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NOVEL OXATHIAZINONES AS GASTRIN LIGANDS : UNEXPECTED PRODUCTS FROM THE SCHOTTEN-BAUMANN REACTION OF ARYLSULPHONYL CHLORIDES WITH DERIVATIVES OF ASPARTIC ACID

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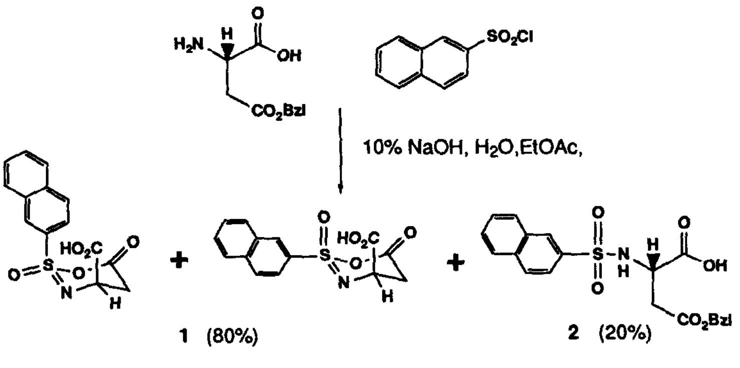
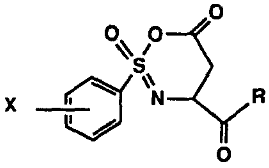
Abstract : The Schotten-Baumann reaction of arylsulphonyl chlorides with aspartic acid derivatives does not give the expected sulphonamide as the sole product. The unexpected products appear to represent the first examples of a previously unreported series of arylsulphoxo-2,3-dehydro-1,2,3-oxathiazin-6-ones and show activity at the gastrin receptor.

The Schotten-Baumann reaction between amines and sulphonyl chlorides is a routine method for the preparation of sulphonamides. Indeed we have employed this reaction many times for the preparation of arylsulphonamide derivatives of oligopeptides and amino acids. In general the reactions proceed smoothly and the predicted products are isolated in good yield. However, we were surprised to find that the reaction between the sodium salt of the 4-benzyloxyaspartic acid with 2-naphthalenesulphonyl chloride did not proceed in line with our expectations. 300 MHz ^1H NMR (d^3-MeOD) of the major product clearly showed that the benzyl group had disappeared. Furthermore, the signal due to the methylene protons (δ 2.8-3.1) was considerably more complex than had been anticipated and this led us to examine the isolated product in greater detail. A 2-D-J-resolved ^1H NMR experiment showed that the crude product was in fact a mixture of three compounds, of which only one was the desired sulphonamide **2** (Scheme 1). Interestingly, the other two appeared to be a mixture of diastereoisomers **1** [1]. Given that the starting amino acid contains one asymmetric centre, there are only two possible ways in which diastereoisomeric products could be obtained from this reaction. The first would be dimerisation of the starting material or product; however mass spectral and FTIR data failed to produce evidence of the presence of any such species. The second possibility is that the molecules contain an asymmetric sulphur atom.



