Synthesis of (2*R*,3*S*,4*R*)-3,4-Dihydroxyproline from D-Ribonolactone; an Approach to the Synthesis of Polyfunctionalised D-Amino Acids from Sugar Lactones. *X*-Ray Molecular Structures of 2-Azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone, 2-Azido-2-deoxy-D-ribono-1,4-lactone, and (2*R*,3*S*,3*R*)-3,4-Dihydroxyproline

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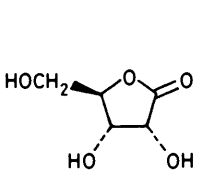
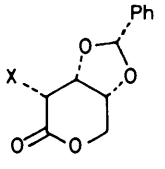
A general approach to the synthesis of polyfunctionalised amino acids from sugar lactones, in which nucleophilic displacement by azide ion of *O*-trifluoromethanesulphonyl esters adjacent to the lactone carbonyl is the key step, is discussed and is exemplified by the synthesis of the D-amino acid (2*R*,3*S*,4*R*)-3,4-dihydroxyproline from D-ribonolactone; unexpectedly, displacement of a triflate by azide in the C-2 position of ribonolactone occurs with retention of configuration. The *X*-ray crystal structures of 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone, 2-azido-2-deoxy-D-ribono-1,4-lactone, and (2*R*,3*S*,4*R*)-3,4-dihydroxyproline are reported. I.r. spectroscopy does not discriminate reliably between 1,4-and 1,5-lactones, and *X*-ray crystallography is necessary to provide reliable structural information in this area.

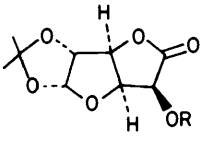
There are well over a thousand naturally occurring non-protein amino acids; more than a hundred of these contain at least two chiral centres—frequently with one or more other functional groups including alcohols, carboxylic acids, and amines—and may be exemplified by such structures as polyhydroxylated prolines and pipecolic acids, hydroxylated aminopimelic, glutamic, and aminoadipic acids.[1] Although these compounds frequently occur in nature in small amounts and are difficult to isolate, it is apparent that the biological roles they play are diverse. Since the amounts available from natural sources frequently cannot normally provide sufficient material for proper biological evaluation, there has recently been considerable effort into the synthesis of chiral polyfunctionalised amino acids.[2,3] Carbohydrates have been used as chiral starting materials for the synthesis of several amino acids,[4,5] with recent examples including a trihydroxypipecolic acid,[6] bulgecinine,[7] and the amino acid constituents of the polyoxins;[8] many are multi-stage syntheses involving considerable manipulation of protecting groups. In a general approach to amino acids from carbohydrates, two common practical problems are (i) the difficulty of introduction of a nitrogen function by nucleophilic substitution in sugars due to the presence of β-oxygen functions and (ii) the oxidation of the carbon destined to become the carboxy group. These disadvantages are avoided, or at least reduced, by the use of readily available sugar lactones, in which the carboxy group is already present (and also protects one of the other alcohol functions) and in which the position α to the carbonyl group should be relatively susceptible to nucleophilic substitution. Both D-ribonolactone (**1**) and D-glucuronolactone are cheap compounds which may efficiently be converted in a single step to derivatives such as (**2**) and (**7**)[9] respectively, in both of which only the hydroxy group adjacent to the lactone carbonyl function is unprotected, and so are suitable candidates for divergent syntheses of amino acids; the protection of functional groups in this approach to amino acids is thus minimal in comparison with the majority of syntheses of amino acids from carbohydrates. The potential of this approach has been demonstrated by the conversion of glucuronolactone into bulgecinine, two trihydroxypipecolic acids, and a dihydroxypipecolic acid.[10]



(**1**)



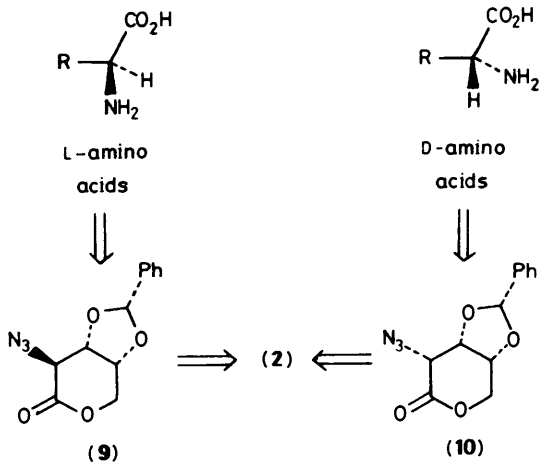
(**2**) X = OH
(**3**) X = $OS(O)_2CF_3$
(**4**) X = OC(S)SMe
(**5**) X = $OS(O)_2Me$
(**6**) X = H



(**7**) R = H
(**8**) R = $OS(O)_2CF_3$

D-Ribonolactone (**1**) has been used as a chiral starting material for the synthesis of natural products,[11,12] several of which utilise Zinner's lactone (**2**) in which only the 2-OH group is unprotected.[13] Replacement of this hydroxy group with azide with inversion of configuration would allow the preparation of an intermediate (**9**) suitable for elaboration to L-amino acids, whereas introduction of azide with retention to give intermediate (**10**) should allow the synthesis of D-amino acids (Scheme 1). This paper describes the use of D-ribonolactone in the synthesis of intermediates for the synthesis of D-amino acids, illustrated by the preparation of (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**); a preliminary account of this work has been published.[14] The enantiospecific synthesis of two other diastereoisomers of 3,4-dihydroxyproline, in which the two hydroxy groups are *trans* to each other, starting from allylglycine, has been reported recently.[15]

For the synthesis of the D-amino acid (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**), a suspension of D-ribonolactone (**1**) was treated with benzaldehyde in conc. hydrochloric acid to give Zinner's lactone (**2**),[16] which has been shown[17] to have the 1,5-lactone structure with only the C-2 OH group unprotected, in 89% yield on a 20 g scale.[18] The lactone (**2**) is insoluble in the majority of organic solvents and so could not be converted to the trifluoromethanesulphonyl derivative (**3**) by the normal procedure;[19] accordingly, the free hydroxy group was esterified by addition of trifluoromethanesulphonic anhydride to a



