## 360. *Amino-acids and Peptides. Part XVII.*[1] *The Preparation of the Methyl and Benzyl Esters of Amino-acids by Means of Dialkyl Sulphites.*

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The toluene-*p*-sulphonates of the methyl and benzyl esters of some amino-acids have been prepared by the action of dialkyl sulphites on the acid in the presence of toluene-*p*-sulphonic acid.

We have obtained high yields of the toluene-*p*-sulphonates of the methyl and the benzyl esters of certain amino-acids by the action of the dialkyl sulphite on the amino-acid in the presence of just over one equivalent of anhydrous toluene-*p*-sulphonic acid.[2] Voss and Blanke [3] prepared alkyl esters of simple carboxylic acids by the use of alkyl sulphites, but Voss and Wulkan [4] found that the methanesulphonates of betaine esters were formed when glycine, alanine, and tyrosine were heated with dimethyl sulphite for some hours at 130° (without added acid). We have not observed any *N*-alkylation under our conditions. Iselin, Rittel, Sieber, and Schwyzer [5] obtained high yields of the *p*-nitrophenyl esters of acylamido-acids by the use of di-*p*-nitrophenyl sulphite in the presence of pyridine, but this reaction does not proceed with alkyl sulphites.

The success of the reaction depends largely on the nature of the amino-acid. In those cases reported in the Tables high yields were obtained of products which were shown by paper chromatography to contain at most traces of amino-acid toluene-*p*-sulphonate (a common impurity in the product from other methods), an exception being the methylation of isoleucine, from which the crude product required purification by liberation of the free amino-ester and reprecipitation by toluene-*p*-sulphonic acid. Optical activity is retained; the leucine and phenylalanine methyl ester toluene-*p*-sulphonates were converted into optically active hydrochlorides, the proline methyl ester was converted into fully active *p*-nitrobenzoyl-L-proline methyl ester, and the optical rotations of the benzyl ester toluene-*p*-sulphonates agree reasonably well with those reported in the literature. The methylation of alanine, valine, and lysine could not be completed, nor could the benzylation of alanine,

[1] Part XVI, Williams and Young, *J.*, 1963, 881.
[2] Preliminary communication, Theobald, Williams, and Young, Proc. 3rd European Peptide Symposium, Basel, 1960, *Chimia* (*Switz.*), 1960, **14**, 371.
[3] Voss and Blanke, *Annalen*, 1931, **485**, 258.
[4] Voss and Wulkan, *Ber.*, 1937, **70**, 388.
[5] Iselin, Rittel, Sieber, and Schwyzer, *Helv. Chim. Acta*, 1957, **40**, 373.

TABLE 1.

Toluene-*p*-sulphonates of amino-acid methyl esters.

| No. | Amino-acid | Dimethyl sulphite (moles) | Time (hr.) | Yield (%) | Solvent for recrystallisation | M. p. |
|---|---|---|---|---|---|---|
| 1. | Glycine | 5 | 6·5 | 97 | Ethanol–ether | 116—117° |
| 2. | L-Leucine [d] | 5 | 3·5 | 97 | Acetone | 175·5—176 |
| 3. | DL-Isoleucine [e] | 10 | 6·5 | 94 | Chloroform–ether | 163—164 |
| 4. | L-Phenylalanine | 5 | 4·5 | 99 | Ethyl acetate | 162—162·5 |
| 5. | L-Proline | 5 | 6·5 | 91 | Methanol–ether | 116·6—117·5 |
| 6. | L-Aspartic acid | 10 | 5 | 98 | Ethyl acetate | 95—96 |
| 7. | L-Glutamic acid | 15 | 6·5 | 98 | Ethyl acetate | 130—130·5 |