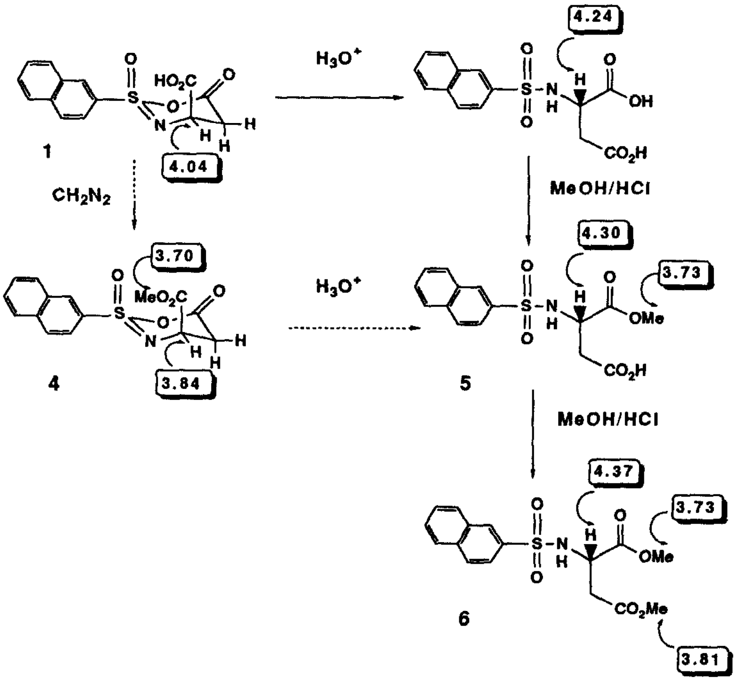
Scheme 1

Sulphonylation of analogous compounds is known to occur on oxygen and in most cases spontaneous rearrangement occurs at temperatures above 0°C so that the N-sulphonylated product is the only species isolated [2]. If we propose that sulphonylation occurs on the β-carboxyl group following ester hydrolysis, intramolecular attack of the amine and elimination of water would give a six-membered oxathiazinone system of the general structure **3**. This would imply that the products of the Schotten-Baumann reaction (Scheme 1) were the oxathiazinones **1**, a hitherto unreported ring system.



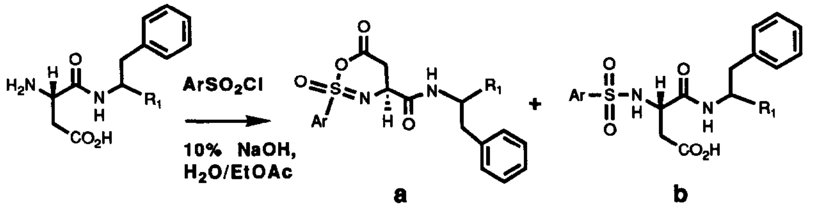
3

In order to investigate this proposal further, the reaction product was treated with methanolic HCl and the reaction monitored by ^{1}H NMR [3]. The movement of the signal due to the methine proton was used to diagnose the course of the reaction and the results obtained are consistent with initial hydrolysis of the six-membered ring followed by esterification of the diacid to give mixtures of the mono- and dimethyl esters, **5** & **6**. In a complementary experiment, the oxathiazinones **1** were treated with diazomethane. The product of this reaction was clearly a monomethyl ester **4** (δ 3.70 ppm $CO_2C\underline{H}_3$), indicating that the product only contained one free carboxylic acid. Subsequent treatment with methanolic HCl gave mixtures of mono- and dimethyl esters **5** & **6** identical to those obtained from the previous experiment. If we combine all these results we can propose a plausible mechanism for the initial hydrolysis and subsequent esterification of the compound which is consistent with our proposed structure for the oxathiazinone **1** (Scheme 2).



Scheme 2

Formation of the oxathiazinone does not appear to be limited to this substrate and we have observed the presence of similar products arising from the sulphonylation of other derivatives of aspartic acid (Table 1).



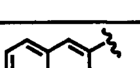
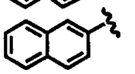
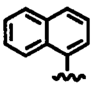
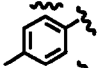
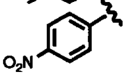
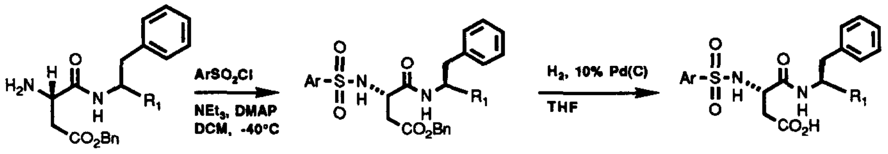
	Ar	R₁	Oxathiazinone a	:	Sulphonamide b
7.		H	99		1
8.		CONH₂	1		99
9.		CONH₂	54		46
10.		CONH₂	23		77
11.		CONH₂	88		12

Table 1

For example, reaction of L-aspartyl-L-phenylalaninamide with *p*-toluenesulphonyl chloride under these conditions gives rise to two distinct products (**10a** & **10b**). The major product **10b** precipitates on acidification of the reaction mixture with concentrated hydrochloric acid and a second distinct product **10a** is obtained from concentration of the aqueous phase. The ^{1}H NMR spectra (d^3-MeOD) of **10a** is similar, but not identical to that of the precipitate **10b**. Hence, whilst the two spectra are clearly non-identical they both appear to be consistent with that of the expected sulphonamide when examined in isolation [4]. This behaviour was mirrored by the *p*-nitrobenzenesulphonyl analogues and in each case it appeared that material isolated by precipitation from the reaction mixture **11b** belonged to a different class to that obtained from concentration of the aqueous phase **11a**. Authentic samples of the relevant sulphonamides were obtained using the route outlined in Scheme 3 and products obtained were identical to the precipitated products .



