serial flow. In a larger scale biopharmaceutical manufacturing operation, these complexities can decrease process robustness, cleaning efficiency, and limit adoption of single-use technologies. For example, configuration of a PCC process within a single unit operation leads to inefficient utilization of capital equipment and a loss of flexibility compared to traditional single column chromatography unit operations, especially for the adoption of equipment enabling single-use flowpaths.

[0006] Fault tolerance is also an issue for connected multi-column systems, and most particularly where two or more unit operations have been interconnected and contained within a single skid. The interconnection of these components and flow paths can have a significant impact on unit operations and the manufacturing process as a whole. When any component of this interconnected system is impacted, the entire unit operation(s) can be brought to a stop and the entire run up to that point can be lost.

[0007] The invention described herein overcomes these deficiencies by defining a mode of operation whereby the equipment and software configuration is smaller scale, platform-friendly, and modularized, the units operated in parallel rather than in a serial configuration, resulting in a system that is flexible, expandable, operated by external control, and not requiring complex set up and flow of material. Additionally, the described system displays superior fault tolerance compared to multi-column serial units, in that if a single unit is unable to function, only the single column is impacted and can be easily isolated and extracted, without the entire run being lost and/or brought to a stop.

Summary

[0008] In accordance with an example, a continuous parallel chromatography system is described herein that includes a plurality of chromatography column skids, where each chromatography column skid is suitable for independent batch processing. Each chromatography column skid includes a chromatography column, an inlet selectively coupled to an upstream source, at least one pump, at least one filter, and an outlet sensor. The system further includes a control circuit configured to control the operation of the plurality of chromatography column skids through one or more full cycles of a chromatography process, where each full cycle includes a long step and a plurality of shorter steps, each of the plurality of shorter steps having a processing time shorter than or equal to a processing time of the long step. The control circuit is configured to receive a synchronization signal associated with a first one of the plurality of chromatography column skids that the long step is complete and, in response to reception of the synchronization signal, direct operation of a second one of the plurality of chromatography column skids to commence operation of the long step.

[0009] In some forms, the control circuit can be configured to direct operation of the plurality of shorter steps on the first one of the plurality of chromatography column skids to complete the

the first downstream chromatography column skid that the long elution step is complete and, in response to reception of the synchronization signal, direct operation of the second chromatography column skid to commence operation of the long elution step.

[0014] In accordance with an aspect, a method for purification of a recombinant protein from one or more contaminants is described that includes subjecting the protein to an affinity chromatography unit operation, subjecting the elution pool from the affinity chromatography unit operation to low pH viral inactivation and neutralization, and subjecting the neutralized pool to one or more polishing chromatography unit operations. At least one of the affinity chromatography unit operation or at least one of the one or more polish chromatography unit operations are operated according to a parallel chromatography process configured to control the operation of a plurality of chromatography column skids through one or more full cycles of a chromatography process, where each full cycle includes a long step and a plurality of shorter steps and each of the plurality of shorter steps has a processing time shorter than or equal to a processing time of the long step. The parallel chromatography process includes the steps of: receiving a synchronization signal associated with a first one of the plurality of chromatography column skids that the long step is complete at a control circuit of a parallel chromatography system and in response to reception of the synchronization signal, directing operation with the control circuit of a second one of the plurality of chromatography column skids to commence operation of the long step.

[0015] In some forms, the affinity chromatography unit operation is performed according to the parallel chromatography process and, in further forms, the long step can be loading a column of the chromatography column skids and the plurality of shorter steps can be two or more of: an equilibration step, one or more washing steps, an elution step, a regeneration step, or a flushing step. In other forms, at least one of the one or more polish chromatography unit operations can be performed according to the parallel chromatography process and, in further forms, the long step can be eluting a column of the chromatography column skids and the plurality of shorter steps can be two or more of: an equilibration step, a loading step, one or more washing steps, a regeneration step, and a flushing step. In yet further forms, both the affinity chromatography unit operation and at least one of the one or more polish chromatography unit operations can be performed according to the parallel chromatography process.

[0016] In some forms, the parallel chromatography process can include the step of directing operation of the plurality of shorter steps on the first one of the plurality of chromatography column skids with the control circuit to complete the full cycle of the chromatography process therefor after reception of the synchronization signal and, in further forms, the parallel chromatography process can include the steps of: receiving a second synchronization signal associated with the second one of the plurality of chromatography column skids at the control

circuit that the long step is complete and, in response to reception of the second synchronization signal, directing operation of the first one of the plurality of chromatography column skids with the control circuit to commence a second operation of the long step.

[0017] In some forms, the method can include one or more of the following: at least one of the one or more polish chromatography unit operations can include first and second polish chromatography unit operations with a first capture chromatography column connected in series with a second capture chromatography column; the one or more polish chromatography unit operations can include separate first and second polish chromatography unit operations; the first polish chromatography unit operation precedes or follows the second polish chromatography unit operation; the virus inactivated elution pool can be neutralized using a neutralizing buffer system capable of minimizing volume expansion of a viral inactivated eluate pool while maintaining conductivity less than or equal to 10 ms/cm, wherein the buffer system includes a titrant that does not have buffer capacity in the target pH range and a buffering agent which buffers at a desired pH; the viral inactivation can be 30 minutes or more; the method can include subjecting the neutralized pool to a depth filtration operation; the method can include subjecting an elution of the one or more polish chromatography unit operations to one or more of: an ultrafiltration/diafiltration unit operation or a virus filtration unit operation; the affinity chromatography unit operation can be Protein A chromatography, Protein G chromatography, or Protein L chromatography; at least one of the one or more polish chromatography unit operations can be cation exchange chromatography, anion exchange chromatography, multi-modal chromatography, or hydrophobic interaction chromatography.

[0018] Any of the above forms can be a method for producing an isolated, purified, recombinant protein of interest. In accordance with further aspects, an isolated, purified, recombinant protein of interest of the above method is disclosed and a pharmaceutical composition including the isolated, purified, recombinant protein of interest of the above method is disclosed.

[0019] In accordance with a further aspect, a system for neutralizing a viral inactivated pool to a target pH with minimal volume expansion is disclosed that includes a titrant that does not have buffer capacity in a target pH range and a buffering agent which buffers at the target pH, where the conductivity of the neutralized viral inactivation elution pool is maintained at less than or equal to 10 ms/cm.

[0020] In some forms, the system can include one or more of the following examples: the titrant can have a pKa greater than 7 and the buffering agent can have a pKa in the range of 4.5 to 6.0; the titrant can be acetate and the buffering agent can be Tris Base; the titrant can be in the range of greater than 0.1 M to less than 0.3 M sodium acetate; the buffering agent can be in the range of greater than 0.00005 M to less than 0.2 M Tris Base; the viral inactivation pool can be obtained by eluting an affinity chromatography column using a buffer comprising an acid