Scheme 1.

suspension of compound (**2**) in pyridine at −10 °C to form the corresponding triflate (**3**) as an easily handled compound which can be recrystallised from ethanol. The *O*-trifluoromethanesulphonate (**8**)[20] derived from D-glucuronolactone is similarly readily manipulated and also may be recrystallised from ethanol, and it may be that these kinds of triflates are highly convenient synthetic intermediates for nucleophilic displacements. Treatment of the *ribo*-triflate (**3**) with sodium azide in dimethylformamide (DMF) at room temperature formed the azido lactone, 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**), [64% yield from (**2**) on a 10 g scale], in which the stereochemistry at C-2 (established by *X*-ray crystallography) had been unexpectedly retained during the nucleophilic displacement; treatment of the triflate with tetrabutylammonium azide in ethyl acetate gave only the same azide (**10**) in lower yield, together with other products not containing the azide functionality. Since the coupling constants between the protons attached to C-2 and C-3 in the alcohol (**2**) ($J_{H2,H3}$ 3.1 Hz), the azide (**10**) ($J_{H2,H3}$ 3.3 Hz), the triflate (**3**) ($J_{H2,H3}$ 3.4 Hz), and the dithiocarbonate (**4**) ($J_{H2,H3}$ 2.9 Hz) are very similar, the triflate (**3**) also probably has the *ribo* configuration; also, all the other proton coupling constants in the triflate (**3**) and the azide (**10**) are essentially the same, supporting the hypothesis that the compounds have much the same geometry. Thus the displacement of triflate in (**3**) by azide proceeds efficiently with overall retention of configuration. It is clear from the crystal structure of the *ribo*-azide (**10**) (Figure 1) that the 1,3-dioxolane ring holds the lactone in a boat conformation, in which the azide is in the bowsprit position; the epimeric *arabino*-azide (**9**) would thus have the azide in the flagpole position in a boat conformation. One possible explanation of the observed retention in the conversion of (**3**) into *ribo*-azide (**10**) is equilibration of the initially formed *arabino*-azide (**9**) to the non-flagpole position under the reaction conditions; any pathways involving neighbouring group participation would require lactone ring opening and reclosure and may be considered to be less likely. Although the displacement of the triflate group in (**3**) by azide occurs rapidly and efficiently at room temperature, reaction of the corresponding mesyl ester (**5**) under the same or more vigorous conditions, gave no products containing an azide function;[21,*] this provides a further example of the value of triflate as an excellent leaving group in nucleophilic displacements in carbohydrate chemistry.

* The major products are derived from competitive elimination initiated by abstraction of C–H followed by fragmentation with loss of benzaldehyde.

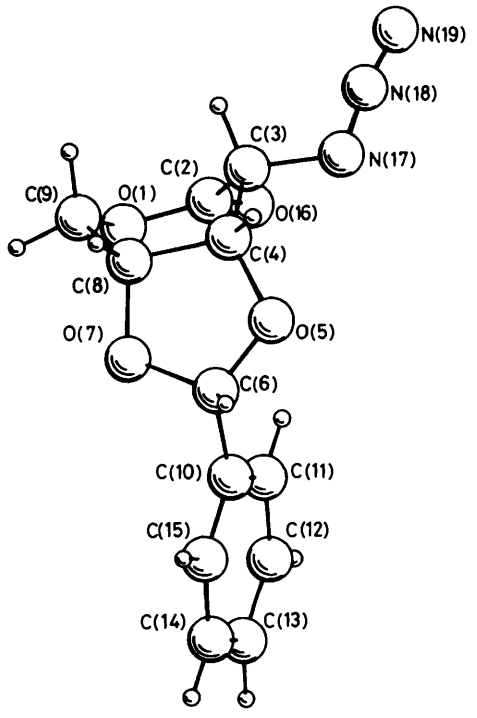


Figure 1. *X*-Ray molecular structure of 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**) with the crystallographic numbering scheme

Hydrolysis of the 1,5-lactone (**10**) with aqueous trifluoroacetic acid (TFA) caused removal of the benzylidene protecting group and the conversion of the 1,5- into the 1,4-lactone (**11**) in 94% yield. Although the stereochemical integrity at C-2 of aldonolactones is sometimes vulnerable to epimerisation under such conditions,[22] the structure of 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**) was established by *X*-ray crystallography (Figure 2) and clearly showed that the *ribo* configuration had been maintained during this transformation. Selective esterification of the primary hydroxy group in the azido diol (**11**) with methanesulphonyl chloride in pyridine at −20 °C gave the mesyl ester (**12**) in 71% yield.

^{13}C N.m.r. spectroscopy may be used to establish the size of the lactone ring in these compounds (Tables 1 and 2). Thus the chemical shifts of C-3 and C-4 in the 1,5-lactones (**2**), (**3**), (**4**), (**6**),

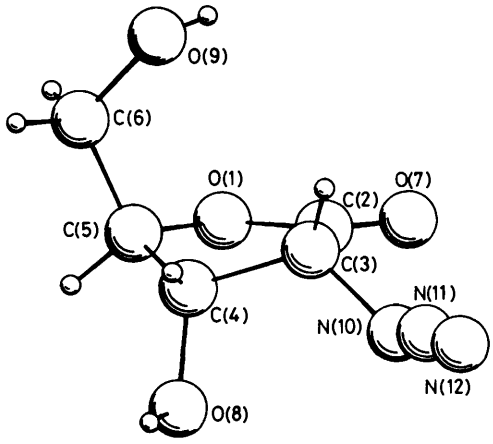
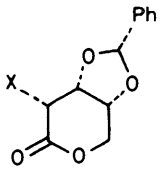


Figure 2. *X*-Ray molecular structure of 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**) with the crystallographic numbering scheme

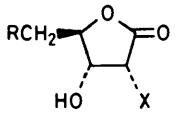
Table 1. I.r. and ^{13}C n.m.r. data for 1,5-lactones (**2**)—(**6**), (**10**)



Lactone[a]	C-3, C-4[b]	C-5	$\nu_{max.}$(C=O)/cm^{-1}
(**2**) X = OH	73.3, 76.7	66.8	1 750
(**3**) X = $OS(O)_2CF_3$	73.7, 74.3	67.8	1 770
(**4**) X = OC(S)SMe	73.6, 74.4	67.6	1 785
(**6**) X = H	72.4, 72.5	68.2	1 745
(**10**) X = N_3	73.1, 76.3	67.3	1 755

[a] Data for (**2**), (**4**), and (**6**) from ref. 18; the structures of these compounds were incorrectly assigned as 1,4-lactones. [b] No assignment of C-3 and C-4 resonances is claimed.

