was the high potency of (5), comparable with that of the saturated derivatives (2) and (3) on GABA binding *in vitro* and postsynaptic receptors *in vivo*.

Experimental

General experimental details have been given in Part III.[5] Thin-layer chromatography (t.l.c.) in butanol/acetic acid/water, 4:1:1, on Merck kieselgel 60 precoated t.l.c. plates gave for the following compounds the R_F values (after visualization with ninhydrin): (4), HCl 0·27; (5) 0·28; (6) 0·30; (10) 0·23; (2) 0·30 and (3) 0·37.

cis-*4-Aminocyclopent-2-ene-1-carboxylic Acid* (*4*)

Following the procedure of Jagt and van Leusen,[7] a solution of tosyl cyanide[11] (1·0 g, 5·5 mmol) in freshly distilled cyclopentadiene (20 ml) was stirred at room temperature for 30 min and evaporated to dryness without heating. The crude product was refluxed with 2 M hydrochloric acid (60 ml) for 2 h and the oily layer removed by extraction with methylene chloride (2 × 20 ml). The aqueous layer was evaporated to dryness under vacuum, dissolved in water (20 ml) and absorbed onto a column of Dowex 50(H^+) ion-exchange resin (50 ml). After washing with water the amino acid was eluted with 1 M ammonium hydroxide and evaporated to dryness. Crystallization from acetic acid/ethyl acetate gave the neutral amino acid (590 mg, 84%), m.p. 156–170° (dec.), which appeared to be of high purity by ^{1}H n.m.r. spectroscopy and t.l.c. but slowly turned brown on storage and could not be obtained analytically pure. The compound was recrystallized from acetic acid as its hydrochloride, cis-*4-aminocyclopent-2-ene-1-carboxylic acid hydrochloride*, m.p. 167–170° (Found: C, 44·0; H, 6·2; Cl, 21·5; N, 8·9. $C_6H_{10}ClNO_2$ requires C, 44·0; H, 6·2; Cl, 21·7; N, 8·6%). ν_{max} 3100, 1700, 1610w, 1220, 845, 770w, 755w, 700w cm^{-1}. ^{1}H n.m.r. δ (D_2O, external $SiMe_4$) 6·7, m, 1H, =CH; 6·45, m, 1H, =CH; 4·9, m, $CHNH_2$; 4·2, m, $CHCO_2H$; 3·4–2·9, m, 1H, CH_2; 2·8–2·3, m, 1H, CH_2.

4-Aminocyclopent-1-ene-1-carboxylic Acid (*5*)

The hydrochloride of amino acid (4) (1·0 g, 6 mmol) was refluxed under nitrogen in 2 M sodium hydroxide solution (100 ml) for 2 h. After cooling, the solution was brought to pH 5 with hydrochloric acid and absorbed on Dowex 50(H^+) ion-exchange resin (50 ml). The column was washed with water, and after elution with 1 M ammonium hydroxide the product was evaporated to dryness. Two recrystallizations from aqueous ethanol gave *4-aminocyclopent-1-ene-1-carboxylic acid* (5), m.p. >300° (Found: C, 56·7; H, 7·1; N, 11·0. $C_6H_9NO_2$ requires C, 56·7; H, 7·1; N, 11·0%). ν_{max} 2180, 1635, 1120w, 1100w, 760, 730s cm^{-1}. ^{1}H n.m.r. δ (D_2O, external $SiMe_4$) 6·7, m, =CH; 4·5, m, $CHNH_2$; 3·8–2·8, complex, 4H, $2CH_2$.

Dehydrobromination of 1-Bromo-3-phthalimidocyclopentane-1-carboxylic Acid (*9*)

cis-3-Aminocyclopentane-1-carboxylic acid[12] (14 g, 11 mmol) was mixed with phthalic anhydride (16 g, 11 mmol) and heated to 160° for 30 min. The product was allowed to cool, dissolved in chloroform and crystallized by the addition of 60–80° light petroleum to yield cis-*3-phthalimidocyclopentane-1-carboxylic acid* (8) (16 g, 61%), m.p. 132–134° (Found: C, 64·6; H, 5·1; N, 5·3. $C_{14}H_{13}NO_4$ requires C, 64·9; H, 5·1; N, 5·4%). ν_{max} 1760w, 1705s, 705 cm^{-1}.

