Chemistry of Glutamic Acid Analogues. Structure of an Isoxazolo[5,4-*f*]indolizine Derivative

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The chemistry and X-ray analysis of (*RS*)-ethyl 4,6,7,9-tetrahydro-3-methoxy-5,7-dioxoisoxazolo[5,4-*f*]indolizine-4a(5*H*)-carboxylate, a derivative of a new ring system, is reported.

As part of our studies on the "receptor-active conformation(s)" of the central excitatory neurotransmitter glutamic acid,[1,2] the 3-isoxazolol amino acid α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) was synthesized.[3] AMPA was shown to be a specific and very potent agonist acting at a population of receptors, which probably represent the postsynaptic glutamic acid receptors.[1] Subsequently, 4,5,6,7-tetrahydro-3-hydroxy-isoxazolo[5,4-*c*]pyridine-5-carboxylic acid (5-HPCA) was synthesized and shown to interact very effectively with the glutamic acid receptors with a specificity similar to that of AMPA.[4]

Thus, in light of the conformational restrictions imposed on 5-HPCA this compound is assumed to essentially reflect "receptor-active conformations" of AMPA and glutamic acid.

5-HPCA was synthesized by stepwise deprotection of *2*, which was synthesized by *intra*-molecular *N*-alkylation of *1*[4] (Scheme 1). At reaction temperatures between −10 and 60 °C the reaction product contained, in addition to *2*, an acidic and strongly UV-absorbing compound. As no definite conclusion on the structure of this compound, which is the major product at reaction temperatures above *ca.* 30 °C, could be reached by spectroscopic methods, an X-ray crystal structure analysis was undertaken. The structure of the compound was shown to be (*RS*)-ethyl 4,6,7,9-tetrahydro-3-methoxy-5,7-dioxoisoxazolo[5,4-*f*]indolizine-4a(5*H*)-carboxylate (*3*). Compound *3* is evidently formed by a Claisen type reaction between the acetyl group and one of the ethoxycarbonyl groups of *2*.

Glutamic acid   AMPA   5-HPCA

Scheme 1.

(a)   (b)
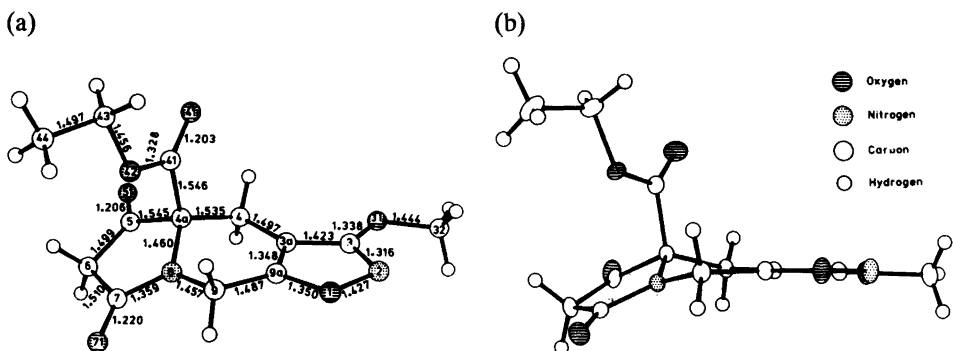

● Oxygen
◉ Nitrogen
○ Carbon
○ Hydrogen
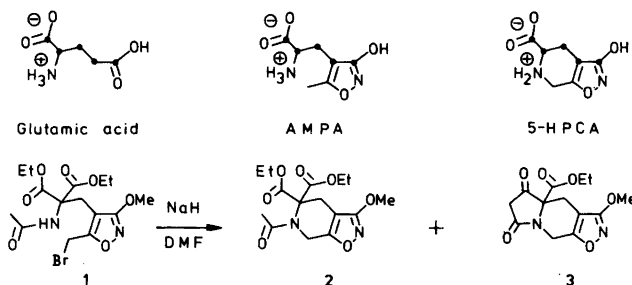
Fig. 1. ORTEP drawings[5] of (*3*). (a). The numbering of the atoms and bond lengths are displayed. E.s.d.'s are 0.001−0.002 Å. (b) 50 % probability ellipsoids for the non-hydrogen atoms are displayed; hydrogen atoms are represented as spheres of arbitrary radius.

