Mechanistic extension of a hydrophobic interaction chromatography model to account for pH changes and mixed-mode binding

INTRODUCTION
Hydrophobic interaction chromatography (HIC) is one of the common separation methods for the purifica-
tion of biomolecules. It is especially powerful as a polishing step for monoclonal antibodies to remove aggregates and impurities such as host cell proteins. The separation principle is based on the irreversible interaction between hydrophobic moieties on proteins and the adsorption ligands in the hydrophobic ligands of the stationary phase. Adsorption, elution, and desorption of the proteins are demonstrated by bind-and-elute ex-
periments in column format and calculation of the confi-
nement between well-ordered and like ordered water molecules as well as identifiability of isotherm parameters in Tables 1 and 2 (Karlsruhe Institute of Technology (KIT), Karlsruhe, Germany).

MODEL EXPERIMENTS
The result of the batch chromatogra-
phy experiments for glucose oxidase, BSA, lysozyme, and ten other protein species. Only L_n and

BATCH EXPERIMENTS
The result of the batch chromatography experiments for glucose oxidase, BSA, lysozyme, and ten other protein species. Only L_n and c_m were adjusted to account for the difference in elution capacity and volume basis.

MODEL COMPARISON
To compare the performance of the presented model with previous reported isotherms for HIC, the binding characteristics of the HIC model parameter were used to calibrate the model (Deitcher et al., 2010). The obtained parameters are shown in Table 1.

PH EXTENSION
Modeling pH-dependence is a necessity for practical development of industrial HIC pro-
cesses. Biological and biomolecular structures and their adsorption behavior of lysozyme, hem-
oglobin, and a single parameter were used to simulate pH-dependence of the model. The model was used for protein con-
centration over time. The challenge for practical model-based processes is to match the observed peak shapes. The newly constructed model proved good results with a much simpler model and, thus, less parameters.

MIXED MODE EXTENSION
We combined our HIC model with the Stolz Max Action (SMA) iso-
dispersing hydrophilic interaction model similar to Hilt et al. (2020). A case study with the glucose oxi-
dase investigated identifiability of the parameters on the model performance here (GE Healthcare) and compared the model's ability to match the observed peak shapes. The newly constructed model provided good results with a much simpler model and, thus, less parameters.

STOP EXPERIMENTING GO SILICO
Tobias Hahn1, Gang Wang2, Theresa Lauffer1, Manuela Catala Perlavaz2, Thiemo Huuk1, Jürgen Huubuch4
1 GoSilico GmbH, Karlsruhe, Germany
2 Karlsruhe Institute of Technology (KIT), Karlsruhe, Germany

1. The approach by Wang, Hilt, and Huubuch (2019) presents an equi-
lar model that can be tuned to any partic-
larly identical isotherm parameter set
2. The result of the batch chromatogra-
phy experiments for glucose oxidase, BSA, lysozyme, and ten other protein species. Only L_n and c_m were adjusted to account for the difference in elution capacity and volume basis.
3. The mixed model model by Hilt et al. required a complex pore model to obtain the fits shown in Figure 8.

GoSilico is a start-up company from Karlsruhe, Germany, that develops software and hardware for accelerated bioprocess development.

www.go-silico.com