Phthalimido acid (8) (17 g, 62 mmol) was mixed with red phosphorus (0·7 g) and bromine (7 ml, 136 mmol) and heated on a mantle for 4 h. After cooling, water was added and then decanted off. The remaining product was dissolved in ethyl acetate, precipitated with 60–80° light petroleum and recrystallized from ethyl acetate to give 1-bromo-3-phthalimidocyclopentane-1-carboxylic acid (9) (10 g, 45%), m.p. 167–170°. ν_{max} 1735, 1700s, 710 cm^{-1}.

The bromo phthalimido acid (9) (2·7 g) was refluxed for 16 h in 48% hydrobromic acid (50 ml). Normal isolation procedures as previously described on Dowex 50(H^+) ion-exchange resin and crystallization from ethanol/water gave the major product with the higher R_F on t.l.c. (660 mg, 65%), m.p. 266–268°. Analysis of the product by g.l.c. of the trifluoroacetyl methyl esters[5] and by ^{1}H n.m.r. and i.r. spectroscopy showed the product to be a 2:1 mixture of (5) and (6).

[11] Cox, J. M., and Ghosh, R., *Tetrahedron Lett.*, 1969, 3351.
[12] Berger, H., Paul, H., and Hilgetag, G., *Chem. Ber.*, 1968, **101**, 1525.

On repeating the procedure with 20% hydrobromic acid, (9) (2·0 g) formed a lower R_F compound as the major product. A similar isolation procedure and crystallization from ethanol gave *3-amino-1-hydroxycyclopentane-1-carboxylic acid* (10) (230 mg, 26%), m.p. 276–280° (dec.) (Found: C, 49·7; H, 7·7; N, 9·2. $C_6H_{11}NO_3$ requires C, 49·7; H, 7·7; N, 9·6%). ν_{max} 3100 (br), 2150, 1640, 1600, 1520, 1220, 1180, 800, 750, 605 cm^{-1}. ^1H n.m.r. δ (D_2O) 4·4, m, 1H, $CHNH_2$; 3·4–2·1, complex, 6H, $3CH_2$.

3-Bromocyclopent-1-ene-1-carbonitrile (12)

Cyclopent-1-ene-1-carbonitrile[13] (23·3 g, 250 mmol) and freshly crystallized *N*-bromosuccinimide (49 g, 290 mmol) in carbon tetrachloride (250 ml) were refluxed under a tungsten lamp for 2·5 h. The reaction was then shown to be complete by n.m.r. spectroscopy. The solution was cooled, filtered, washed with water (3 × 200 ml), dried (Na_2SO_4) and the solvent evaporated to give crude nitrile (12) (43·7 g). ^1H n.m.r. δ ($CDCl_3$) 6·85, m, =CH; 5·2, m, CHBr; 3·1–2·3, complex, $2CH_2$; with an impurity at 6·95. In another preparation on half this scale, distillation reduced the impurity in the main fraction (b.p. 155–165°/1·5 mm) to 15–20% but also reduced the yield to 36%.

Methyl 3-(N-Benzyloxycarbonylamino)cyclopent-1-ene-1-carboxylate (14)

The crude bromo nitrile (12) (43·7 g) was dissolved in diethyl ether (100 ml) and added over 5 min to vigorously stirred liquid ammonia (600 ml) at −60 to −70°. The reaction was followed by n.m.r. spectroscopy (δ 5·2 replaced by 4·2) and was complete in 80 min. The ammonia was then removed as quickly as possible under vacuum on a rotary evaporator. After the addition of ice-water (100 ml), conc. hydrochloric acid (500 ml) was added cautiously and the resultant gummy mixture hydrolysed under reflux for 2·5 h. The solvent was then removed under vacuum and the product dissolved in water (150 ml), filtered and absorbed on Dowex 50(H^+) ion-exchange resin (350 ml). After washing with water, elution with 1 M ammonium hydroxide and evaporation gave the crude amino acid (6) (15 g) with an impurity at δ 7·3.

