299f Effect of Culture Conditions on the Glycosylation of Human Secreted Alkaline Phosphatase (Seap) Expressed in Tobacco Nt1 Cell Suspension Cultures

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Plants have recently emerged as an alternative system for the production of recombinant proteins and currently there is commercial production of a handful of industrial enzymes and some therapeutic proteins are now in clinical trials. Plant cell cultures have intrinsic benefits over whole plants because they can be maintained in a bioreactor under more controlled and reproducible conditions than in an agricultural environment and can serve as models of whole plant systems. Plant cells can also fold and assemble complex proteins correctly and perform post-translational modifications, which may be required for some proteins. Among these post-translational modifications, N-linked glycosylation is of particular interest for the case of therapeutic proteins. The differences between mammalian and plant glycans need to be addressed because non-mammalian glycans may affect the pharmacokinetic characteristics of proteins and two oligosaccharide residues added onto glycoproteins by plants are regarded as potential immunogens to humans. In this study, human secreted alkaline phosphatase (SEAP) was expressed in batch cultures of tobacco NT1 cell suspensions as a model system. SEAP is only glycosylated in one of its two putative N-glycosylation sites. In this work we will discuss the glycosylation profile of SEAP produced by cells under regular culture conditions and the effect of extracellular additives such as salicylic acid and ammonia on the glycoforms found in this model glycoprotein.