DECIPHERING *IN VIVO* GLYCOBIOLOGY WITH GENETICALLY-ENCODED, MULTIVALENT LIQUID GLYCAN ARRAY (LiGA) AND LIQUID LECTIN ARRAY (LiLA)

Ratmir Derda

Department of Chemistry, University of Alberta, Canada
ratmir@ualberta.ca

A major barrier to studying the role of glycans *in vivo* is the fundamental lack of one-to-one correspondence between sequence of DNA and structures of carbohydrates. Investigations of carbohydrates, thus, cannot rely on DNA sequencing directly. To solve this challenge, we introduce genetically-encoded platform technologies termed Liquid Glycan Array (LiGA) [1-4] and Liquid Lectin Array (LiLA) [5-6]. Glycan arrays, made by ligation of carbohydrates to glass or bead surfaces and complementary array of lectins on glass slides are the workhorse tools in glycobiology. These technologies cannot measure interactions of glycans and GBPs in their natural environment on the surface of cells in tissues *in vivo*. In contrast, Liquid arrays introduce one-to-one correspondence between DNA sequence and carbohydrate structure of glycan or glycan binding protein displayed on phage. They enable unsupervised profiling of interactions of glycans with receptors on the surface of cells *ex vivo* and *in vivo*.
LiGA is produced by chemical [1] and chemoenzymatic ligation [2] of carbohydrates to bacteriophage (phage) particles. Genetically encoded, monodisperse carriers based on 700 mm long M13 phage particles display 50-1000 copes of glycans on their surface. The identity and presentation (density) of glycans are encoded by the DNA barcode inside the phage genome. LiLA in turn is produced by enzymatic ligation of lectins to bacteriophage using SpyCatcher-SpyTag technology [5-6]. LiGA and LiLA uncovered an optimal structure/density combination for recognition for a wide collection of lectins and immune lectin proteins (DC-SIGN and Siglec family proteins) expressed on live cells [1,2,4,5]. LiLA can detect presence of specific glycoisoforms on live cells. Injection of the LiGA or LiLA into mice identified glycan:GBP interactions necessary for homing to specific organs. This work provides an unprecedented quantitative evaluation of the interaction of complex glycans with GBPs *in vitro* and *in vivo*.

References:
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