| | | | Found (%) | | | | Required (%) | | |
|---|---|---|---|---|---|---|---|---|---|
| No. | $[\alpha]_D$ [b] | $R_F$ [c] | C | H | N | Formula | C | H | N |
| 1. | — | 0·52 | 45·6 | 5·7 | 5·4 | $C_{10}H_{15}NO_5S$ | 45·6 | 6·1 | 5·3 |
| 2. | +11·6° (*c* 6·9) | 0·83 | 53·0 | 7·1 | 4·4 | $C_{14}H_{23}NO_5S$ | 52·6 | 7·5 | 4·4 |
| 3. | — | 0·82 | 52·6 | 7·7 | 4·1 | $C_{14}H_{23}NO_5S$ | 52·6 | 7·5 | 4·4 |
| 4. | +13·1° (*c* 7·5) | 0·80 | 57·8 | 5·9 | 4·2 | $C_{16}H_{19}NO_5S$ | 58·1 | 5·9 | 4·0 |
| 5. | −17·8° (*c* 6·7) | 0·65 | 51·6 | 6·2 | 4·9 | $C_{13}H_{19}NO_5S$ | 51·6 | 6·6 | 4·6 |
| 6. | +10·8° (*c* 8·8) | 0·70 | 46·7 | 5·6 | 4·2 | $C_{13}H_{19}NO_7S$ | 46·7 | 6·0 | 4·2 |
| 7. | +15·8° (*c* 8·0) | 0·73 | 48·4 | 6·0 | 3·8 | $C_{14}H_{21}NO_7S$ | 48·1 | 6·3 | 4·0 |

TABLE 2.

Toluene-*p*-sulphonates of amino-acid benzyl esters.

| No. | Amino-acid | Dibenzyl sulphite (moles) [a] | Time (hr.) | Yield (%) | Solvent for recrystallisation | M. p. Obs. | M. p. Lit. [g] |
|---|---|---|---|---|---|---|---|
| 1. | Glycine | 5 | — [h] | 86 | Methanol–ether | 133—133·5° | 132—134° |
| 2. | L-Valine | 10 | 14 | 90 | Methanol–ether | 156—160 | 158—160 |
| 3. | L-Leucine | 5 | 6 | 93 | Methanol–ether | 156—157 | 158·5—160 |
| 4. | L-Phenylalamine | 5 | 6 | 88 | Ethyl acetate | 169—170·5 | 170·5—171·5 |
| 5. | L-Aspartic acid | 10 | 6 | 95 | Water–methanol (9 : 1) | 158—158·5 | 158—160 |
| 6. | L-Glutamic acid | 10 | 8 | 92 | Ethyl acetate | 141—142 | 144—145 |

| | $[\alpha]_D$ | | | Found (%) | | | | Calc. (%) | | |
|---|---|---|---|---|---|---|---|---|---|---|
| No. | Obs. [b] | Lit. | $R_F$ [c] | C | H | N | Formula | C | H | N |
| 1. | — | — | 0·80 | 57·4 | 5·6 | 4·0 | $C_{16}H_{19}NO_5S$ | 57·0 | 5·7 | 4·2 |
| 2. | −3·7° (*c* 5·0 in MeOH) | −3·6° [i] +1·2° [g] (MeOH) | 0·85 | 60·3 | 6·5 | 3·5 | $C_{19}H_{25}NO_5S$ | 60·2 | 6·9 | 3·7 |
| 3. | +4·5° (*c* 1·9 in DMF) | — [i] | 0·86 | 60·8 | 6·8 | 3·7 | $C_{20}H_{27}NO_5S$ | 61·2 | 6·7 | 3·6 |
| 4. | −5·6° (*c* 3·7 in MeOH) | −7·2° (MeOH) [g] | 0·89 | 64·5 | 6·0 | 3·4 | $C_{23}H_{25}NO_5S$ | 64·7 | 5·9 | 3·3 |
| 5. | +7·3° (*c* 2·0 in $CHCl_3$) | +6° [j] ($CHCl_3$) (D, −7·4) [i] | 0·87 | 61·7 | 5·7 | 2·8 | $C_{25}H_{27}NO_7S$ | 61·8 | 5·6 | 2·9 |
| 6. | +6·7° (*c* 2·5 in MeOH) | +7·6 (MeOH) [g] | 0·86 | 62·1 | 5·9 | 2·5 | $C_{26}H_{29}NO_7S$ | 62·5 | 5·9 | 2·8 |

Footnotes to Tables 1 and 2.

DMF = Dimethylformamide.

