135-137 °C. There is thus obtained the desired **6** in 80% yield at a purity level of 94%. That **6** and **7** have the same relative stereochemistry at carbons 2 and 3, but differ only in the C_3-C_4 relationship, is strongly suggested by their partial equilibration with alumina.[15]

Reduction of **6** with DIBAL in benzene-toluene afforded the isomers **8**. Although readily accomplished, there is no need to separate these hydroxy epimers.[14] Each compound undergoes a Ferrier-type rearrangement[16] (isopropyl alcohol, *p*-TsOH, 0 °C) to give (96%) the β-disposed *anomer* **9**. The tendency of the Ferrier rearrangement to produce an axial glycoside is undoubtedly responsible for the stereospecific formation of **9**.[14]

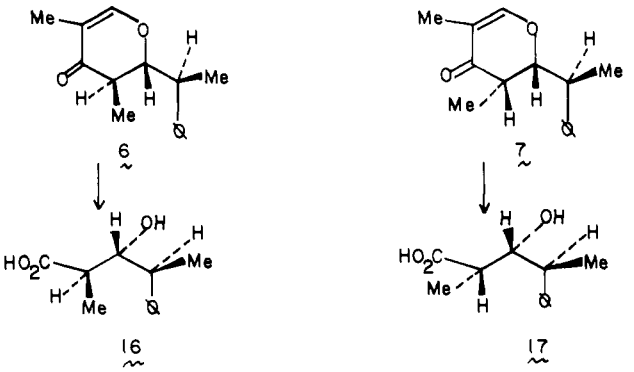
Flanked as it is by two β functions, the double bond in **9** suffers catalytic reduction (H_2/Pd-Al_2O_3; EtOAc) from its α face to afford **10**.[14] Compound **10**, mp 34-38 °C, was subjected to the action of ozone in aqueous acetic acid containing a trace of trifluoroacetic acid (4 h, room temperature). This was followed by reaction with hydrogen peroxide in aqueous acetic acid. There was thus obtained a 56% yield of **11**, mp 116-117 °C.[14] *Thus, in the ozonolysis process, the acetal linkage in* ***10*** *had suffered a Deslongchamps type of degradation,*[17] *concurrently with the more classical oxidative cleavage of its phenyl ring.*[18] The overall yield for this straightforward synthesis of **11** is currently 29% (unoptimized).

The C_6 epimer of **11**, i.e., compound **15**, can also be obtained in a stereospecific fashion from **8**. Toward this end, **8** is subjected to Ferrier rearrangement[16] by using aqueous HCl in dioxane. The resultant hemiacetal **12** on Jones oxidation gives the unsaturated lactone **13**.[14] Catalytic reduction of **13** (H_2/Pd/C; EtOH) affords, stereospecifically, the compound **6** epi system, **14**,[14] mp 66-69 °C. Thus, either the *cis*- or *trans*-4,6-dimethyl systems are available stereospecifically from a common intermediate. The structure of **14** was supported by its transformation (ozone, aqueous acetic acid, room temperature, 4 h, followed by hydrogen peroxide) to lactone **15**.[14]

We have also discovered a surprising medium effect on the stereochemistry of the cyclocondensation reaction. As noted above, when the reaction was carried out in methylene chloride at -78 °C, the ratio **6**/**7** was 4.3:1. However, when the reaction is conducted in carbon tetrachloride at room temperature, the ratio **6**/**7** becomes 1:4. Again, we could observe no loss in Cram's rule specificity.

The effect seems to arise primarily from the change in solvents since, in methylene chloride at -10 °C, the trans compound **6** still predominates. Obviously a great deal of experimentation awaits us in seeking to improve upon the cis:trans specificities and in determining the origin of this medium effect. The possibility that it reflects a change of mechanism in the cyclocondensation reaction will be explored.

Finally, we note the possibility of using the dihydropyrones as masked and manageable equivalents of β-aldols. This dimension of the new methodology was demonstrated. Ozonolysis of **6** or **7** in methanol at -78 °C followed by reaction with alkaline hydrogen peroxide resulted in the smooth formation of the known[19,20] **16** (mp 106.5-107.5 °C) and **17** (mp 136-138 °C), respectively. There is no erosion in stereochemical homogeneity in this simple unveiling procedure. Plans to exploit the synthetic equivalency between the now readily available dihydro-γ-pyrones and β-hydroxycarbonyl systems of defined stereochemistry abound, and their implementation is currently being pursued.