Table 2. I.r. and ^{13}C n.m.r. data for 1,4-lactones (**1**), (**11**)—(**13**)

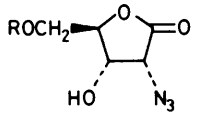


Lactone[a]	C-3	C-4	C-5	$\nu_{max.}$(C=O)/cm^{-1}
(**1**) R = X = OH	69.7	85.7	60.9	
(**11**) R = OH, X = N_3	72.2	88.5	62.0	1 765
(**12**) R = OMs, X = N_3	71.2	84.7	69.9	1 780
(**13**) R = X = H	73.1	84.6	18.6	1 760

[a] Data for (**1**) and (**13**) from ref. 18.

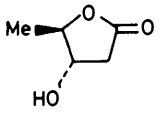
and (**10**) are all within the range δ_C 72.4—76.7 (Table 1), whereas in the 1,4-lactones (**1**), (**11**), (**12**), and (**13**) C-3 resonates in the range δ_C 69.7—73.1 and C-4 resonates at significantly lower field in the range δ_C 84.6—88.5 (Table 2). Also, the signal for C-5 in the 1,5-lactones is at lower field (δ_C 66.8—68.2) than that for C-5 in the unfunctionalised lactones (**1**) and (**11**).

It is noteworthy that the carbonyl stretching frequency is not reliable for establishing the size of the lactone ring in this class of compound; very high carbonyl frequencies (up to 1 790 cm^{-1}) have been observed for 1,5-lactones, and may indicate generally that the lactone is in an approximate boat conformation.[23]



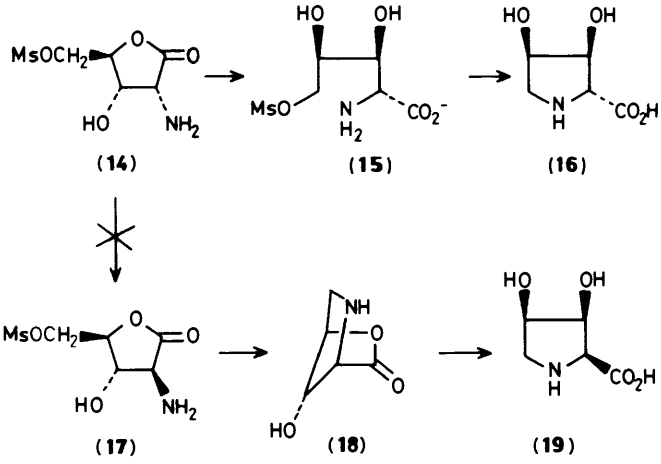
(**11**) R = H

(**12**) R = $S(O)_2Me$



(**13**)

Hydrogenation of compound (**12**) in the presence of palladium black followed by treatment of the resulting amino mesyl ester (**14**) with aqueous sodium hydroxide gave, after purification by ion-exchange chromatography, (2*R*,3*S*,4*R*)-dihydroxyproline (**16**) in 51% yield (20% overall yield from ribonolactone). The formation of compound (**16**) in this sequence requires initial hydrolysis of the lactone ring to form the carboxylate ion (**15**) which undergoes intramolecular ring closure (Scheme 2); it was possible that epimerisation to (**17**) had occurred prior to hydrolysis of the lactone ring, and that this may have led to the bicyclic lactone (**18**), which on hydrolysis would yield the all-*cis* amino acid (**19**). Therefore the structure of compound (**16**) was



(**14**) (**15**) (**16**) (**17**) (**18**) (**19**)

Scheme 2.

unambiguously shown to be (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (Figure 3) by *X*-ray crystallography; the racemic form of this amino acid has been previously prepared by osmium tetraoxide oxidation of 2,5-dihydropyrrole-1-carboxylic acid but no n.m.r. data were reported.[24]

Although the majority of naturally occurring non-protein amino acids have the L-configuration, there are many polyfunctionalised D-amino acids which are natural products;[1] the azido-1,5-lactone (**10**) and azido-1,4-lactone (**11**) are readily prepared by the above procedures on a 10 g scale in 57 and 54% yield from ribonolactone, and they may be convenient intermediates for the enantiospecific synthesis of a number of D-amino acids.

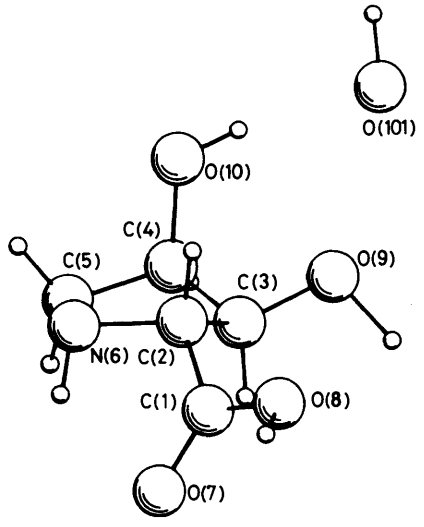


Figure 3. *X*-Ray molecular structure of (2*R*,3S,4*R*)-3,4-dihydroxyproline (**16**) with the crystallographic numbering scheme

We consider that this simple sequence of reactions illustrates the need for the use of *X*-ray crystallographic structure determination in this area of synthesis; other spectroscopic techniques, in particular vicinal proton coupling constants, are not satisfactory for determining the stereochemistry of the substituents in the five-membered-ring compounds.