Table 1. Valency and torsion angles (°). Estimated standard deviations are given in parentheses.

Valency angles (°)	C9a−O1−N2	107.8(1)	C5−C6−C7	104.8(1)
	O1−N2−C3	104.3(1)	C6−C7−N8	108.4(1)
	N2−C3−C3a	113.4(1)	C6−C7−O71	127.2(1)
	N2−C3−O31	122.7(1)	N8−C7−O71	124.4(1)
	C3a−C3−O31	123.9(1)	C7−N8−C4a	115.3(1)
	C3−O31−C32	114.9(1)	C7−N8−C9	122.7(1)
	C9a−C3a−C3	102.7(1)	C9−N8−C4a	120.7(1)
	C9a−C3a−C4	124.4(1)	N8−C9−C9a	105.5(1)
	C3−C3a−C4	132.9(1)	C3a−C9a−C9	127.4(1)
	C3a−C4−C4a	107.6(1)	O1−C9a−C9	120.7(1)
	C4−C4a−N8	113.4(1)	O1−C9a−C3a	111.9(1)
	C5−C4a−N8	101.3(1)	C4a−C41−O41	124.4(1)
	C4−C4a−C5	113.0(1)	C4a−C41−O42	109.2(1)
	C4−C4a−C41	112.0(1)	O41−C41−O42	126.2(1)
	C5−C4a−C41	105.7(1)	C41−O42−C43	117.7(1)
	N8−C4a−C41	110.7(1)	O42−C43−C44	106.1(1)
	C4a−C5−C6	108.4(1)	⟨C(N,O)−C−H⟩	110(1)
	C4a−C5−O51	123.3(1)	⟨H−C−H⟩	110(2)
	C6−C5−O51	128.3(1)		
Torsion angles (°)	C9a−O1−N2−C3	−0.2(2)	C4a−C5−C6−C7	−8.9(2)
	O1−N2−C3−C3a	1.0(2)	C5−C6−C7−N8	0.7(2)
	N2−C3−C3a−C8a	−1.5(2)	C6−C7−N8−C4a	8.5(2)
	C3−C3a−C9a−O1	1.3(2)	C7−N8−C4a−C5	−13.5(1)
	C3a−C9a−O1−N2	−0.8(2)	O51−C5−C6−C7	173.1(2)
	N8−C4a−C4−C3a	−38.2(2)	O71−C7−C6−C5	−178.4(1)
	C4a−C4−C3a−C9a	11.9(2)	O41−C41−C4a−C4	29.8(2)
	C4−C3a−C9a−C9	2.9(2)	O42−C41−C4a−N8	−27.2(1)
	C3a−C9−C9a−N8	8.7(2)	C4a−C41−O42−C43	−170.5(1)
	C9a−C9−N8−C4a	−39.2(2)	C41−O42−C43−C44	161.9(1)
	C9−N8−C4a−C4	58.0(2)	C32−O31−C3−N2	1.0(2)
	N8−C4a−C5−C6	13.0(1)		

Structural data and the overall shape of the molecule of *3* in the solid state are displayed in Table 1 and Fig. 1. The isoxazole ring is planar within the limits of experimental error. The six-membered ring adopts a half-chair conformation. The C4a and N8 atoms are at distances of 0.35 and −0.26 Å, respectively, from the best plane formed by the four remaining atoms. The five-membered ring of the tetrahydroindolizine moiety adopts a flattened envelope conformation. The C4a atom is on the flap of the envelope and deviates 0.21 Å from the best plane formed by the four remaining atoms of the ring. The nitrogen atom of the tetrahydroindolizine moiety is pyramidal, and, thus, the ring fusion is towards *trans*. The N8 atom is −0.10 Å out of the plane of its ligands, and the sum of the bond angles around it (358.7) differs from 360°. The ester group is in an axial position.

Inter-molecular distances between the non-hydrogen atoms less than 3.2 Å from $O71(x,\frac{1}{2}-y,\frac{1}{2}+z)$ to C5, C6, C7, or N8 are 2.990(2), 3.059(2), 3.151(2), 3.150(2) Å, respectively, and from $O42(2-x,\bar{y}, 1-z)$ to O51 3.187(2) Å.

