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COMPARISON OF COMMERCIAL CHO MEDIA FORMULATIONS USING MATERIAL-ORIENTED RECURRENT SPECTRAL LIBRARIES

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Chinese hamster ovary (CHO) cells are commonly used for the production of biological therapeutics. Metabolic profiles of media components can be used to monitor process variability and look for markers that discriminate between batches of product. Currently, there exists no database of CHO media components or a method for the comparison of commercial media formulations. We are creating material-oriented libraries of CHO media components that include LC-MS/MS and GC-MS data. The libraries represent all metabolites that can be detected by LC-MS/MS and GC-MS and consist of recurrent spectra that cover all known fragmentation conditions and precursors. Recurrent spectra occur repeatedly in the sample and are processed to produce high-quality consensus spectra for the library. These spectra represent all detectable metabolites, both known and unknown. In addition, we are developing tools and methods for the profiling of complex biological materials that result in compound identifications. Current profiling studies largely rely on molecular feature extraction for identifications. Especially in LC-MS, multiple molecular features can result from a single compound due to the formation of in-source fragmentation ions, adducts, multiple charge states, etc. Commercial CHO cell media formulations were precipitated with 80% methanol, evaporated to dryness under a nitrogen stream and resuspended in pure methanol to remove salts. The samples were then analyzed by LC-MS/MS and GC-MS. LC-MS/MS and GC-MS data were searched against the NIST Libraries (NIST14/2014/EPA and MS/MS for GC and LC, respectively). Following searching to generate identifications, spectra were clustered and annotated using in-house developed software. Consensus spectra were then made from the spectra and further annotated with additional identifications for the spectral libraries.