Experimental

M.p.s were recorded on a Kofler block. I.r. spectra were recorded on a Perkin-Elmer 297 spectrophotometer. ^1H N.m.r.

spectra were run at 300 MHz on a Bruker WH 300 spectrometer (500 MHz on a Bruker AM 500 spectrometer); ^{13}C N.m.r. spectra were recorded on a Bruker AM 250 (62.9 MHz) or a Bruker AM 500 (125.0 MHz) spectrometer. All n.m.r. spectra were obtained using deuteriochloroform as solvent unless otherwise stated; for n.m.r. spectra in D_2O, 1,4-dioxane (δ_C 67.6) was used as the internal standard. Mass spectra were recorded on VG Micromass ZAB 1F or MM 30 F spectrometers; in order to obtain satisfactory mass spectra for these highly polar compounds, it was generally necessary to use DCI or FAB techniques. Microanalyses were performed by the microanalytical services of the Dyson Perrins Laboratory. T.l.c. was performed on glass plates coated with silica gel Blend 41, and compounds were visualised with a spray of 5% v/v sulphuric acid in ethanol or 5% dodecamolybdophosphoric acid in ethanol. Flash chromatography was carried out on Merck Kieselgel 60, 230—400 mesh. Dowex 50x 8—100 ion-exchange resin was obtained from the Aldrich Chemical Company. D-Ribonolactone (**1**) was supplied by the Aldrich Chemical Company and was converted into 3,4-*O*-(*R*)-benzylidene-D-ribono-1,5-lactone (**2**) in 89% yield as previously described.[18]

3,4-O-(R)-*Benzylidene*-2-O-*trifluoromethylsulphonyl*-D-*ribono*-1,5-*lactone* (**3**) *and* 2-*Azido*-3,4-O-(R)-*benzylidene*-2-*deoxy*-D-*ribono*-1,5-*lactone* (**10**).—3,4-*O*-(*R*)-Benzylidene-D-ribono-1,5-lactone (**2**) (13.96 g, 59.1 mmol) was suspended in dry pyridine (150 ml) and the reaction mixture was cooled to −10 °C nitrogen. Trifluoromethanesulphonic anhydride (20 g, 1.2 mol equiv., 72 mmol) was added to the stirred reaction mixture at between −10 and −20 °C during 15 min; the resulting suspension was stirred at the same temperature for a further 1 h, at which time the reaction mixture had become homogeneous. The pyridine solution was diluted with ethyl acetate (300 ml), then washed successively with water (300 ml) and saturated aqueous sodium hydrogen carbonate (300 ml), and dried (sodium sulphate), and the solvent was removed to give the required *triflate* (**3**) as a yellow solid, which was used directly without further purification for the conversion into the azido lactone (**10**). A small amount of the triflate was recrystallised from ethanol to give 3,4-*O*-(*R*)-*benzylidene*-2,O-*trifluoromethylsulphonyl*-D-*ribono*-1,5-*lactone* (**3**) as white needles, m.p. 172—173 °C (decomp.); $[\alpha]_D^{20}$ − 128.6° (*c* 0.56 in EtOAc); $\nu_{max.}$(Nujol) 1 770 (C=O), 1 455, 1 410, and 1 090 cm^{-1}; δ_H 7.4—7.5 (5 H, m, ArH), 5.87 (1 H, s, PhC*H*), 5.35 (1 H, d, $J_{2,3}$ 3.4 Hz, 2-H), 5.00 (1 H, dd, $J_{3,4}$ 8.0 Hz, 3-H), 4.79 (1 H, d, 4-H), 4.68 (1 H, d, $J_{5,5'}$ 13.6 Hz, 5-H), and 4.38 (1 H, dd, $J_{4,5'}$ 1.7 Hz, 5′-H); δ_C[$(CD_3)_2SO$] 67.84 (t, C-5), 73.7 and 74.3 (2 d, C-3 and -4), 80.9 (d, C-2), 103.6 (d, PhCH), 119.0 (q, CF_3, not proton decoupled), 127.2 (d), 128.5 (d), 130.3 (d), 135.3 (s), and 164.8 (s, C-1); *m/z* (DCI, NH_3) 386 (100%, *M* + NH_4^+) (Found: C, 42.4; H, 3.0. $C_{13}H_{11}F_3O_7S$ requires C, 42.39; H, 2.99%).

The crude triflate (**3**) was dissolved in DMF (100 ml) and the solution was stirred with sodium azide (6.4 g, 98.5 mmol) at room temperature for 1.5 h. The DMF was then removed from the reaction mixture by evaporation and the residue was partitioned between ethyl acetate (300 ml) and water (300 ml). The organic layer was dried (sodium sulphate), and the solvent was removed to give, after crystallisation from ethanol, 2-*azido*-3,4-O-(R)-*benzylidene*-2-*deoxy*-D-*ribono*-1,5-*lactone* (**10**) (9.8 g, 64% from alcohol), m.p. 145 °C (decomp.); $[\alpha]_D^{20}$ −245.5° (*c* 0.42 in $CHCl_3$); $\nu_{max.}$($CHCl_3$) 2 110 (N_3), 1 755 (C=O), 1 460, 1 405, 1 160, and 1 090 cm^{-1}, δ_H 7.4—7.5 (5 H, m, ArH), 5.78 (1 H, s, PhC*H*), 4.91 (1 H, dd, $J_{3,4}$ 8.0, $J_{2,3}$ 3.3 Hz, 3-H), 4.67 (1 H, dd, 4-H), 4.61 (1 H, d, $J_{5,5'}$ 13.4 Hz, 5-H), 4.25 (1 H, dd, $J_{4,5'}$ 1.7 Hz, 5′-H), and 3.95 (1 H, d, 2-H); δ_C 58.9 (d, C-2), 67.3 (t, C-5), 73.1 and 76.3 (2 d, C-3 and -4), 104.6 (d, PhCH) 127.1 (d), 128.6 (d), 130.3 (d), 134.4 (s), and 166.4 (s, C-1); *m/z* (DCI, NH_3) 279 (60%, *M* + NH_4^+) and 234 (100, *M* + H^+ − N_2) (Found: C, 55.1; H, 4.2; N, 16.0. $C_{12}H_{11}N_3O_4$ requires C, 55.17; H, 4.21; N, 16.09%).