EXPERIMENTAL

Melting points, determined in capillary tubes, are corrected. Elemental analyses were made by Mr. G. Cornali, Microanalytical Laboratory, Leo Pharmaceutical Products, Ballerup, Denmark. Perkin-Elmer grating IR and UV spectrophotometers, Model 247 and 402, respectively, a JEOL FX 900 90 MHz ^{1}H NMR spectrometer, and a Finnigan 3100 D mass spectrometer were used.

(RS)-Ethyl 4,6,7,9-tetrahydro-3-methoxy-5,7-dioxoisoxazolo[5,4-f]indolizine-4a(5H)-carboxylate (3) and ethyl 3-methoxy-5-ethoxycarbonyl-6-acetyl-4,5,6,7-tetrahydroisoxazolo[5,4-c]pyridine-5-carboxylate (2). To a stirred suspension of sodium hydride (36 mg; 1.5 mmol) in dry *N,N*-dimethylformamide (2ml) was added *1* [4] (316 mg; 0.75 mmol) during a period of 1 min at 60 °C. Stirring was continued at 60 °C for 75 min. The solution was evaporated at 15 mmHg, and upon addition of water (5 ml) and glacial acetic acid (0.2 ml), the residue was extracted with chloroform (3×10 ml). The combined and dried (MgSO$_4$) organic phases were evaporated and the residue subjected to column chromatography [silica gel (Woelm 0.063−1.00 mm):15 g; eluent: toluene−ethyl acetate−glacial acetic acid (25:25:1)] to give *2* (7 mg; 3 %), the IR spectrum of which was identical with that of an authentic sample of *2*,[4] and *3* (108 mg; 49 %), m.p. 145.0−147.0 °C. Anal. C$_{13}$H$_{14}$N$_2$O$_6$: C, H, N. IR (KBr): 2980−2850 (m−s, several bands), 1775 (s), 1725 (s), 1690 (s), 1655 (s), 1520 (m) cm^{-1}. UV [methanol (log ε)]: 272 (3.43) and 212 (4.03) nm. ^{1}H NMR (CDCl$_3$; TMS was used as an internal standard): δ 5.18 (1 H, dd, *J* 17 Hz and 2.0 Hz), 4.23 (2 H, q, *J* 7 Hz), 4.16 (1 H, dm, *J* 17 Hz), 3.99 (3 H, s), 3.49 (1 H, dd, *J* 16 and 2.0 Hz), 3.32 (1 H, dd, *J* 20 and 1.0 Hz), 3.19 (1 H, dd, *J* 20 and 1.0 Hz), 2.58 (1 H, dt, *J* 16 and 2.0 Hz), 1.25 (3 H, t, *J* 7 Hz). MS [70 eV; *m/z* (% rel. int.)]: 294 (15, M), 266 (40, [M−CO or C$_2$H$_4$]), 221 (100, [M−CO$_2$C$_2$H$_5$]), 193 (35, [M−CO$_2$C$_2$H$_5$−CO]).

X-Ray crystallographic analysis of (RS)-ethyl 4,6,7,9-tetrahydro-3-methoxy-5,7-dioxoisoxazolo[5,4-f]indolizine-4a(5H)-carboxylate (3). Compound *3* crystallized (ethyl acetate−light petroleum) in the monoclinic space group $P2_1/c$ with a=13.059(5), b=14.014(4), c=7.225(1) Å, β=95.05(2)° and D_c=1.484 g cm^{-3} for Z=4, D_m (flotation)=1.38 g cm^{-3} (20 °C). A single crystal (0.1×0.2×0.6 mm) was used for determination of the unit cell parameters and for the collection of intensity data. The measurements were performed at *ca.* 110 K with an Enraf-Nonius CAD-4 diffractometer. The temperature was kept constant within 0.5 K during the experiment. Graphite monochromated Mo*K*α radiation (λ=0.71073 Å) was used. A total of 5784 independent reflections with θ<35° were measured with θ−2θ scans, of these 3252 had net intensities greater than 2σ(*I*) and were regarded as observed reflections and used in the refinement procedure. The estimated standard deviation σ(*I*) of an intensity was calculated from counting statistics. No absorption correction was made. (μ=1.11 cm^{-1}).

