NEW TOTAL SYNTHESIS OF PATULIN

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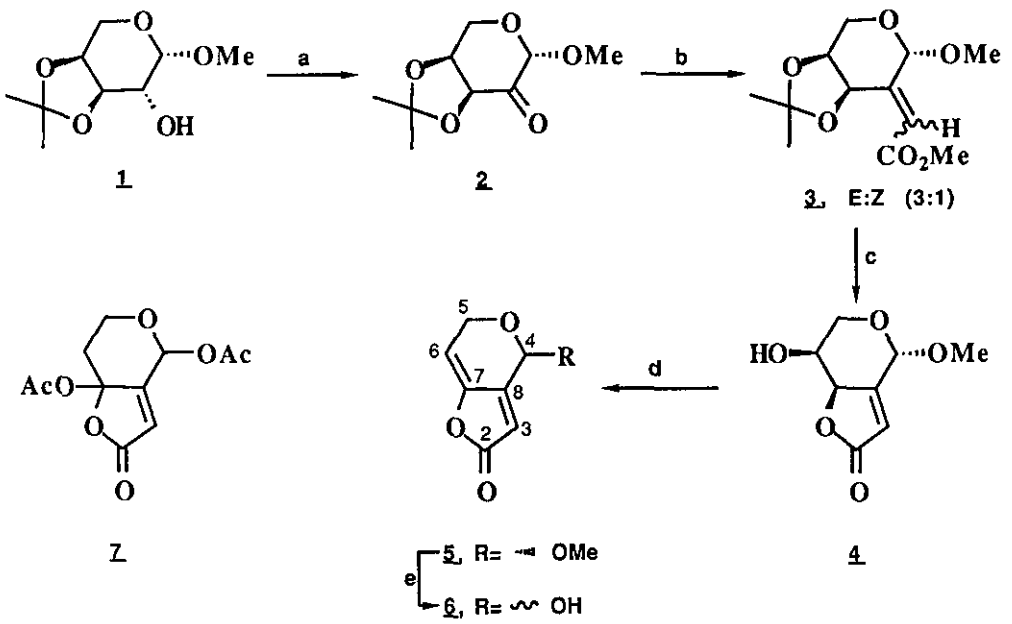
Abstract-The total synthesis of the mycotoxin patulin from L-arabinose is described.

Patulin (6), a potent antibiotic and mycotoxin produced by Penicillium and Aspergillus species, is synthesized biologically from acetylcoenzyme A[1]. Its α,β-unsaturated five-membered lactone structure was determined by total synthesis by Woodward and Singh[2], though they only obtained a very low yield. Since then no improvement in the yield nor new synthesis has been reported despite the large number of reports of its detection and isolation from different sources[1]. Here we report a new synthesis of patulin that affords a 23% overall yield from L-arabinose, an inexpensive, readily available aldopentose. Since arabinose has a cis-configuration at C-3 and C-4, the hemiacetal ring of 6 with its double bond, can be formed from its pyranoside form.



1 2 3, E:Z (3:1) 4 5, R= OMe 6, R= OH 7

a) PCC, molecular sieves, Cl_2CH_2; b) $Ph_3P{=}CHCO_2Me$, benzene, 50°C;
c) MeOH-HCl, reflux; d) MsCl, Py; e) TFA:H_2O (9:1), 50°C

Methyl-3,4-O-isopropylidene-α-L-arabinopyranoside (1) was prepared from L-arabinose as previously described[3] except for a modification of the isopropylidenation step that raised the yield to 70% from L-arabinose. Oxidation of 1 with pyridinium chlorochromate (PCC) gave an 87% yield of the ketone 2. Compound 2 was previously prepared by oxidation of methyl-3,4-O-isopropylidene-α-L-arabinopyranoside by different reagents[4,5]. Wittig olefination of 2 with (carbomethoxymethylene)triphenylphosphorane afforded 3 as a 3:1 mixture of E-Z stereoisomers that could not be separated by chromatography, and hydrolysis of the isopropylidene group of 3 yielded the lactone 4 (68%). When 4 was treated with methanesulphonyl chloride in pyridine at 0°C and left 1 day at room temperature, spontaneous anti-elimination occurred affording (S)-O-methylpatulin (5) in 85% yield, whereas in Woodward and Singh's route to patulin, conversion of the diacetate 7 to acetylpatulin occurs in only 1-2% yield. Hydrolysis of the methoxy group of 5 was carried out in 90% aqueous trifluoroacetic acid (TFA) at 50°C during 1 h to afford patulin (6) in 79% yield. Lower TFA/H_2O ratios meant longer reaction times and the formation of polymers.