2-*Azido*-2-*deoxy*-D-*ribono*-1,4-*lactone* (**11**).—2-Azido,3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**) (5 g, 19.2 mmol) was dissolved in stirred aqueous TFA (67% v/v; 39 ml) at 50 °C during 1.5 h. Evaporation of the solvent, followed by flash chromatography of the residue [ethyl acetate–hexane (3:2)], afforded 2-*azido*-2-*deoxy*-D-*ribono*-1,4-*lactone* (**11**) (3.1 g, 94%), m.p. 84—85 °C (from ether); $[\alpha]_D^{20}$ + 54.2° (*c* 0.28 in EtOAC); $\nu_{max.}$($CHCl_3$) 3 400br, 2 105 (N_3), and 1 765 cm^{-1} (C=O); δ_H [$(CD_3)_2CO$] 5.28 (1 H, br, d, D_2O exchange, OH), 4.61 (1 H, d, $J_{2,3}$ 5.3 Hz, 3-H), 4.50 (1 H, t, 4-H), 4.44 (1 H, d, 2-H), 3.83 (2 H, d, $J_{4,5}$ 3.0 Hz, 5-H), and 2.98 (1 H, br s, D_2O exchange, OH); δ_C [$(CD_3)_2SO$] 2.04 (t, C-5), 62.26 (d, C-2), 72.17 (2 d, C-3), 88.51 (d, 4-C), and 174.5 (s, C-1); *m/z* (DCI, NH_3) 191 (100%, *M* + NH_4^+) and 146 (10, *M* + H^+ − N_2) (Found: C, 34.7; H, 4.0; N, 24.15. $C_5H_7N_3O_4$ requires C, 34.68; H, 4.05; N, 24.28%).

2-*Azido*-2-*deoxy*-5-O-*methylsulphonyl*-D-*ribono*-1,4-*lactone* (**12**).—A solution of 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**) (1.07 g, 6.17 mmol) in dry pyridine (40 ml) at −20 °C under nitrogen was treated with methanesulphonyl chloride (0.49 ml, 1.1 mol equiv.), and the reaction mixture was kept overnight. The bulk of the pyridine was removed under reduced pressure and the residue was diluted with ethyl acetate (100 ml); the solution was then washed successively with aqueous hydrochloric acid (2M; 50 ml), water (100 ml), and saturated aqueous sodium hydrogen carbonate (100 ml), and dried (sodium

Table 3. Crystal data for 2-azido-3,4-*O*(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**), 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**), and (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**)

	(**10**)	(**11**)	(**16**)
Formula	$C_{12}H_{11}N_3O_4$	$C_5H_7N_3O_4$	$C_5H_{11}NO_5$
M_r	261.34	173.128	165.146
Crystal size/mm	0.2 × 0.1 × 0.8	0.5 × 0.7 × 0.8	1.0 × 0.1 × 0.1
Crystal class	tetragonal	monoclinic	orthorhombic
a/Å	8.752(1)	6.324(1)	4.949(1)
b/Å	8.752(1)	11.013(3)	7.755(1)
c/Å	15.755(3)	5.135(1)	18.693(4)
α/°	90	90	90
β/°	90	94.47(4)	90
γ/°	90	90	90
Space group	$P4_1$	$P2_1$	$P2_12_12_1$
Z	4	2	4
D_c/Mg m^{-3}	1.438	1.613	1.529
F(000)	544	180	352
V/Å^3	1 206.8	356.54	717.43
Radiation	Cu-K_α	Mo-K_α	Cu-K_α
10^{-2} μ/m^{-1}	8.9	1.3	11.4
$(\sin\theta/\lambda)_{max.}$	0.61	0.81	0.59
Total I[a]	3 635	2 258	1 225
Unique I[b]	1 083	1 304	572
$R_m/10^{-2}$	7.12	7.54	7.08
n[c]	3	3	3
$R/10^{-2}$	4.1	5.0	4.4
$R_w/10^{-2}$	5.3	6.8	4.8
Shift/error[d]	0.00	0.00	0.00
$\Delta_{max.}$/e Å^{-3}[e]	<0.05	0.28	0.24
Ext. para.	87	203.0	19.0
Weights	680, 928, 285, 25	unit weights	unit weights

[a] Total number of reflections measured. [b] Number of unique reflections with intensity significantly above the background intensity. [c] Criterion for recognising observed reflections $I > n\sigma(I)$. [d] Ratio of maximum least-squares shift to error in final refinement cycle. [e] Maximum height in final difference electron density synthesis.

Table 4(i). Fractional atomic co-ordinates with e.s.d.s in parentheses (atomic labelling as in Figure 1) for 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**)

Atom	*x*	*y*	*z*
C(2)	0.321 9(5)	0.307 9(5)	−0.581 4(4)
C(3)	0.339 3(4)	0.139 2(4)	−0.579 6(3)
C(4)	0.297 8(4)	0.078 7(4)	−0.491 8(3)
C(6)	0.135 6(4)	0.146 6(4)	−0.386 8(3)
C(8)	0.388 1(4)	0.163 9(5)	−0.423 5(4)
C(9)	0.480 1(5)	0.293 7(5)	−0.458 9(4)
C(10)	0.003 6(4)	0.246 7(4)	−0.365 4(3)
C(11)	−0.059 7(4)	0.345 3(4)	−0.424 0(3)
C(12)	−0.175 2(4)	0.444 6(5)	−0.400 0(4)
C(13)	−0.227 7(4)	0.444 6(5)	−0.316 1(4)
C(14)	−0.165 2(5)	0.345 2(5)	−0.258 4(4)
C(15)	−0.052 2(5)	0.244 9(5)	−0.282 1(4)
O(1)	0.392 7(4)	0.384 6(3)	−0.519 1(3)
O(5)	0.142 9(2)	0.116 9(3)	−0.475 2(3)
O(7)	0.273 4(3)	0.227 0(3)	−0.368 8(3)
O(16)	0.250 3(5)	0.377 7(4)	−0.633 8(4)
N(17)	0.240 4(4)	0.063 2(5)	−0.641 0(3)
N(18)	0.277 7(4)	0.081 0(5)	−0.716 3(3)
N(19)	0.298 7(7)	0.081 4(8)	−0.786 6(4)

Table 4(ii). Bond lengths (Å) for the non-hydrogen atoms with e.s.d.s in parentheses (atomic labelling as in Figure 1) for 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**)

