# Mathematical Modelling of Monoclonal Antibody Glycosylation Process in Mammalian Cell

Sha Sha; Dr. Seongkyu Yoon*
University of Massachusettes Lowell

## ABSTRACT

Tremendous efforts have been put into understanding and control of glycosylation reaction pathways so as to result in uniform glycan sturctures in biopharmaceutical products. Mathematical modeling approaches are a paradigm recently emerging in research area striving toward this goal. The current state of the art shows several types of mathematical tools being incorporated. One is the development of reaction network models which aims to describe simplified but the essence of glycosylation mechanism. This method aims to accurately simulate the glycosylation process in mamalian cells, and provide a capability to observe glycosylation output deviation under varied culture conditions. Another method is statistical data analysis method. This method queries the experimental data and illustrate the patterns and correlations inherent in the datasets..

## CONTACT

Sha Sha
University of Massachusettes Lowell
Email: Sha_Sha@student.uml.edu
Phone: 978-761-4830

## INTRODUCTION

Monoclonal antibody (MAb), as one large portion of biopharmaceutical products, is significantly affected of its physiological and clinical properties by the accompanied *N*-linked glycan structures. In pharmaceutical manufacturing, glycan structures of produced MAb are subject to a high degree of heterogeneity. However, due to complex pathways existing in the cell glycosylation process, limited strategies have been achieved to understand and control these glycosylation pathways.

Instead of traditional experimental methodology, mathematical modelling approaches, as a paradigm in system biology realm, are emerging as powerful tools complementary for better understanding and control of mammalian cell glycosylation.

**Two types of mathematical tools are currently in the center of our attention:**

(1) Reaction network modelling of antibody glycosylation process, which depicts the underlying pathways of glycosylation.

(2) Data-derived modelling which tends to make best utilization of large scale experimental data from manufacturing process by extracting valuable information latent in datasets.

## METHODS AND MATERIALS

◆Network model formulation methods

Chemical network models will ususlly be built based on a couple of DOEs. Cell compartments like ER and Golgi where glycosylation occure will be regarded as certain type of chemical reactors. DOE equations are fomulated based on glycosylation network and according to equillibrium status of all components considered in the modelled comparments.

◆Statistical Analysis methods

The statistical analysis methods applied for experimental-derived and glycosylation-related data include hierarchical clustering, principal component analyasis and partial least-squares methods.

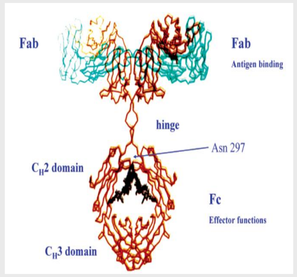


**Figure 1.** Antibody structure

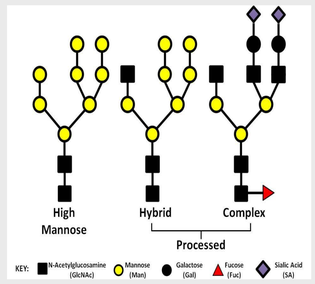


**Figure 2.** Example of glycan structures

## RESULTS

As one example of developing data-derived models, one attempt was given to a set of data from a published paper using multivariate statistic analysis tools to uncover the correlation between a couple of cell culture factors (like glutamine supplements, metabolites accumulation in media) and cell produced antibody glycan profile. Statistical analysis (Fig.3) well classifies glycan sturcture into two groups, distinctive with the glycosylation level. The latent variables for this model might be certain significant causes for glycosylation level variation, such as glycan donor availablilty.

The correlation between measured factors and glycan outputs is revealed by the clusterring pattern of red spots (factors) and green spots (glycan structures). Some factors, especially nucleotide sugars, are shown to be highly correlated with glycosylation level.

As an agreement with the observation from the above statistical analysis, we are taking nutrients supplement (especially Glutamine as an imporant source of sugars) as one important paramter in reaction network model intiation. A network model is currently under out work, as illustrated in Fig.4, to achieve glycosylation process outcome as a function of given inputs parameters like cell cuture conditions and glycosylation-related enzyme kinetic parameters.

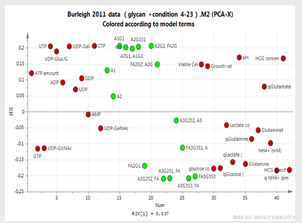


**Figure 3.** Scattering of cell culture factors and glycan structure outputs

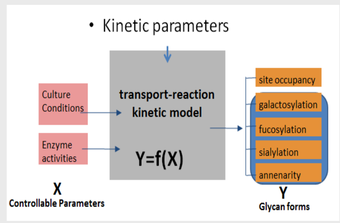


**Figure 4.** Scheme for glycosylation reaction network modelling

## DISCUSSION

As produced by “dry lab”, mathematical models are always nessitated to be validated and evaluated of their performance. Accuracy and robustness should be examined with models adequately.

It is also the essence of system biology study that wet-lab (experimental) should be combined to test the hypothesis emerging from the established models. It is usually a procedure of iterative refinement of model structure and parameters using computation and experimentation that brings models to perfection.

## CONCLUSIONS

Mathematical modelling serves as a tool embracing unique advantage that studies glycosylation process as a system instead of conventional reductionist approach of experimentation which studies individual proteins and moleclar intereations one-at-a-time. As a whole, the prospective functions out of those models will be:

**- prediction of glycan profile with cell culture condition and cell lines**

**- manufacturing online monitoring of mAb glycan profile**

**- post-simulation analysis to generate hypothesis for experimentation and direction for glycoengineering**

## REFERENCES

1. Burleigh, S. C.; van de Laar, T.; Stroop, C. J.; van Grunsven, W. M.; O'Donoghue, N.; Rudd, P. M.; Davey, G. P., Synergizing metabolic flux analysis and nucleotide sugar metabolism to understand the control of glycosylation of recombinant protein in CHO cells. *BMC biotechnology* **2011,** *11*, 95.

2. Butler, M. Optimization of the cellular metabolism of glycosylation for recombinant proteins produced by mammalian cell system. *Cytotechnology* **2006,** 50: 57-76.

3. Puri, A.; Neelamegham, S., Understanding glycomechanics using mathematical modeling: a review of current approaches to simulate cellular glycosylation reaction networks. *Annals of biomedical engineering* **2012,** *40* (4), 816-27.