[a] For each mole of amino-acid. [b] For methyl esters, in methanol; for benzyl esters, in the solvent stated; temperature, 19—23°. [c] In butan-1-ol–water–acetic acid (62 : 26 : 12). [d] The product gave hydrochloride of m. p. 151—151·5°, $[\alpha]_D^{23}$ +13·2° (*c* 6·1 in water), $[\alpha]_D^{25}$ +20·0° (*c* 3·7 in MeOH). Schott, Larkin, Rockland, and Dunn (*J. Org. Chem.*, 1947, **12**, 490) give $[\alpha]_D^{25.9}$ −13·40° (*c* 5 in water); the sign is clearly erroneous. This error is reproduced in Table 10—28 of " Chemistry of the Amino Acids," Greenstein and Winitz, Wiley, New York, 1961, p. 930. Weil and Kuhn (*Helv. Chim. Acta*, 1946, **29**, 784) give $[\alpha]_D^{26}$ 20·85° (*c* 4·5 in MeOH). [e] The crude product contained isoleucine toluene-*p*-sulphonate, and was purified by liberation of the free amino-ester in ether and precipitation with toluene-*p*-sulphonic acid. [f] The product gave hydrochloride of m. p. 160—161°, $[\alpha]_D^{23}$ −4·1° (*c* 3·6 in water); Schwarz, Bumpus, and Page (*J. Amer. Chem. Soc.*, 1957, **79**, 5697) give $[\alpha]_D^{25}$ −4·6° (*c* 5 in water). [g] Zervas, Winitz, and Greenstein, *J. Org. Chem.*, 1957, **22**, 1515. [h] 35 min. at 125—130°, then 3 hr. at 100°. [i] Gibian and Schröder, *Annalen*, 1961, **642**, 145. These authors give $[\alpha]_D^{21}$ +4·4° for L-leucine benzyl ester benzenesulphonate (in DMF). [j] Velluz, Amiard, Bartos, Goffinet, and Heymès, *Bull. Soc. chim. France*, 1956, 1464.

lysine, and proline. In both cases tyrosine gave chromatographically impure products, presumably due to *O*-alkylation. The procedure appears to be capable of extension, since di-*p*-nitrobenzyl sulphite gave an excellent yield of L-leucine *p*-nitrobenzyl ester toluene-*p*-sulphonate.

In our experience the most satisfactory general method for the preparation of the hydrochlorides of the methyl esters of amino-acids remains that of Brenner and Huber,[6] using thionyl chloride with methanol; it is likely that this esterification is effected by methyl chlorosulphite or dimethyl sulphite. Toluene-*p*-sulphonates are, however, frequently more readily purified than are the corresponding hydrochlorides; thus, L-proline methyl ester hydrochloride is known only as an oil, whereas we describe here the crystalline (but hygroscopic) toluene-*p*-sulphonate. In such cases, the present procedure has an advantage.

For the preparation of amino-acid benzyl ester toluene-*p*-sulphonates, Zervas, Winitz, and Greenstein[7] recommend the use of benzene in the azeotropic procedure,[8] and report yields of 80—90% in most instances. For the amino-acids listed in Table 2, our method gives comparable yields and the choice must depend on convenience. It should be added that pure dimethyl and dibenzyl sulphites are stable for some months at 0—5°.

## Experimental

M. p.s were taken on a Kofler block. Optical rotations were measured on an ETL-NPL automatic polarimeter. Thionyl chloride was purified by distillation from quinoline and then from linseed oil, the fraction of b. p. 76—78° being used. Toluene-*p*-sulphonic acid hydrate was dehydrated at 100° at a water-pump for 4 hr., and the product was recrystallised from ethyl acetate. Dimethyl sulphite was prepared by the method of Voss and Blanke [3] and had b. p. 124—127°. $R_F$ values refer to the solvent butan-1-ol–water–acetic acid (62 : 26 : 12).

*Dibenzyl Sulphite.*—The method of Richter [9] was modified as follows. Purified thionyl chloride (54 ml.) was added dropwise during 2 hr. to a well-stirred solution of redistilled benzyl alcohol (153 ml.) and dry pyridine (121 ml.) in dry ether (1 l.), at 15—25°. Stirring was continued for another 1 hr., and the pyridine hydrochloride was filtered off and washed with ether; the combined filtrates were washed with dilute hydrochloric acid, 2N-sodium carbonate, and water, and dried ($MgSO_4$). The ether was removed and the residue distilled (with some decomposition) at 0·4 mm.; the fraction of b. p. 160—165°/0·4 mm., $n_D^{25}$ 1·5600, was used (yield 60—70%). Richter [9] records b. p. 193—199°/15 mm., with much decomposition.

*Di*-p-*nitrobenzyl Sulphite.*—This was prepared analogously to dibenzyl sulphite, except that the reaction was stopped 20 min. after the final addition of thionyl chloride, by adding water and ethyl acetate, which dissolved the oil that had separated. The organic layer was washed, dried, and evaporated to give a crystalline solid (79%). Recrystallisation from chloroform–light petroleum (b. p. 60—80°) gave the *ester* as pale yellow prisms, m. p. 82·5—83° (Found: C, 47·9; H, 3·5; N, 7·9. $C_{14}H_{12}N_2O_7S$ requires C, 47·7; H, 3·4; N, 7·95%).