C(2)–C(3)	1.484(5)
C(2)–O(1)	1.342(5)
C(2)–O(16)	1.203(5)
C(3)–C(4)	1.526(5)
C(3)–N(17)	1.458(5)
C(4)–C(8)	1.528(5)
C(4)–O(5)	1.421(4)
C(6)–C(10)	1.489(5)
C(6)–O(5)	1.418(4)
C(6)–O(7)	1.424(4)
C(8)–C(9)	1.500(6)
C(8)–O(7)	1.434(4)
C(9)–O(1)	1.455(6)
C(10)–C(11)	1.380(5)
C(10)–C(15)	1.400(5)
C(11)–C(12)	1.386(5)
C(12)–C(13)	1.400(7)
C(13)–C(14)	1.372(7)
C(14)–C(15)	1.374(6)
N(17)–N(18)	1.240(5)
N(18)–N(19)	1.123(5)

Table 4(iii). Bond angles (°) for the non-hydrogen atoms with e.s.d.s in parentheses (atomic labelling as in Figure 1) for 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**)

O(1)–C(2)–C(3)	115.9(3)
O(16)–C(2)–C(3)	124.9(4)
O(16)–C(2)–O(1)	119.2(4)
C(4)–C(3)–C(2)	109.7(3)
N(17)–C(3)–C(2)	112.3(3)
N(17)–C(3)–C(4)	107.5(3)
C(8)–C(4)–C(3)	110.2(3)
O(5)–C(4)–C(3)	108.2(3)
O(5)–C(4)–C(8)	104.4(3)
O(5)–C(6)–C(10)	111.4(3)
O(7)–C(6)–C(10)	108.7(3)
O(7)–C(6)–O(5)	104.4(3)
C(9)–C(8)–C(4)	112.7(3)
O(7)–C(8)–C(4)	104.4(3)
O(7)–C(8)–C(9)	107.9(3)
O(1)–C(9)–C(8)	111.9(3)
C(11)–C(10)–C(6)	121.9(3)
C(15)–C(10)–C(6)	118.4(3)
C(15)–C(10)–C(11)	119.6(3)
C(12)–C(11)–C(10)	120.1(3)
C(13)–C(12)–C(11)	119.8(4)
C(14)–C(13)–C(12)	119.7(4)
C(15)–C(14)–C(13)	120.8(4)
C(14)–C(15)–C(10)	119.9(4)
C(9)–O(1)–C(2)	116.5(3)
C(6)–O(5)–C(4)	105.4(2)
C(8)–O(7)–C(6)	106.4(3)
N(18)–N(17)–C(3)	114.8(3)
N(17)–N(18)–N(19)	170.8(5)

Table 5(i). Fractional atomic co-ordinates with e.s.d.s in parentheses (atomic labelling as in Figure 2) for 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**)

Atom	*x*	*y*	*z*
C(2)	0.706 0(6)	−0.433 8(4)	0.946 0(7)
C(3)	0.728 3(6)	−0.487 2(4)	1.222 0(7)
C(4)	0.503 6(7)	−0.529 0(3)	1.266 7(7)
C(5)	0.365 6(6)	−0.446 4(3)	1.088 4(7)
C(6)	0.287 9(7)	−0.331 4(3)	1.210 0(9)
O(1)	0.499 4(4)	−0.412 9(3)	0.878 3(5)
O(7)	0.842 2(5)	−0.412 2(4)	0.808 2(6)
O(8)	0.466 0(6)	−0.650 6(3)	1.178 7(6)
O(9)	0.457 9(6)	−0.255 6(3)	1.309 4(6)
N(10)	0.889 0(7)	−0.582 4(5)	1.223 5(8)
N(11)	0.984 6(5)	−0.607 1(4)	1.431 2(7)
N(12)	1.085 1(8)	−0.640 8(6)	1.608 8(9)

Table 5(ii). Bond lengths (Å) for the non-hydrogen atoms with e.s.d.s in parentheses (atomic labelling as in Figure 2) for 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**)

C(2)–C(3)	1.530(5)
C(2)–O(1)	1.345(5)
C(2)–O(7)	1.181(5)
C(3)–C(4)	1.528(6)
C(3)–N(10)	1.460(6)
C(4)–C(5)	1.518(5)
C(4)–O(8)	1.427(5)
C(5)–C(6)	1.511(5)
C(5)–O(1)	1.469(5)
C(6)–O(9)	1.424(5)
N(10)–N(11)	1.215(5)
N(11)–N(12)	1.133(5)

Table 5(iii). Bond angles (°) for the non-hydrogen atoms with e.s.d.s in parentheses (atomic labelling as in Figure 2) for 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**)

O(1)–C(2)–C(3)	108.8(3)
O(7)–C(2)–C(3)	127.8(4)
O(7)–C(2)–O(1)	123.3(4)
C(4)–C(3)–C(2)	103.6(3)
N(10)–C(3)–C(2)	107.1(3)
N(10)–C(3)–C(4)	116.0(4)
C(5)–C(4)–C(3)	103.1(3)
O(8)–C(4)–C(3)	111.7(3)
O(8)–C(4)–C(5)	107.3(3)
C(6)–C(5)–C(4)	116.3(3)
O(1)–C(5)–C(4)	104.9(3)
O(1)–C(5)–C(6)	108.5(3)
O(9)–C(6)–C(5)	112.2(3)
C(5)–O(1)–C(2)	111.7(3)
N(11)–N(10)–C(3)	117.7(4)
N(12)–N(11)–N(10)	171.7(5)

Table 6(i). Fractional atomic co-ordinates with e.s.d.s in parentheses (atomic labelling as in Figure 3) for (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**)

Atom	*x*	*y*	*z*
C(1)	0.182(1)	0.145 0(7)	0.273 3(3)
C(2)	0.348(1)	0.170 8(7)	0.206 7(2)
C(3)	0.276(1)	0.043 4(6)	0.146 4(3)
C(4)	0.347(1)	0.138 9(8)	0.078 7(3)
C(5)	0.262(1)	0.321 6(7)	0.093 8(3)
N(6)	0.294(1)	0.344 6(6)	0.173 8(2)
O(7)	−0.040 8(8)	0.221 0(6)	0.274 9(2)
O(8)	0.262 4(9)	0.041 7(5)	0.319 8(2)
O(9)	0.415 3(8)	−0.113 8(5)	0.153 2(2)
O(10)	0.636 0(8)	0.135 1(6)	0.068 9(2)
O(101)	0.805 9(9)	−0.226 0(7)	0.039 0(2)