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(15) Of course, the equilibration of **6** and **7** per se does not establish the identity of their configurations at C_3 since the mechanism of equilibration might, in principle, involve β elimination of the oxygen from position 3 and recyclization. However, preliminary results in our laboratory (N. Kato) indicate that the recyclization process of the 1-hydroxypenta-1,4-dien-3-one (produced under different conditions) does not smoothly occur under our reaction conditions.

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Two-Dimensional NMR Investigation of Amide Proton Exchange in H_2O Solution

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This communication demonstrates the feasibility of using the Redfield[1] technique for H_2O suppression in a two-dimensional NMR study[2] of exchanging amide protons. The basic 2-D technique for studying exchange has been described in detail and applied to systems other than those which involve amide protons exchanging with an H_2O solvent.[2-4] The Redfield technique for suppressing the H_2O resonance and enhancing dynamic range has been widely used in one-dimensional NMR studies, including saturation-transfer experiments designed to elucidate exchange mechanisms.[5] Combination of the 2-D and Redfield techniques significantly extends NMR capability for studies of an important class of chemical exchange kinetics.

The Redfield excitation operates by stimulating the resonances of interest while leaving the intense H_2O resonance relatively unexcited.[1] However, in the 2-D experiment ($-90°_x$-evolution-$90°_x$-mixing-$90°_x$-detection-preparation-)$_n$ it is crucial that both resonances of the exchanging pair be excited by the first two 90° pulses of the sequence.[2,3] This is so, because for each resonance, frequency labeling (which gives the second chemical shift dimension) and kinetic development of the spin system (which allows the exchange process to be observed) must occur before observation of the free induction decay. Thus, only the third or acquisition pulse in the sequence can be a Redfield pulse. In terms of technical difficulty it may be thought that this situation is analogous to that encountered in Redfield under-water decoupling, where the

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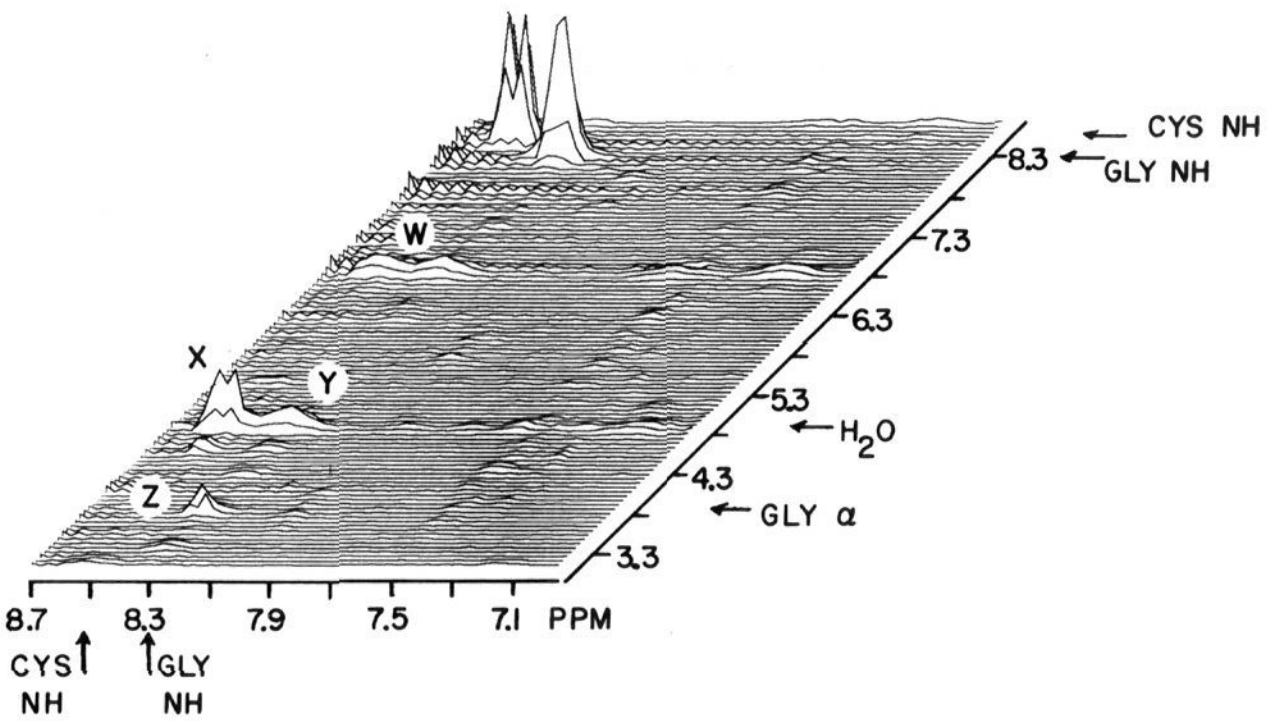

Figure 1. Two-dimensional ^1H NMR spectrum of glutathione (0.14 M, 87% H_2O/13% D_2O, pH_m 5.3, 19 °C). Data were collected as described in the text by utilizing single-phase detection, 1400-Hz bandwidth, 512 data points, and 76 scans per FID. Sine bell apodization was used in each dimension, and the spectrum shown is an absolute value spectrum. Chemical shifts are relative to TSP.