R_1 = H, $CONH_2$

Scheme 3

A number of other possible reactions that are open to these aspartic acid derivatives, eg racemisation of one or other of the chiral centres, simple salt formation, β-lactam or succinimide formation were similarly ruled out by independent synthesis.

The products of the Schotten-Baumann reaction can clearly be divided into two classes on the basis of their ^1H NMR spectra by examining the positions of the peaks assigned to the methine protons. In each case the chemical shift of the ring junction methine proton occurs ca. 0.25 ppm downfield of the α-methine proton in the analogous sulphonamide. A consistent pattern of changes was also observed in the positions of the methylene protons within the series; however these did not prove as good a structural diagnostic. The infrared spectrum of the original diastereoisomeric mixture of compounds **1** shows strong carbonyl stretches at 1729 and 1713 cm^{-1} which can be attributed to the carboxylic acid and ring carbonyl groups respectively. However, despite careful examination of the spectra of the dipeptide derivatives, there does not appear to be any structural correlation with the vibrational frequencies of either the C=O or S=O stretches and the structure of the products cannot be assigned on this basis. MNDO calculations showed that the carbonyl stretching bands were expected to be closer together in the oxathiazinone series when compared with the analogous sulphonamides. However, this method can only be used on a qualitative basis and the absolute calculated frequencies were not in good agreement with experiment, as expected for this method [5].

A variety of these compounds was examined in the isolated mouse stomach assay [7] and the oxathiazinones were shown to behave as either full or partial agonists at the gastrin receptor (Table 2). Gastrin is one of the three primary stimulants of gastric acid secretion [8] and thus these structures may well be useful intermediates in the preparation of gastrin agonists and antagonists. By contrast the analogous sulphonamides are generally inactive at concentrations below $3x10^{-5}M$ and it is interesting to note that the differences in physical properties that are reflected as small differences in the spectra of the two classes of compound are clearly evident in this biological system (Table 2).

Compound	log A_{50}	% Pentagastrin control
8a	-8.1	67
8b	-5.6	96
10a	-5.3	56
10b	inactive < $3x10^{-5}M$	-----

Table 2

The biological properties of the 2-naphthyloxathiazinone (**8a**) prompted us to examine the course of the Schotten-Baumann reaction more closely. Hence, we examined the reaction between H-Asp-Phe-NH_2 and 2-naphthalenesulphonyl chloride by analytical HPLC [C8 column (35% $MeCN/H_2O$ + 0.1% AcOH) at 1ml min^{-1}]. This study showed that the two products were apparently formed in equal proportions over the first hour of the reaction. However, the sulphonamide was clearly the dominant product after 4h and was the only product isolated after 24h at ambient temperature. Preliminary investigations did not provide any evidence of interconversion between the two products and suggest either that formation of the sulphonamide is autocatalytic or that of the oxathiazinone is suppressed. Modifying the reaction conditions by changing the nature of the organic solvent did little to alter the ratio of oxathiazinone to sulphonamide. Interestingly the products of the reactions with dipeptide derivatives have been consistently isolated as single diastereoisomers in all cases to date and this presumably reflects remote chiral induction from the phenylalanine residue. During these studies we observed that sulphonylation of the (R,R)-enantiomer of H-Asp-Phe-NH_2 gave the same product distribution as that obtained with the (S,S)-compound. However, sulphonylation of the (R,S) and (S,R) isomers was much slower and only minimal amounts of either product were isolated. This is consistent with the involvement of phenylalanine in controlling the stereochemical outcome at sulphur.

In conclusion, we have clearly shown that sulphonylation of aspartic acid derivatives under standard Schotten-Baumann conditions can produce a series of compounds some of which are not those expected from literature precedent and we have proposed that the unexpected products belong to a novel series of oxathiazinones. In some cases mixtures of compounds are formed, but in others the oxathiazinone predominates to the exclusion of the expected sulphonamide products. A comprehensive substructure search of the Chemical Abstracts database has not produced any references to the 2,3-dehydro-1,2,3-oxathiazin-6-one ring system and hence we can conclude that this is the first report of this novel ring system. Six-membered rings containing three or more heteroatoms do not occur widely in Nature. However, those that have been synthesised have been widely screened for biological activity [6] and indeed representative examples of our novel heterocyclic products have been shown to be biologically active at the gastrin receptor. The proposed oxathiazinone structure is consistent with all available physical data and studies are currently underway to crystalise the amorphous solids in order to carry out an X-ray analysis. Whilst this ring system has not been reported previously it is almost certain that these types of compounds have been isolated before and that their structural identity may well have been misassigned due to the spectral similarities with the anticipated sulphonamide products.