Table 6(ii). Bond lengths (Å) for the non-hydrogen atoms with e.s.d.s in parentheses (atomic labelling as in Figure 3) for (2*R*, 3*S*, 4*R*)-3,4-dihydroxyproline (**16**)

C(1)–C(2)	1.508(7)
C(1)–O(7)	1.249(7)
C(1)–O(8)	1.248(6)
C(2)–C(3)	1.541(7)
C(2)–N(6)	1.506(6)
C(3)–C(4)	1.509(7)
C(3)–O(9)	1.406(6)
C(4)–C(5)	1.505(8)
C(4)–O(10)	1.440(7)
C(5)–N(6)	1.514(6)

Table 6(iii). Bond angles (°) for the non-hydrogen atoms with e.s.d.s in parentheses (atomic labelling as in Figure 3) for (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**)

O(7)–C(1)–C(2)	116.1(5)
O(8)–C(1)–C(2)	119.0(5)
O(8)–C(1)–O(7)	124.7(5)
C(3)–C(2)–C(1)	113.0(4)
N(6)–C(2)–C(1)	111.0(4)
N(6)–C(2)–C(3)	103.5(4)
C(4)–C(3)–C(2)	104.1(4)
O(9)–C(3)–C(2)	112.0(5)
O(9)–C(3)–C(4)	112.7(5)
C(5)–C(4)–C(3)	103.8(5)
O(10)–C(4)–C(3)	109.3(5)
O(10)–C(4)–C(5)	108.7(6)
N(6)–C(5)–C(4)	105.5(5)
C(5)–N(6)–C(2)	108.4(4)

sulphate). Evaporation of the solvent, followed by flash chromatography of the residue [ethyl acetate–hexane (2:1)], gave unchanged starting material (0.18 g, 7%) and 2-*azido*-2-*deoxy*-5-O-*methylsulphonyl*-D-*ribono*-1,4-*lactone* (**12**) (1.1 g, 71% yield), m.p. 68—69 °C (from $CHCl_3$); $[\alpha]_D^{20}$ +37.3° (*c* 0.48 in EtOAc); $\nu_{max.}$(film) 3 450br, 2 110 (N_3), 1 780 (C=O), 1 350, and 1 165 cm^{-1}; δ_H ($CDCl_3$) 4.67 (1 H, m, 4-H), 4.52 (1 H, dd, 3-H), 4.50—4.45 (3 H, m, 2-, 5-, and 5′-H), 3.09 (3-H, s, Me), and 2.9 (1 H, br s, OH); δ_C (CD_3OD) 37.39 (q, CH_3), 61.61 (d, C-2), 69.06 (t, 5-C), 71.17 (d, C-3), 84.70 (d, C-4), and 174 (s, C-1); *m*/*z* (DCI, NH_3) 269 (100%, $M + NH_4^+$) and 224 (10, $M + H^+ - N_2$) (Found: C, 28.4; H, 3.5; N, 16.4. $C_6H_9N_3O_6S$ requires C, 28.7; H, 3.6; N, 16.7%).

(2R,3S,4R)-3,4-*Dihydroxyproline* (**16**).—2-Azido-2-deoxy-5-*O*-methylsulphonyl-D-ribono-1,4-lactone (**12**) (0.72 g, 2.8 mmol) was dissolved in ethyl acetate (8 ml) and the solution was stirred at room temperature under hydrogen in the presence of palladium black (0.1 g). After 24 h, the ethyl acetate was removed under reduced pressure and the residue was suspended in water (30 ml). Aqueous sodium hydroxide (2M; 1.4 ml, 2.8 mmol) was added and the suspension was stirred at room temperature for 24 h until only catalyst remained undissolved. The solution was then filtered and evaporated to afford a brown syrup, which was purified by ion-exchange chromatography (Dowex 50x 8—100 ion-exchange resin, H^+ form, eluted with 1M-aqueous pyridine) to give, after freeze drying, (2R,3S,4R)-3,4-*dihydroxyproline* (**16**) (0.21 g, 51%) as a white solid (from aqueous acetone or aqueous ethanol) which decomposes (without melting) at 247 °C, $[\alpha]_D^{20}$ −6.8° (*c* 0.43 in water); $\nu_{max.}$(KBr) 3 401, 3 240, 3 099, 3 036, 2 927, 2 713, 2 565, 2 427, 1 652, 1 615, and 1 568 cm^{-1}, δ_H (D_2O) 4.2—4.25 (2 H, m, 3- and 4-H), 3.85 (1 H, d, $J_{2,3}$ 5.0 Hz, 2-H), 3.41 (1 H, dd, $J_{4,5'}$ 4.9 Hz, 5′-H), and 3.17 (1 H, dd, $J_{4,5}$ 4.2, $J_{5,5'}$, 5-H); 12.4 Hz, 5-H); δ_C (D_2O) 48.21 (t, C-5), 64.13 (d, C-2), 69.77 (d, C-4), 73.92 (d, C-3), and 171.9 (s, C-1); *m*/*z* (FAB$^+$) 148 (100%, $M + H^+$) (Found: C, 40.9; H, 6.3; N, 9.8. $C_5H_9NO_4$ requires C, 40.82; H, 6.12; N, 9.52%).

X-*Ray Crystal Structure Analyses*.—The crystal data for 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**), 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**), and (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**) are given in Table 3. Crystals were prepared as described above and recrystallised as follows: 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone from ethanol, 2-azido-2-deoxy-D-ribono-1,4-lactone from acetone, and (2*R*,3*S*,4*R*)-3,4-dihydroxyproline from aqueous ethanol.