Redfield part of the protocol is designed to minimize stimulation of the H_2O resonance, but the decoupling rf unavoidably stimulates it too much, often making the experiment impossible.[6]

The situation encountered in the 2-D experiment is, in fact, more favorable than that faced in Redfield under-water decoupling for several reasons. Since the acquisition pulse in the 2-D experiment examines the spin system in a nonequilibrium state,[2,3] residual excitation of Z-axis H_2O magnetization by a Redfield pulse nutates proportionately less signal into the transverse plane. In addition, the optimum mixing periods[2] appropriate for observation of amide proton exchange (vide infra) may be long enough to permit significant decay of transverse H_2O magnetization.

Using a $(-90°_x$–evolution–$90°_x$–mixing–$90°_{x,\text{Redfield 2-1-4}}$–detection–preparation–$)_n$ sequence, we have successfully observed NH exchange in 90% H_2O solutions. However, this sequence yields undesirable axial resonances. A phase alternation scheme has been developed[3] for suppressing such resonances as well as reducing unwanted transverse magnetization without a homospoil pulse. Limitations of the pulse programmer in our spectrometer, however, prevent us from conveniently implementing this scheme along with a Redfield 2-1-4 pulse. Therefore, in order to take advantage of the phase alternation procedure, we forego the 2-1-4 modification[1] of the Redfield pulse and use a simple long, soft 90° pulse to excite the amides and suppress the H_2O when acquiring data.

Figure 1 shows the results of this compromise pulse sequence $[(-90°_x$–evolution–$90°_x$–mixing–$90°_{x,\text{soft}}$–detection–preparation–$90°_{-x}$–evolution–$90°_x$–mixing–$90°_{-x,\text{soft}}$–detection–preparation–$)_n]$ for glutathione (γ-Glu-Cys-Gly) in 90% H_2O solution. The portion of the 2-D spectrum near the H_2O resonance is not shown, because it exhibits the usual distortions which characterize Redfield spectra. The effect of these distortions in the amide region of the spectrum is greatly reduced by sine bell apodization[7] in both dimensions.

This work was performed on a NTC-200 spectrometer, utilizing the NTC-1180 computer and NTC-293A′ pulse programmer. Programmable transition between high and low power modes of transmitter operation is possible and required by the experiments discussed above. The 2-D spectrum was accumulated in the single-phase detector mode of receiver operation. Conditions for optimizing the off-diagonal exchange resonances were determined according to the theory of Jeener et al.[2] by using data from one-dimensional spin–lattice relaxation and saturation-transfer experiments for the cysteine amide. Note that the resonances in the top left corner of Figure 1 are the diagonal resonances corresponding to the cys-NH and the gly-NH. Assignments of the NH resonances are obvious from their multiplet structures in a 1-D spectrum of sufficient resolution. The glutamic acid amide resonance is missing in Figure 1, because it is too exchange broadened to survive the sine bell apodization used in processing the data.

The off-diagonal resonances X and Y in Figure 1 indicate magnetization exchange between the cysteine and glycine amide protons of glutathione and the H_2O solvent, for they occur at the expected locations defined by the chemical shifts of these resonances. At present the contributions from chemical exchange and intermolecular cross relaxation have not bee separated, although such differentiation is possible in principle.[2,3] The location of off-diagonal resonance Z indicates that it arises from dipolar and/or spin–spin interactions between the glycine amide and α protons.[8] The resonances labeled W appear to be artifacts related to residual excitation of H_2O by the acquisition pulse. In conclusion, then, the study of amide proton exchange in aqueous solutions by the important 2-D NMR technique appears to be feasible, particularly if advantage can be taken of the more sophisticated modifications[1] of the Redfield H_2O suppression technique.

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Structure of Ristocetin A: Configurational Studies of the Peptide

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From studies of a number of examples[1] it is now apparent that the vancomycin-group glycopeptide antibiotics contain heptapeptides in which residues II and IV–VII (numbered from the N terminus) are similar or identical, whereas wide variations occur in residues I and III. In ristocetin A these residues are contained within ristomycinic acid (**1**), which has two substituted phenyl-

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