Half of this product was purified through the *N*-benzyloxycarbonyl methyl ester (14) as follows. Crude amino acid (6) (7·5 g) was esterified with methanolic hydrochloric acid (300 ml) for 0·5 h. After removal of the solvent the crude methyl ester hydrochloride (0·5 g, about 50 mmol) was dissolved in water (130 ml), filtered and stirred with ice cooling. Potassium carbonate (20·7 g, 150 mmol) was added followed by carbobenzoxy chloride (12·8 g, 75 mmol) and the mixture stirred for 0·5 h at 5–10° and 1 h at room temperature. The product was extracted with methylene chloride (2 × 150 ml), washed with water (3 × 200 ml) and evaporated to an oil (15·7 g). Chromatography on B.D.H. silica gel with chloroform gave crystalline fractions [6·2 g, 18% from (11)] which yielded, after three recrystallizations from benzene/cyclohexane, *methyl 3-(N-benzyloxycarbonylamino)cyclopent-1-ene-1-carboxylate* (14) (1·0 g), m.p. 84–85° (Found: C, 65·4; H, 6·3; N, 5·0. $C_{15}H_{17}NO_4$ requires C, 65·4; H, 6·2; N, 5·1%). ν_{max} 3400, 1720, 1680, 740, 720, 690 cm^{-1}. ^1H n.m.r. δ ($CDCl_3$) 7·4, s, 5H, ArH; 6·65, m, =CH; 5·2, s, $ArCH_2$; 5·2–4·7, m, 2H, NH and CHN; 3·8, s, OCH_3; 2·8–2·1, m, 3H, 3 × ring CH; 2·0–1·4, 1H, ring CH.

3-Aminocyclopent-1-ene-1-carboxylic Acid (6)

The *N*-protected amino ester (14) (500 mg) was refluxed under nitrogen in 6 M hydrochloric acid (30 ml) for 1 h. After removal of the solvent under vacuum, the amino acid was absorbed on a column of Dowex 50(H^+) ion-exchange resin (12 ml), washed with water, and eluted with 1 M pyridine. Evaporation of the solvent produced a crystalline powder (210 mg, 91%) which was recrystallized from aqueous ethanol to give *3-aminocyclopent-1-ene-1-carboxylic acid* (6) (110 mg), m.p. >280° (dec.) (Found: C, 56·4; H, 7·1; N, 10·8. $C_6H_9NO_2$ requires C, 56·7; H, 7·2; N, 11·0%). ν_{max} 2170, 1640, 1550s, 1320, 760 cm^{-1}. ^1H n.m.r. δ (D_2O, external $SiMe_4$) 6·7, apparent q, J 2 Hz, =CH; 4·9, m, $CHNH_2$; 3·4–2·7, m, 3H; 2·6–2·2, m, 1H.

Reduction of (14) to (2) and (3)

The *N*-protected amino ester (14) (500 mg) was reduced in ethanol (40 ml) over a 10% Pd/C catalyst (100 mg) in a Parr hydrogenator at 4 atm pressure for 3 h. Filtration and evaporation of the product yielded the crude saturated amino ester as an oil (260 mg); ν_{max} 3400 (br), 1725 cm^{-1}.

[13] Wheeler, O. H., and Lerner, I., *J. Am. Chem. Soc.*, 1956, **78**, 63.

Hydrolysis by refluxing in 6 M hydrochloric acid (20 ml) for 3 h followed by isolation of the free amino acid product with ion-exchange resin as above gave a slowly crystallizing gum (220 mg, 94%). T.l.c. analysis indicated an approximately 1:1 mixture of *cis*- and *trans*-3-aminocyclopentane-1-carboxylic acids (2) and (3). Two recrystallizations from ethanol/ethyl acetate (125 mg then 36 mg) gave almost pure *trans* isomer (3), m.p. 234–238° (dec.) (lit.[12] 235–236°), with t.l.c. and a ^{1}H n.m.r. spectrum identical to that of an authentic sample. Furthermore, the mass spectrum (70 eV) of this product was identical to that for both (2) and (3). *m/e* 129 (M^+, 7%), 100 (15), 84 (35), 83 (20), 67 (10), 57 (40), 56 (100), 44 (14).

Acknowledgments

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