X-Ray data were collected with an Enraf-Nonius CAD4 diffractometer following the procedures recommended in the manufacturer's manual. The data were corrected for Lorentz and polarisation effects and for absorption. All calculations were carried out on a VAX 11/750 computer using MULTAN80[25] [for (**11**) and (**16**)] or MITHRIL[26] [for (**10**)] for direct methods and CRYSTALS[27] for all other calculations. Atomic scattering factors were taken from standard tables.[28] The positions of all non-hydrogen atoms were given by the appropriate direct methods routine and the C–H hydrogen-atom positions were either obtained geometrically [for (**10**) and (**11**)] or located from difference maps [for (**16**)]. The OH hydrogen-atom positions were located from difference maps. The trial structures were then refined by full-matrix least-squares, with the hydrogen-atom positions linked to those of the atoms to which they were attached. Hydrogen-atom temperature factors were refined isotropically, except those of (**16**) where four of the hydrogen-atom temperature factors were set at U_{iso} 0.1 and the remaining hydrogen-atom temperature factors were refined anisotropically. All non-hydrogen-atom positional parameters and temperature factors were refined anisotropically. Figures (1), (2), and (3) show the *X*-ray molecular structures of 2-azido-3,4-*O*-(*R*)-benzylidene-2-deoxy-D-ribono-1,5-lactone (**10**), 2-azido-2-deoxy-D-ribono-1,4-lactone (**11**), and (2*R*,3*S*,4*R*)-3,4-dihydroxyproline (**16**) respectively with their crystallographic numbering schemes. Atomic co-ordinates, bond lengths, and bond angles are given in Tables 4—6.*

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* *Supplementary data* [see section 5.6.3 of Instructions for Authors (1987), in the January issue]. H-Atom co-ordinates and thermal parameters have been deposited at the Cambridge Crystallographic Data Centre.

References

1 B. Hunt in 'Non-Protein Amino Acids,' Ch. 4 in 'Chemistry and Biochemistry of Amino Acids,' ed. G. C. Barratt, Chapman and Hall, London, 1985; I. Wagner and H. Musso, *Angew. Chem., Int. Ed. Engl.*, 1983, **22**, 816.
2 W. R. Ewing, B. D. Harris, K. L. Bhat, and M. M. Joullie, *Tetrahedron*, 1986, **42**, 2421.
3 A. L. Castelhano, S. Horne, R. Billedeau, and A. Krantz, *Tetrahedron Lett.*, 1986, **27**, 2435.
4 S. Hannessian, 'Total Synthesis of Natural Products: The Chiron Approach,' Pergamon, Oxford, 1983.
5 M. Miyashita, N. Chida, and A. Yoshikoshi, *J. Chem. Soc., Chem. Commun.*, 1984, 195.
6 R. C. Bernotas and B. Ganem, *Tetrahedron Lett.*, 1985, **26**, 4981.
7 T. Wakamiya, K. Yamanoi, M. Nishikawa, and T. Shiba, *Tetrahedron Lett.*, 1985, **26**, 4759.
8 P. Shenbagamurthi, H. A. Smith, J. M. Becker, and F. Naider, *J. Med. Chem.*, 1986, **29**, 802, and references therein.
9 T. Kitihara, T. Ogawa, T. Naganuma, and M. Matsui, *Agric. Biol. Chem.*, 1974, **38**, 2189.
10 B. P. Bashyal, H.-F. Chow, and G. W. J. Fleet, *Tetrahedron Lett.*, 1986, **27**, 3205; B. P. Bashyal, H.-F. Chow, L. E. Fellows, and G. W. J. Fleet, *Tetrahedron*, 1987, **43**, 415; B. P. Bashyal, H.-F. Chow, and G. W. J. Fleet, *ibid*, 1987, **43**, 423.
11 A. G. M. Barrett and H. G. Sheth, *J. Chem. Soc., Chem. Commun.*, 1982, 170.
12 J. Mann and A. Thomas, *J. Chem. Soc., Chem. Commun.*, 1985, 737.
13 S. Y. Chen and M. M. Joullie, *Tetrahedron Lett.*, 1983, **24**, 5027.
14 J. C. Dho, G. W. J. Fleet, J. M. Peach, K. Prout, and P. W. Smith, *Tetrahedron Lett.*, 1986, **27**, 5203.
15 Y. Ohfune and N. Kurokawa, *Tetrahedron Lett.*, 1985, **26**, 5307.
16 H. Zinner, H. Voight, and J. Voight, *Carbohydr. Res.*, 1968, **7**, 38.
17 N. Baggett, J. G. Buchanan, M. Y. Fatah, C. H. Lachut, K. J. McCullough, and J. M. Webber, *J. Chem. Soc., Chem. Commun.*, 1985, 1826.
18 S. Y. Chen and M. M. Joullie, *J. Org. Chem.*, 1984, **49**, 2168.
19 G. W. J. Fleet, T. K. M. Shing, and S. M. Warr, *J. Chem. Soc., Perkin Trans. 1*, 1984, 905.
20 R. Cauk, H. Honig, J. Nimpf, and H. Weidmann, *Tetrahedron Lett.*, 1980, **21**, 2135.
21 G. W. J. Fleet and B. P. Bashyal, unpublished results.
22 K. Bock, I. Lundt, and C. Pedersen, *Carbohydr. Res.*, 1981, **90**, 17.
23 S. Mirza, L.-P. Molleyers, and A. Vasella, *Helv. Chim. Acta*, 1985, **68**, 988; R. A. Edwards, G. W. J. Fleet, R. H. Jones, and T. K. M. Shing, unpublished results; K. H. Overton, N. G. Weir, and A. Wylie, *J. Chem. Soc. C*, 1965, 1482.
24 C. R. Hudson, A. V. Robertson, and W. R. J. Simpson, *Aust. J. Chem.*, 1968, **21**, 769.
25 P. Main, MULTAN80, A System of Computer Programs for the Automatic Solution of Crystal Structures from X-ray Diffraction Data, Dept. of Physics, University of York, 1980.
26 C. J. Gilmore, MITHRIL, A Computer Program for the Automatic Solution of Crystal Structures from X-Ray Data, Dept. of Chemistry, University of Glasgow, 1983.
27 D. J. Watkins, J. R. Carruthers, and P. W. Betteridge, CRYSTALS User Guide, Chemical Crystallography Laboratory, University of Oxford, 1985.
28 International Tables for X-Ray Crystallography, Vol. IV, Kynoch Press, Birmingham, 1974.

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