because OCH₃⁻5 in this case is located in similar environments as OCH₃⁻5 of IIIb and OCH⁻⁵' of 1b.

The above mode of linkage gains further support from mass spectral studies of IVb. The presence of m/e 311 as a major peak in IVb and IIIb, but very minor one in Vb, is of considerable significance. This may be attributed to facile carbon-carbon cleavage in IVb and IIIb due to steric reasons.

Lanthanide-induced shift studies by Eu(FOD)₃ have also been carried out to evaluate proton chemical shifts of IVb. S-values of every proton are listed in the Table. We have recently reported that H-6 of flavone nucleus on addition of Eu(FOD)₃ shows a considerable downfield shift (2.76 ~ 5.80 ppm in S-values) in comparison with H-8 or H-3 (less than 1.14 ppm) while the side phenyl protons are shifted to a very small extent. A singlet at 6.88 ppm assigned to H-8' gave a small S-value (0.50 ppm). A large S-value of OCH₃⁻5 (10.38 ppm) would mean that complexation occurs mostly at this side of the molecule. The S-value of H-2' (1.42 ppm) is larger than usual (~0.50 ~ 0.56 ppm) perhaps because the side phenyl group (at C-3) is attached to 6-position of the other flavone nucleus. These observations are in accord with the previous findings and are compatible with the structure of robustaflavone (Iva). Further, paramagnetic induced shift studies disentangled the signals of H-2' (7.81d) and H-6' (7.87q) which were found overlapping with a doublet of H-2'' (6'') (7.87d).

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The Synthesis of a Decapeptide with Glycosidase Activity

Copolymers of Gnu and hydrophobic amino acids have been shown to have substantial lysozyme-like and non-specific glycosidase activities. These copolymers were synthesized as they would have unionized and ionized carboxyl functions in their hydrophobic and hydrophilic regions respectively. Contact with a polysaccharide substrate was expected to lead to protonation of proximally placed glycosidic oxygen atoms by unionized carboxyl functions. Bond cleavage would then occur if the resulting carbonium ion could be stabilized by a suitably placed carboxylate anion.

The extension of this concept is the synthesis of a small peptide with suitably disposed carboxyl functions of which one should be in a hydrophobic environment and at least one in a hydrophilic environment. The design of such a peptide requires anticipation of its conformation. In the absence of adequate information on the conformation of peptide sequences in solution, we projected our synthesis to achieving conformational control in the solid state.

In an analysis of amino acid sequence whose three-dimensional structures have been determined by X-ray crystallography, Kotokluch and Scheraga have identified the α-helical and non-helical character of nearly 80% of the individual peptides in protein molecules. According to the rules formulated, the initiation of an α-helix requires 4 helix-making amino acids in a row so that the helix grows towards the C-terminal end unless interrupted by 2 helix-breaking amino acids in succession. If these characteristics are also manifest in smaller peptide sequences, the decapptide Glu-Phe-Ala-Ala-Glu-Glu-Ala-Ala-Ser-Phe (I) might be expected to have a tendency to form an α-helix as the only helix-breaking amino acid in the sequence is Ser-9. Further, if this peptide were to adopt an α-helix conformation, Glu-6 would have its carboxyl function in a hydrophobic environment as it would be flanked above and below by the benzene rings of Phe-2 and Phe-10 (Figure 1) and the adjacent Ala-7 methyl would also contribute to its hydrophobic environment. Gnu-5, on the other hand, would be in a hydrophilic environment and the carboxyl of this amino acid residue or of Gnu-1 with the carboxyl of Gnu-6 could provide the catalytic site of this enzyme model.

Decapptide I was synthesized conventionally. Z-Ser(Bu) was condensed with Phe-OBu in the presence of DCC to give Z-Ser(Bu)-Phe-OBu (II), crystallized from C₆H₆-C₂H₄, mp 105-106 °C, [c]D₂ --19.7 °, yield 76%. It was treated with H₂/Pd black to yield Glu(OBut) -Z-Ala-OBSu to obtain tripeptide Z-Ala-Ser(Bu)-Phe-OBu (III), crystallized from CH₂Cl₂-C₆H₄, mp 114-115 °C, [c]D₂ --19.7 °, yield 76%. Z-Glu(Obu) was condensed with Ala-OMe with the aid of DCC. The product Z-Glu(Obu)-Ala-OMe crystallized from CH₂Cl₂-C₆H₄, mp 105-106 °C, [c]D₂ --49.7 °, yield 85%, was treated with H₂/Pd black to yield Glu[Obu]-

The synthetic sample of IVb was obtained in a yield of 10 to 15% through Wessely-Moser rearrangement of hexa-O-methylubflavone (Vb) followed by methylation. Both the samples had the same m.p. 305-308 °C and showed no depression on admixture. RF values, fluorescence in UV-light, UV, IR and NMR spectral data of the 2 samples were also in good accordance. Judging from the RF value (TLC) it was deduced that robustaflavone was present in AgII and its monomethyl ether in AgIV.

