

## **Specificity of Jacalin Binding**

Jacalin is a lectin present in the seeds of the Jackfruit, *Artocarpus integrifolia*. Recent papers <sup>1,2</sup> reported that Jacalin specifically bound human secretory IgA and that this lectin could be used to separate human IgA from other serum glycoproteins, including other immunoglobulin classes, and that agarose bound Jacalin could be used to separate IgA<sub>1</sub> and IgA<sub>2</sub>.

We have recently affinity purified Jacalin. Our preliminary investigation shows it to have a molecular weight of approximately 60,000 with a rather complex subunit structure. In addition to confirming its affinity toward galactose, we have examined the specificity of this lectin toward several glycoproteins. Using glycoproteins of known carbohydrate structure, we can now provide some information on the structural requirements of the oligosaccharide for binding Jacalin.

We first determined that fetuin, a glycoprotein present in fetal bovine serum, would bind to Jacalin covalently attached to agarose. Fetuin, however, contains two types of oligosaccharides, those linked N-glycosidically to asparagine and those attached by O-glycosidic linkage to serine or threonine. Another glycoprotein, which contains an N-glycosidically linked oligosaccharide similar to fetuin is alpha1 acid glycoprotein. However, neither native alpha1 acid glycoprotein nor the desialylated form bound to Jacalin. This indicated that Jacalin does not bind galactosyl ( $\beta$  1-3,4) N-acetylglucosamine, the terminating sequence in the oligosaccharide of alpha1 acid glycoprotein. The O-glycosidically linked oligosaccharide in fetuin consists of galactose ( $\beta$  1-3) N-acetylgalacto-samine, with sialic acid substitutions on the 3 position of galactose and the 6 position of N-acetylgalactosamine. To determine if sialic acid was required for binding we tested asialofetuin.

The desialylated form of fetuin bound equally well, showing that binding was not dependent on the presence of the sialic acid residues. Removal of galactose from the N-acetylgalactosamine residue of fetuin reduced but did not abolish the ability to bind to Jacalin, indicating the N-acetylgalactosamine is an important part of the carbohydrate structure required for binding. Studies are underway to further characterize these structural requirements.

Jacalin apparently does not bind to galactosyl-N-acetylglucosamine but recognizes galactose ( $\beta$  1-3) N-acetylgalactosamine, a structure identical to the receptor for peanut agglutinin and sometimes referred to as the Thomsen-Friedenreich antigen or "T antigen". However, peanut agglutinin required this O-linked oligosaccharide to be devoid of sialic acid, whereas Jacalin will bind to the fully sialylated disaccharide. In addition to its potential value in the isolation of IgA, Jacalin, in combination with peanut agglutinin, may provide important information on glycoproteins containing O-linked oligosaccharides or provide an easy method to fractionate these glycoconjugates.

- 1. Roque-Barreira, M.C. and Campos-Neto, A., J. Immunol. <u>134</u>, 1740 (1985)
- 2. Gregory, R.L., Rundegren, J and Arnold, R.R., J. Immunol. Meth., <u>99</u>, 101 (1987)

Additional references available upon request.