EXPERIMENTAL

Melting points were determined using a Büchi apparatus and are uncorrected. Ir spectra were recorded with a Perkin-Elmer 710B spectrophotometer, uv spectra with a Kontron Uvicon 820 spectrophotometer, nmr spectra (in the solvent specified and with Me_4Si as internal standard) on a Bruker WP-250 operating at 250 MHz (for ^1H) or 62.85 MHz (for ^{13}C) and mass spectra on a Kratos MS-50 apparatus. Optical rotations were measured on a Perkin-Elmer 141 polarimeter (concentrations are given in g/100 ml). The adsorbent used for flash column chromatography was silica gel (Merck Kieselgel 60, 230-400 mesh) and the plates used for tlc were coated with silica gel (Merck Kieselgel 60 HF_{254}). Solutions in organic solvents were dried with anhydrous sodium sulphate; solvents were removed under reduced pressure. Dichloromethane and pyridine were distilled from P_2O_5 and KOH, respectively. Methyl α-L-arabinopyranoside was prepared from L-arabinose in 81% yield[3].

Methyl 3,4-O-isopropylidene-α-L-arabinopyranoside (1).- A mixture of anhydrous copper(II) sulphate (30 g) and methyl α-L-arabinopyranoside (11.5 g, 70.12 mmol) in acetone (AR grade, 500 ml) was stirred with (98%) sulphuric acid (0.5 ml) at room temperature for 1 day. The mixture was neutralized with concentrated aqueous ammonium hydroxide, filtered through a celite plug which was then washed with acetone, and the solvent was removed. The residue was dissolved in dichloromethane (300 ml) and dried , and the solvent was evaporated to give methyl 3.4-O-isopropylidene-α-L-arabinopyranoside (1) (12.3 g, 86%), as a syrup which could be used for the next stage without any further purification. $[\alpha]_D$ +200° (c 1.5, chloroform).

Methyl 3,4-O-isopropylidene-α-L-*erythro*-pentopyranosid-2-ulose (2).- A mixture of methyl 3,4-O-isopropylidene-α-L-arabinopyranoside (1) (12.3 g, 60.3 mmol), pyridinium chlorochromate (27.0 g, 125.3 mmol) and powdered 3Å molecular sieves (21.4 g) in dry dichloromethane (250 ml) was stirred at room temperature for 3 h. The mixture was diluted with ether and filtered through a silica gel plug (eluted with 1:1 ethyl acetate-hexane) to give methyl 3,4-O-isopropylidene-α-L-*erythro*-pentopyranosid-2-ulose (2) (10.6 g, 87%). mp 88-91°C (lit[5] 87-88°C); $[\alpha]_D$ +148° (c 1.33, chloroform); ir ν_{max} (KBr) 1760 cm^{-1}.

Wittig reaction of methyl-3,4-O-isopropylidene-α-L-*erythro*-pentopyranosid-2-ulose.- Methyl-3,4-O-isopropylidene-α-L-*erythro*-pentopyranosid-2-ulose (2) (10.60 g, 52.5 mmol) was dissolved in benzene (150 ml) and stirred at 50°C with (carbomethoxymethylene)triphenylphosphorane (19.9 g, 59.6 mmol) for 6 h. The solvent was removed and the residue was purified by flash chromatography (eluted with 3:7 ethyl acetate-hexane) to afford the 3:1 mixture of E:Z alkene isomers 3 (11.35 g, 84%), as a colourless oil. $[\alpha]_D$+129.7° (c 3.21, $CHCl_3$); ir ν_{max} (KBr, film) 1740, 1695 cm^{-1}; ^1H nmr ($CDCl_3$) ∂ 6.4-3.8 (5H, m), 3.75 (2.2H, s, CO_2CH_3), 3.72 (0.8H, s, CO_2CH_3), 3.65 (1H, m), 3.48 (3H, s, OCH_3), 1.54 (2.2H, s, CH_3), 1.51 (0.8H, s, CH_3), 1.40 (3H, s, CH_3); ms m/z (relative intensity) 243 (M^+-CH_3, 0.3), 227 (M^+-OMe, 0.7), 126 (100), 69 (58). Found C, 55.62; H, 7.25. $C_{12}H_{18}O_6$ requires C, 55.81; H, 7.02%.

Synthesis of the lactone 4.- The mixture of alkenes 3 (926 mg, 3.59 mmol) described above was dissolved in methanol-1.2 N hydrochloric acid (49 ml : 1 ml) and refluxed for 1.5 h. The solvent was removed and the residue was partitioned between saturated aqueous sodium bicarbonate solution (10 ml) and dichloromethane (4 x 60 ml). The organic layer was dried and evaporated to give the lactone 4 (453 mg, 68%) as a white solid. mp 129-131°C (CH_2Cl_2-Et_2O); $[\alpha]_D$ +241° (c 1.55, $CHCl_3$); ir ν_{max} (KBr) 3450,1740 cm^{-1}; ^1H nmr ($CDCl_3$) ∂ 6.01 (1H, d, $J_{3,7}$= 1.5 Hz, H-3), 5.50 (1H, s, H-4), 5.15 (1H, dd, $J_{7,6}$= 3.9 Hz, $J_{7,3}$= 1.8 Hz, H-7), 4.38 (1H, m, H-6), 4.02 (1H, d, J_{gem}= 12.7 Hz, H-5), 3.86 (1H, dd, J_{gem}=12.7 Hz, $J_{5',6}$= 2.3 Hz, H-5'), 3.50 (3H, s, OCH_3); ^{13}C nmr ($CDCl_3$) ∂ 172.5 (C-2), 159.6 (C-8), 115.2 (C-3), 95.4 (C-4), 78.2 (C-7), 69.4 (C-6), 61.2 (C-5), 55.2 (OCH_3); ms m/z (relative intensity) 187 (M^++1, 0.4), 168 (2), 155 (14), 126 (100), 111 (20), 99 (31), 69 (70). Found C, 51.48; H, 5.36. $C_8H_{10}O_5$ requires C, 51.61; H, 5.41%.