Zusammenfassung. Isolierung und Strukturaufklärung eines neuen Typs der Biflavone aus Agathis robusta

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1 Communication No. 1722 from the Central Drug Research Institute constitutes synthetic substitute enzymes Pt. V, presented in part at the I.U.P.A.C. symposium on Natural Products, New Delhi, Feb. 1972.
2 Abbreviations in accordance with IUPAC-IUB Commission on Biochemical Nomenclature, Biochemistry 17, 1726 (1972).
7 Satisfactory C, H and N, analysis obtained for all peptides synthesized. [c]D₂ and [c]D₂ are reported for 10% solutions in MeOH at 25° and in DMF at 34° respectively.
Ala-OMe (IV). Condensation of IV with Z-Glu(OBu\textsuperscript{t})-ONSu yielded Z-Glu(OBu\textsuperscript{t})-Glu(OBu\textsuperscript{t})-Ala-OMe (V), crystallized from CH\textsubscript{2}Cl\textsubscript{2}-C\textsubscript{6}H\textsubscript{14}, mp 117–118\degree, (\(\alpha\))\textsubscript{D}\textsuperscript{a} = 54.5\degree, yield 69\%. V was treated with NH\textsubscript{3},NH\textsubscript{2},H\textsubscript{2}O (98\%) to yield Z-Glu(OBu\textsuperscript{t})-Glu(OBu\textsuperscript{t})-Ala-N\textsubscript{2}H\textsubscript{2} (VI), crystallized from MeOH-Et\textsubscript{2}O, mp 159–160\degree, (\(\alpha\))\textsubscript{D}\textsuperscript{b} = 5\degree, yield 70\%. III was decarbobenzoxylated with H\textsubscript{2}/Pd black and condensed with the azide from VI to yield the hexapeptide Z-Glu(OBu\textsuperscript{t})-Glu(OBu\textsuperscript{t})-Ala-Ala-Ser(Bu\textsuperscript{t})-Phe-OBu\textsuperscript{t} (VII), crystallized from EtOAc, mp 226–227\degree, (\(\alpha\))\textsubscript{D}\textsuperscript{b} = 6.3\degree, yield 60%.

Incorporation of \(^{3}\text{H}\) in I. HBr

<table>
<thead>
<tr>
<th>Tritiation time (h)</th>
<th>Counts (mg/5 min)</th>
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<tr>
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<tr>
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</tr>
<tr>
<td>12</td>
<td>2222</td>
</tr>
<tr>
<td>24</td>
<td></td>
</tr>
</tbody>
</table>

\textsuperscript{a} Tritiation carried out in presence of TFA

Z-Ala-Ala-OMe\textsuperscript{b} was decarbobenzoxylated with HBr/AcOH and the free base condensed with Z-Phe-ONSu to give Z-Phe-Ala-Ala-OMe (VIII), crystallized from EtOAc, mp 192–193\degree, (\(\alpha\))\textsubscript{D}\textsuperscript{a} = 23.8\degree, yield 82\%. VIII in turn was treated with HBr/AcOH and the free base condensed with Z-Glu(OBu\textsuperscript{t})-ONSu to give Z-Glu(OBu\textsuperscript{t})-Phe-Ala-Ala-OMe (IX), crystallized from EtOAc, mp 156–157\degree, (\(\alpha\))\textsubscript{D}\textsuperscript{a} = 31\degree, yield 72\%. IX was converted to Z-Glu(OBu\textsuperscript{t})-Phe-Ala-Ala-N\textsubscript{2}H\textsubscript{2} (X), crystallized from MeOH, mp 220–221\degree, (\(\alpha\))\textsubscript{D}\textsuperscript{b} = 20\degree, yield 85\%, by treatment with NH\textsubscript{3},NH\textsubscript{2},H\textsubscript{2}O (98\%).

VII was treated with H\textsubscript{2}/Pd black and the free base condensed with the azide from (X) to yield Z-Glu(OBu\textsuperscript{t})-Phe-Ala-Ala-Glu(OBu\textsuperscript{t})-Glu(OBu\textsuperscript{t})-Ala-Ala-Ser(Bu\textsuperscript{t})-Phe-OBu\textsuperscript{t} (XI), recrystallized from DMF, mp 249–250\degree dec., (\(\alpha\))\textsubscript{D}\textsuperscript{b} = 19.4\degree (C = 1, DMSO), yield 76\%. The protected decapptide XI was chromatographically homogeneous and after HCl hydrolysis, its amino acid composition was found to be 0.91 Ser, 3.03 Glu, 4 Ala, 1,95 Phe in an amino acid analyser. Treatment of XI with HBr/TFA yielded I.HBr, recrystallized from DMF-Et\textsubscript{2}O, mp 213–214\degree dec., (\(\alpha\))\textsubscript{D}\textsuperscript{a} = 59\degree (C = 1, CF\textsubscript{3}CO.CF\textsubscript{3}.3H\textsubscript{2}O).

Decapptide I was examined for its ability to hydrolyse chitin and dextran. For this purpose, it was converted into a trisodium salt and dispersed in 0.01 M phosphate buffers. Figure 2 shows the rate of release of reducing sugar, measured colorimetrically by the PARK and JOHNSON method, on treatment of a suspension of chitin in phosphate buffer with the decapptide and with hen egg-white lysozyme at pH 5.8 and 7.0. Decapptide I also hydrolysed dextran at about the same rate as it hydrolysed chitin. The observed activity of decapptide I is significant but of a low order. Ideal conditions for studying the enzymatic activity of such peptides have, however, still to be established.

The observation of glycosidase activity suggests that the decapptide has an \(\alpha\)-helical conformation. This view is supported by the IR-spectra of the compound. N-H stretching is at 3270 cm\textsuperscript{-1} both in KBr and in dimethyl formamide solution and suggests that only intramolecular

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