s1.m5.p7 The structure of Trichoderma reesei hydrophobin HFBII. Johanna Hakanpaa,^a Markus Linder^b and Juha Rouvinen^a, ^aDepartment of Chemistry, University of Joensuu PO BOX 111 80101 Joensuu, Finland, and ^bVTT Biotechnology, PO BOX 1500 02044 VTT, Finland. E-mail: juha.rouvinen@joensuu.fi

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Hydrophobins are small proteins, about 100 amino acid residues in size, secreted by filamentous fungi. They are characterized by eight conserved cysteine residues in the sequence and their ability to change the character of a surface by spontaneous self-assembly on a hydrophobic-hydrophilic interface. Hydrophobins are also among the most surface-active biomolecules known. The biological properties of hydrophobins have been studied widely but the molecular bases of their function has remained largely unknown in lack of a three-dimensional structural model.

We have determined the crystal structure of *Trichoderma reesei* hydrophobin HFBII to an atomic resolution of 1.0 Å [1], [2]. The structure is novel, containing four antiparallel b-stands and an a-helix. The β -stands form a small barrel, inside which two of the four disulfide bridges are located. The remaining bridges connect the N-terminal loop and the α -helix to the β -barrel. A flat, hydrophobic patch is found on the surface of the protein giving rise to the amphiphilic nature of the molecule. In the crystal structure, two hydrophobin molecules pack together, partly concealing the hydrophobic patches in between them. This makes HFBII, in spite of its name, quite water-soluble.

Solving the structure of hydrophobin HFBII has changed conceptions about these proteins dramatically. Hydrophobins were stated to be largely unstructured in solution and thought to function through large conformational changes, which seems unlikely in the light of the determined structure, which is quite compact. Also the assumption of the cysteines forming disulfide bridges consecutively was found incorrect. These findings underline the importance of experimental structure determination.

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s1.m5.p8 The Structure of 2 EF-hand Calcium Binding Allergen Che a 3. <u>Walter Keller</u>,^{*a*} Petra Verdino,^{*ab*} Rodrigo Barderas,^{*c*} Rosalia Rodriguez,^{*c*} Kerstin Westritschnig^{*d*} and Rudolf Valenta^{*d*}, ^{*a*}Institute of Chemistry / Structural Biology, Karl-Franzens-University Graz, Austria, ^{*b*}The Scripps Research Institute, Department of Molecular Biology, La Jolla, USA, ^{*c*}Department of Biochemistry and Molecular Biology, Faculty of Chemistry, Complutense University, Madrid, Spain, and ^{*d*}Department of Pathophysiology, Molecular Immunopathology Group, Medical University Vienna, Austria. *E-mail: Walter.keller@uni-graz.at*

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Calcium-binding proteins are ubiquitous in living organisms, taking part in a variety of cellular processes e.g. muscle contraction, cell cycle control, signal transduction and differentiation. In general, calcium binding proteins can be divided in two categories: calcium buffer proteins and calcium sensors, depending on their ability to either interact with a ligand in a calcium dependent manner or to just bind calcium and thereby regulating the intracellular calcium level. The most prevalent calcium binding motif is the EF-hand motif, which generally occurs in intramolecular EF-hand pairs.

Recently a novel family of calcium binding allergens were characterized [1], which exhibited two EF-hand motifs in there sequence and showed specific IgE binding in a calcium dependent manner. Members of this 2EF-hand protein family have been discovered in the pollen of various grasses, weeds and trees, exhibiting a strong cross-reactivity between each other, but not with other prevalent EF-hand proteins like Calmodulin, Calbindin or Parvalbumin.

We have solved the 3D-structure of two members of this pollen allergen family, Phl p 7 [2] and Che a 3, with X-ray crystallographic methods. In this paper we will discuss the general structure of the allergens and compare the surface properties, which are responsible for the specific recognition by IgE antibodies.

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