*General Esterification Procedure.*—The amino-acid (0·01 mole), anhydrous toluene-*p*-sulphonic acid (0·011 mole), and the dialkyl sulphite (see Tables) were heated on a boiling-water bath for the stated time, during which dissolution occurred. The cooled solution was poured slowly into dry ether, and the amino-ester toluene-*p*-sulphonate crystallised. Paper chromatography normally showed it to contain little or no amino-acid toluene-*p*-sulphonate.

*Conversion of Toluene*-p-*sulphonates into Hydrochlorides.*—The amino-ester toluene-*p*-sulphonate was shaken with 2N-sodium carbonate and ether; the ether extract was dried ($MgSO_4$), and the hydrochloride was precipitated by the addition of hydrogen chloride in ether.

L-*Valine Methyl Ester Toluene*-p-*Sulphonate.*—This was separated from the crude product (containing much valine) by shaking it with 2N-sodium carbonate and ether; the ether extract was dried ($MgSO_4$), and a solution of anhydrous toluene-*p*-sulphonic acid in ether was added. The *toluene*-p-*sulphonate*, recrystallised from acetone, had m. p. 175—176°, $[\alpha]_D^{23}$ +13·4° (*c* 10 in MeOH) (Found: C, 51·6; H, 7·2; N, 4·7. $C_{13}H_{21}NO_5S$ requires C, 51·5; H, 6·9; N, 4·6%). The hydrochloride had m. p. 172—173°, $[\alpha]_D^{23}$ +15·6° (*c* 4·8 in $H_2O$) [lit.,[10] m. p. 167·5—168°, $[\alpha]_D^{21}$ +15·5° (*c* 2 in $H_2O$)].

p-*Nitrobenzoyl*-L-*proline Methyl Ester.*—L-Proline methyl ester toluene-*p*-sulphonate (1·00 g.) was added to ethyl acetate (15 ml.) containing *p*-nitrobenzoyl chloride (0·62 g.). The suspension

[6] Brenner and Huber, *Helv. Chim. Acta*, 1953, **36**, 1109.
[7] Zervas, Winitz, and Greenstein, *J. Org. Chem.*, 1957, **22**, 1515.
[8] Cipera and Nicholls, *Chem. and Ind.*, 1955, 16.
[9] Richter, *Ber.*, 1916, **49**, 2339.
[10] Smith, Spackman, and Polglase, *J. Biol. Chem.*, 1952, **199**, 801.

was stirred vigorously while a solution of sodium carbonate (1·8 g.) in water (10 ml.) was added. After 30 min., the organic layer was separated and dried ($MgSO_4$), and the solvent was removed, leaving a crystalline product (0·75 g., 82%). Recrystallisation from ethyl acetate–light petroleum gave the *ester*, m. p. 107—108°, $[\alpha]_D^{22}$ −83·0° (*c* 1·2 in EtOH) (Found: C, 56·0; H, 5·1; N, 9·7. $C_{13}H_{14}N_2O_5$ requires C, 56·1; N, 5·0; N, 10·1%). The ester prepared by the action of diazomethane on *p*-nitrobenzoyl-L-proline had m. p. 106·5—108°, $[\alpha]_D^{22}$ −81·2° (*c* 2·5 in EtOH).

L-*Leucine* p-*Nitrobenzyl Ester Toluene*-p-*sulphonate*.—L-Leucine (66 mg., 0·5 mmole), anhydrous toluene-*p*-sulphonic acid (103 mg., 0·6 mmole), and di-*p*-nitrobenzyl sulphite (700 mg., 2 mmoles) were heated at 100° for 7 hr. On cooling the mass solidified and was triturated with ether and then filtered off, giving a product (200 mg., 91%) shown by paper chromatography to be free from unchanged leucine. Recrystallisation from methanol–ether gave the *toluene-p-sulphonate*, m. p. 207—208°, $[\alpha]_D^{23}$ −0·7° (*c* 1 in MeOH) (Found: C, 55·0; H, 6·1; N, 6·1. $C_{20}H_{26}N_2O_7S$ requires C, 54·8; H, 5·9; N, 6·4%).

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