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Introduction

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Glycobiology is the study of the biological significance of the complex carbohydrate moieties, glycans, attached to proteins or lipids. Until quite recently the functions of glycans in biology received less attention than those of nucleic acids or proteins. In part, this situation was due to the difficulties of structural analysis of often large, branched oligosaccharides. Typically, glycans contain five or more monosaccharide constituents, joined in a wide variety of glycosidic linkages. Fortunately, in recent years the techniques of glycan analysis have improved greatly, both in speed and sensitivity, producing a wealth of structural information. In parallel to these advances major pathways of assembly and degradation of the glycans, carried out by a host of highly specific glycosidases and glycosyltransferases, have been elucidated biochemically and genetically. Glycans are produced by the sequential action of multiple glycosyltransferases. The rule of 'one enzyme-one glycosidic linkage' proposed many years ago appears to have been upheld: each transferase forms a specific linkage between one monosaccharide, transferred from a specific sugar nucleotide, to a preferred oligosaccharide acceptor. The specificity of glycosyltransferases, especially for acceptor substrate, provides a key regulatory influence on glycan structures. Thus, the diversity of glycan structures, although large, is nevertheless finite, leading to the belief that such structures contain information that can be read by cognate proteins such as antibodies and lectins. Concurrently, progress in chemical and enzymatic synthesis of defined oligosaccharides has provided reagents to characterise the specificity of enzymes involved in glycan assembly and degradation, to isolate and define the specificities of lectins and to study protein-carbohydrate and carbohydrate-carbohydrate interactions of biological significance.

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Glycobiologists now know unequivocally that glycan structures are characteristic of cell or tissue type, as governed by the cell- or tissue-type-specific expression of multiple glycosyltansferases. In higher organisms glycan structures often characterise different embryological stages and the appearance of different cell lineages. Glycan appendages often participate in the fine tuning of fundamental processes mediated by proteins in cell adhesion, motility and growth, and in specific interactions with different proteins, such as growth factors and pathogens. Direct evidence for the role of glycans is also being obtained by modulating glycosylation pathways in cells or in genetically manipulated transgenic animals. Mutations in genes involved in glycan assembly and degradation are increasingly being correlated with important human diseases. This impressive growth of interest justifies the present special issue on developmental and medical glycobiology. The following reviews will focus on some of the recent exciting advances in understanding the role of glycans in development, the association of specific glycosylation patterns with disease and the potential of this emerging knowledge in devising novel therapeutic approaches in the treatment of genetic and infective diseases.

Traditionally, glycans attached to proteins are classified as either N-linked, containing an *N*-acetylglucosamine (GlcNAc)-asparagine linkage or O-linked, containing an *N*-acetylgalactosamine (GalNAc)-serine or threonine linkage. Although early studies suggested an intrinsic glycosylating potential in intracellular organelles such as mitochondria or nuclei, the prevailing consensus was that *N*- and *O*-glycan assembly occurred exclusively within the classical secretory pathway involving the endoplasmic reticulum and Golgi compartments. This view is now outdated by the discovery of glycosylation reactions occurring within the cytoplasm. One of these involves the addition of *O*-βGlcNAc to certain cytoplasmic and nuclear

proteins. A key feature of *O*-GlcNAc modification is its extreme dynamism, analogous to protein phosphorylation. In their review Wells, Vosseller and Hart discuss the extensive evidence relating *O*-GlcNAc modification to nutrient-sensing pathways, and the correlation of elevated *O*-GlcNAc of key proteins to downstream effects on insulin-stimulated glucose uptake into cells in diabetes. Another form of cytoplasmic glycosylation, reviewed by West, differs in several respects from the above: probably α- (rather than β-) linkage of GlcNAc to protein hydroxyproline, relative metabolic stability of the substituents and extension of the GlcNAc residues by other monosaccharides. This type of cytoplasmic glycosylation was discovered in the lower eukaryote *Dictyostelium,* specifically on the Skp1 subunit of the SCF-E3 ubiquitin ligase complex, where it may be required for nuclear targeting of Skp1. However, similar glycosylation may occur more widely in eukaryotes, perhaps modifying proteins additional to the highly conserved Skp1 subunit. The potential role of Skp1 glycosylation in regulation of the ubiquitin-proteasome pathway generally raises fascinating new questions concerning its possible involvement in integrin-mediated cell cycle control, recently discovered to involve regulated degradation of the cyclin kinase inhibitor p27 [1].