Synthesis of (S)-O-methylpatulin (5).- Compound 4 (600 mg, 3.23 mmol) was treated with methanesulphonyl chloride (0.37 ml, 4.57 mmol) in dry pyridine (10 ml) at 0°C and left 1 day at room temperature. The solution was concentrated, diluted with chloroform (100 ml) and washed successively with

aqueous hydrochloric acid (3 M, 3 ml) and brine (2 ml). The chloroform layer was then dried and evaporated to give crude (S)-O-methylpatulin (5), which was purified by flash chromatography (2:8 ethyl acetate-hexane) to give pure 5 (458 mg, 85%) as a white solid. mp 70-73°C (lit[6] 69-71°C, racemic 5). $[\alpha]_D$ +174.7 ° (c 1.04, $CHCl_3$); uv λ_{max} (EtOH) 276 nm; ir ν_{max} (KBr) 1770, 1750 cm^{-1}; ^1H nmr ($CDCl_3$) ∂ 5.90 (1H, br s, H-3), 5.86 (1H, m, H-6), 5.55 (1H, s, H-4), 4.55 (1H, br d, J_{gem}= 17.2 Hz, H-5), 4.30 (1H, dd, J_{gem}= 17.2 Hz, $J_{5',6}$= 4.4 Hz, H-5'), 3.49 (3H, s, OCH_3); ^{13}C nmr ($CDCl_3$) ∂ 168.5 (C-2), 148.7 (C-7), 146.0 (C-8), 110.1 (C-3), 107.4 (C-6), 94.3 (C-4), 58.7 (C-5), 55.9 (OCH_3); ms m/z (rel. int.) 168 (M^+,2), 137 (2), 126 (2), 112 (2), 97 (2), 55 (9), 44(100).

Patulin (6).- O-Methylpatulin (5) (201 mg, 1.19 mmol) was dissolved in 90% aqueous trifluoroacetic acid (30 ml) and heated at 50 °C for 1 h. The solvent was removed (without heating) and the residue was partitioned between saturated aqueous sodium bicarbonate solution (5 ml) and ethyl acetate (5 x 20 ml). The organic layer was dried, evaporated and purified by PTLC (1:1 ethyl acetate-hexane) to give patulin (6) (145 mg, 79%), as a white solid. mp 106-7°C (lit.[6] 110-11°C), $[\alpha]_D$ 0° (c 0.5, EtOAc); uv λ_{max} (EtOH) 276 nm; ir ν_{max} (KBr) 1765, 1750 cm^{-1}; ^1H nmr ($CDCl_3$ + CD_3OD) ∂ 6.02 (2H, br s, H-4, H-3), 5.97 (1H, m, H-6), 4.70 (1H, dd, J_{gem}= 17.4 Hz, $J_{5,6}$= 3.0 Hz, H-5), 4.41 (1H, dd, J_{gem}= 17.4 Hz, $J_{5',6}$= 4.0 Hz, H-5'); ^{13}C nmr ($(CD_3)_2CO$) ∂ 169.5 (C-2), 153.0 (C-7), 147.3 (C-8), 110.7 (C-3), 109.0 (C-6), 89.4 (C-4), 59.7 (C-5); MS m/z (rel. int.) 154 (M^+, 14), 136 (36), 126 (36), 110 (100), 97 (21), 82 (44), 55 (69).

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REFERENCES

1.- a) R. J. Cole and R. H. Cox, "Handbook of Toxic Fungal Metabolites", Academic Press Inc., New York (1981), p. 510-516; b) G. Engel and M. Teuber, "Mycotoxins. Production, Isolation, Separation and Purification", Ed. by V. Betina, Vol. 8, Elsevier (1984), p. 291-298; and references cited therein.

2.- R. B. Woodward and G. Singh, J. Am. Chem. Soc., 1950, 72, 1428.

3.- a) J. W. Pratt, N. K. Richtmyer, and C. S. Hudson, J. Am. Chem. Soc., 1952, 74,2200; b) J. Honeyman, J. Chem. Soc., 990 (1946).

4.- A. Rosenthal and M. Sprinzl, Can. J. Chem.,1970, 48, 3253.

5.- S.-H. An and M. Bobek, Tetrahedron Lett.,1986, 27, 3219.

6.- F. Bergel, A. L. Morrison, A. R. Moss, and H. Rinderknecht, J. Chem. Soc., 415 (1944).

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