The structure was solved by direct methods.[6] After refinement[7] of all the non-hydrogen atoms, the positions of the hydrogen atoms were obtained from a difference map. In

Table 2. Fractional atomic coordinates and equivalent isotropic/isotropic temperature factors ($\times 10^2$ Å^2). Estimated standard deviations are given in parentheses.

Atom[a]	x	y	z	U_{eq}/U_{iso} [b]
O1	0.48560(8)	0.13255(8)	0.50786(13)	1.82
N2	0.44206(9)	0.12986(10)	0.31969(15)	1.80
C3	0.52152(10)	0.11755(10)	0.22208(17)	1.42
C3a	0.61661(10)	0.11004(10)	0.33346(16)	1.23
C4	0.72480(10)	0.09303(10)	0.28809(17)	1.33
C4a	0.78923(10)	0.06978(10)	0.47058(16)	1.18
C5	0.90379(10)	0.09669(11)	0.46680(18)	1.53
C6	0.93808(11)	0.14675(11)	0.64490(19)	1.74
C7	0.84058(11)	0.16325(10)	0.73813(18)	1.47
N8	0.76065(8)	0.12604(8)	0.62821(14)	1.18
C9	0.65601(10)	0.12348(11)	0.68344(17)	1.46
C9a	0.58820(10)	0.12129(10)	0.50704(17)	1.34
O31	0.51273(8)	0.11349(8)	0.03631(13)	1.76
C32	0.40947(12)	0.12522(13)	−0.04968(20)	2.04
C41	0.78805(10)	−0.03797(10)	0.51615(17)	1.40
O41	0.77880(10)	−0.10033(8)	0.40146(14)	2.34
O42	0.80637(8)	−0.04984(7)	0.69840(12)	1.46
C43	0.82455(12)	−0.14656(10)	0.76704(20)	1.90
C44	0.87576(14)	−0.13680(13)	0.95975(21)	2.48
O51	0.95407(8)	0.07650(9)	0.34017(14)	2.30
O71	0.83217(9)	0.20379(8)	0.88545(14)	2.19
H4A	0.753(1)	0.147(1)	0.231(2)	1.3(4)
H4B	0.729(1)	0.038(1)	0.203(2)	1.5(4)
H6A	0.972(2)	0.208(2)	0.619(3)	3.5(6)
H6B	0.981(2)	0.102(2)	0.724(3)	3.1(5)
H9A	0.646(1)	0.180(1)	0.758(2)	2.0(4)
H9B	0.645(1)	0.066(1)	0.758(3)	2.3(5)
H32A	0.418(1)	0.117(1)	−0.181(3)	2.0(5)
H32B	0.368(2)	0.080(1)	−0.002(3)	2.4(5)
H32C	0.385(1)	0.189(2)	−0.020(3)	2.5(5)
H43A	0.758(1)	−0.176(1)	0.758(2)	1.6(4)
H43B	0.868(1)	−0.178(1)	0.687(2)	1.6(4)
H44A	0.941(2)	−0.101(2)	0.955(3)	3.7(6)
H44B	0.831(2)	−0.108(1)	1.044(3)	2.3(5)
H44C	0.896(2)	−0.202(2)	1.008(3)	4.0(6)

[a] The hydrogen atoms have numbers corresponding to those of their parent carbon atoms. [b] For the non-hydrogen atoms $U_{eq}=\frac{1}{3}$ ($U_{11}+U_{22}+U_{33}+2\ U_{13} \cos \beta$).

subsequent full-matrix least-squares calculations, an overall scale factor, atomic coordinates for all atoms, anisotropic thermal parameters for the non-hydrogen atoms and isotropic thermal parameters for the hydrogen atoms, were refined. The quantity minimized was $w(|F_o|-k|F_c|)^2$ where $w=(\sigma^2(F_o)+0.0005(F_o)^2)^{-1}$, except for the strong reflection 040 where $w=0$. The refinement converged at $R=0.040$ and $R_w=0.047$ for the observed reflections.

Final fractional coordinates are given in Table 2. Thermal parameters and observed and calculated structure factors are available on request from the author (L.B.).

Calculations were carried out using the MULTAN program[6] and the X-Ray 76 programme system.[7]

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