Acknowledgements

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References and Notes

[1] 300 MHz ^1H NMR (d^3-MeOD) data obtained for the two isomers of oxathiazinone **1** was as follows: *Isomer a* : δ 2.85 (1H, dd, J 18.5 & 7.0 Hz), 2.94 (1H, dd, J 18.5 & 4.5 Hz), 4.02 (1H, dd, J 8.0 & 4.5 Hz), 7.35 (1H, m), 7.53 (2H, m), 7.89 (3H, m), 8.32 (1H, s).
Isomer b: δ 2.86 (1H, dd, J 18.5 & 7.0 Hz), 2.99 (1H, dd, J 18.5 & 4.5 Hz), 4.04 (1H, dd, J 8.0 & 4.5 Hz), 7.35 (1H, m), 7.53 (2H, m), 7.89 (3H, m), 8.32 (1H, s).

[2] Gorden, I.M.; Maskill,H; and Ruasse,M.; *Chem.Soc.Rev.*, **1989**, *18* (2), 123.

[3] To this end a sample of the product was dissolved in methanol saturated with HCl and the reaction stirred at ambient temperature. Aliquots of the reaction mixture were periodically removed and concentrated *in vacuo* in order to examine the progress of the reaction by ^1H NMR. This process was repeated over a seven day period, after which broadening of the peaks in the spectrum made further observation impractical.

[4] For example L-aspartyl-L-phenylalaninamide (1.0 mmol) was suspended in water (2-3ml) and treated dropwise with a 10% aqueous solution of NaOH until the reaction mixture became homogeneous. The *p*-toluenesulphonyl chloride (1.0mmol) was then added as a solution in ethyl acetate and the biphasic reaction mixture stirred at ambient temperature for 10-18h . The two phases were then

separated and the sulphonamide **10b** precipitated by dropwise addition of conc. hydrochloric acid to the aqueous phase. Concentration of the filtrate gave the oxathiazinone **10a**.

p-Toluenesulphoxo-4-carbonylphenylalaninamide-2,3-dehydro-1,2,3-oxathiazin-6-one **10a**

300 MHz ^{1}H NMR (d^3-MeOD) δ 2.35 (3H, s), 2.66 (1H, dd, J 9&17Hz), 2.88 (2H, m), 3.16 (1H, dd, J 5 &14Hz), 4.04 (1H, dd, J 5 & 9Hz), 4.60 (1H, m), 7.25 (7H, m), 7.67 (2H, d); ν_{max} (KBr disk) 1718, 1673 and 1650 cm^{-1}

p-Toluenesulphonamido-L-aspartyl-L-phenylalninamide **10b** 300 MHz ^{1}H NMR (d^3-MeOD) 2.31 (1H, dd, J 7&17Hz), 2.45 (1H, dd, J 6&17Hz), 2.38 (3H, s), 2.85 (1H, dd, J 8&14Hz), 3.10 (1H, dd, J 5 &14Hz), 4.00 (1H, d, J 7Hz), 4.43 (1H, dd, J 7.2&15Hz), 7.25 (7H, m), 7.62 (2H, d), 8.13 (1H, bs).; ν_{max} (KBr disk) 1737, 1691 & 1645 cm^{-1}

[5] QCPE program no.455 Stewart, J.J.P; *J.Comput.-Aided.Mol.Des.;* **1990**, *4*, 1-105; Dieter,K.M.; Stewart, J.J.P.; Theochem., **1988**, *40*, 143.

[6] Landquist, J.K. in *Comprehensive Heterercyclic Chemistry* ; Katritzky, A.R.; and Rees, C.W., Series Ed.; Pergamon Press : Oxford, 1984; Vol. 1, pp 1084-86.

[7] For details of the bioassay in which the compounds were tested see Black, J.W. & Shankley, N.P.; *Br. J. Pharmacol.,* **1985**, *86*, 571.

[8] For example see Kovacs, T.O.G.; Walsh, J.H; Maxwell, V; Wong, H.C.; Azuma, T; and Katt, E; *Gastroenterol.,* **1989**, *97*, 1406.