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976 Profilins, Calcium-Binding Proteins, and Carbohydrate Crossreacting Determinants in 23 Different Pollen Species*Bianca Barletta**Raffaella Tinghino**Patrizia Iacovacci**Claudia Afferni**Cinzia Butteroni**Gabriella Di Felice**Carlo Pini**Adriano Mari**Department of Immunology, Istituto Superiore di Sanità, Rome, Italy*

Profilins (PRF) and pollen-derived calcium binding proteins (CBP) are the best characterized panallergens in terms of their structure and IgE reactivity. Both the panallergens have been identified in pollen from some of the most common allergenic families. Carbohydrate crossreacting determinants (CCD) are widely present IgE-reacting epitopes, accounting for the crossreactivity between molecules with unrelated protein sequence, and seem to have no biological activity. To further define the distribution of PRF, CBP and CCD we used pooled sera with a previously defined strong IgE reactivity to the three components under study and to allergenic molecules used as controls. rBet v 2 (PRF from birch), rJun o 2 (CBP from juniper) and bromelain (a glycoprotein from pineapple) were used as antigens in an ELISA inhibition assay. rPhl p 5 and rBet v 1 were used as controls. Inhibitors were represented by the following 23 pollen species selected among (1) very common allergenic species (rye grass, juniper/cypress, olive, pellitory), (2) less common allergenic species (elm, goosefoot, hazel, lime, maple, mugwort, oak, pine, plantain, poplar, sycamore), (3) rare species or species never described as allergenic (birch, eucalyptus, locust tree, mustard, palm, ragweed, salt cedar, tree of heaven). Fifty-percent inhibition value was calculated by graphical interpolation. Sixteen out of 23 species reached the 50% inhibition of the binding to the rBet v 2 with inhibitor concentrations ranging between 0.2 and 20 µg protein/ml. The remaining (eucalyptus, juniper/cypress, lime, maple, mugwort, mustard and ragweed) were incapable to inhibit the binding at the highest concentration used (200 µg protein/ml). Conversely, all the species inhibited the sIgE binding to the rJun o 2. Fifty-percent inhibition was obtained with concentrations of the inhibitor

ranging from 0.005 µg protein/ml of the rye grass extract to of 200 µg protein/ml of the elm extract. CCDs were present in 21 out of 23 extracts tested and 50 % inhibition was reached with a protein concentration of the inhibitors ranging from 1.7 to 200 µg/ml. As expected, the IgE binding to rPhl p5 was inhibited only by the rye grass extract, whereas the IgE binding to rBet v 1 was inhibited by birch, hazel and oak pollen extracts.

The wide distribution of panallergens (PRF and CBP) among common and rare pollen species supports the hypothesis that the patients sensitized to these molecules have a broad spectrum of pollen reactivity. The presence of CCD in almost all pollen raises the problem of the possible identification of crossreactive glycoproteins in extracts without clinically relevant allergenic components. The lack of inhibition by some extracts obtained from pollen already known to contain profilins (mugwort and ragweed) raises the question of whether the allergenic extracts need to be further characterized in terms of “minor” components.