It has been known for a long time that proteins proceeding through the secretory pathway may be modified by unusual glycopeptide linkages other than those of the classical *N*- and *O*-glycans [2]. Until recently these 'minor' modifications have been literature curiosities but now have shifted centre stage. Shao and Haltiwanger review the *O*-fucose modification of selected serine and threonine residues present in certain protein modules, for example the epidermal growth factor-like repeats of Notch protein. The Notch family of receptors play a central role in signalling pathways that regulate numerous cell fate decisions in development. Interactions of the Notch receptors with ligands is regulated by further glycosylation of protein-bound fucose, initiated by Fringe family *N*-acetylglucosaminyltransferases. Recent data indicate that *O*-fucosylation is required for signalling of the transforming growth factor-β family member Nodal mediated by the epidermal growth factor family-related protein Cripto-1, essential for formation of mesoderm during gastrulation [3]. In their review Hewitt and Grewal describe the association of altered glycosylation of α-dystroglycan with muscular dystrophies. The best characterised of these mutations affects the synthesis of *O*-mannosyl glycans of dystroglycan. Failure to assemble a wild-type *O*-mannoglycan disrupts the dystroglycan-associated complex which joins laminin in the extracellular matrix to dystrophin and associated proteins within the sarcolemmal cytoplasm.

The diversity of glycan structures is expanded considerably when organisms such as the protozoan parasite *Leishmania* are considered. Novel linkages, involving uncommon sugars and cyclic variants such as galactofuranose, are encountered in the glycoconjugates of these organisms. Pederson and Turco show how these novel glycans are important virulence factors in leishmaniasis, a devastating disease worldwide. Apart from being a headache for structural glycobiologists, these unusual glycans offer real opportunities for development of specific and effective therapies.

The final four reviews of this issue deal with lectins, proteins that recognise the structural information encoded within the diverse glycans of glycoproteins and glycolipids. Galectins are a family of β-galactoside-binding proteins expressed widely in the cytoplasm of many cell types. Although some galectins are secreted and play extracellular roles, their presence in the cytoplasm implies additional intracellular functions. Yang and Liu review the roles of galectins in cell growth and death. Some of these roles require interactions of galectins with intracellular receptors and perhaps nuclear targeting. Although protein-protein interactions are involved in some cases, for example interaction between galectin-3 and the tumour supressor Bcl-2 involved in inhibition of apoptosis, the possibility of interaction of galectins with proteins decorated with the glycans discovered by West is intriguing. In their beautifully illustrated review Bostos and Wlodawar describe a bacterial lectin called cyanovirin-N. This binds tightly to oligomannose glycans of the gp140 envelope glycoprotein of the human immunodeficiency virus (HIV) and efficiently blocks viral infectivity, raising hope for a new anti-HIV strategy. Interestingly, a lectin of very similar specificity called DC-SIGN is expressed at the surface of dendritic cells and appears to potentiate viral presentation to CD4-positive cells [4], a function that may be a specific target for inhibition by cyanovirin-N and related antiviral reagents. Roche, Fajac, Grosse, Frison, Rondanino, Mayer and Monsigny describe how the uptake of plasmid DNAs into specific cells may be induced by wrapping the DNA into a complex with synthetic glycopolymers. These complexes may be recognised by endocytic endogenous lectins expressed at the surface of the cells. As with all forms of nonviral gene therapy, many problems remain to work out, including the efficiency of plasmid uptake compared with viral vectors, and gene stability. However, the results are certainly encouraging and may become adaptable to diseases such as cystic fibrosis. Finally, Diekman discusses protein-carbohydrate interactions that play important roles in sperm maturation and gamete recognition. Autoantibodies directed against some of these components appear to be factors in human infertility: these same factors may offer novel targets for development of new contraceptives.

In summary, the reviews collected here admirably illustrate the impressive breadth of modern research in glycobiology, and how this relatively young discipline impacts significantly on large areas of biomedical research.

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