process to a unit in which the non-uniform feed stream (with a protein or product of interest) is replaced with a non-product containing fluid thereby voiding the product to prepare for the cleaning/sanitization process required before the next cycle is captured. This strategy, however, is typically unable to produce homogenization because after repeated circulations, the feed stream remains non-uniform (Figure 11). It is well-known in the art, that a non-uniform feed stream composition results in mutable viscosity and a variable protein concentration, both of which negatively affect the downstream CVI step. Therefore, there is a need for a device or method to reduce or eliminate these complexities.

SUMMARY

[0007] The present disclosure solves the above problems with the methods and devices disclosed herein, which results in faster and better homogenization of a non-uniform feed stream (e.g., from a ProA column) than the existing art.

[0008] The disclosure is directed to a device for an in-line homogenization of a non-uniform feed stream. In our system, the eluted product, which will be defined as the feed stream which contains at least the product of interest, is first stored within a plug flow reactor (PFR) as a non-uniform feed stream in which the concentration of the stream is variable in that it has a maximum and minimum concentration. The contained non-uniform feed stream is then circulated until the difference between the maximum and the minimum concentration is reduced to a sufficient range, the maximum is sufficiently reduced, or the minimum is sufficiently increased.

[0009] In one example, the device for in-line homogenization of a non-uniform feed stream comprises: (a) a plug flow reactor (PFR); (b) bypass line for diverting a percentage of the non-

uniform feed stream from a first location inside the PFR to a second location within the flow path; (c) a pump for circulating the non-uniform feed stream through the flow path. Thus, in one aspect, the present disclosure provides a device for in-line homogenization of a non-uniform feed stream, said device comprising a flow path comprising: (a) a plug flow reactor (PFR); (b) a bypass line for diverting a percentage of the non-uniform feed stream from a first location inside the PFR to a second location within the flow path; (c) a pump for circulating the non-uniform feed stream through the flow path. In a preferred example the non-uniform feed stream comprises a protein or product of interest.

[0010] In another example, the device is in-line placed between a first and a second process step. In an example, the first process step is a bind and elute column (Protein A) chromatography. In one example, the second process step is a virus inactivation step. In another aspect of the device, the flow path is a closed-circuit flow path. In yet another example, the device is tank-less.

[0011] In one example, the PFR comprises a tubing assembly packed in at least one chamber. The device may also include a bypass line as an extension of the flow path that fluidically connects to locations within the flow path. In one example, the second location is downstream from the first location. In another example, the second location is upstream from the first location. Alternatively, the second location is placed inside the PFR downstream or upstream from the first location. In one example of the device, the pump is selected from a group consisting of a centrifugal pump and /or a positive displacement pump.

The device may further comprise a second pump in the flow path, wherein the second pump modulates the non-uniform feedstream flow in the bypass line. In another example, the second pump is a pump selected from the group consisting of a centrifugal pump and/or a positive displacement pump. In one example, the non-uniform feed stream flow in the bypass line is co-current or counter-current to the non-uniform feed stream flow in the flow path.

[0012] In another aspect, a method for in-line homogenizing a non-uniform feed stream is provided, comprising: (a) providing a non-uniform feed stream, (b) flowing the non-uniform feed stream through a flow path comprising a PFR bypass line and a pump; (c) diverting a percentage of the non-uniform feed stream from a first location inside the PFR to

a second location within the flow path; (d) circulating the non-uniform feed stream through the flow path; and (e) homogenizing the non-uniform feed stream. In a preferred example this method comprises a protein or product of interest. In one example of the method, the flow path is in-line between a first and a second process step. In an alternative example, the first process step is a Protein A (ProA) column chromatography. The second process step in the method can be a virus inactivation step. In an example, the method is directed to virus inactivation.

[0013] In one example, the flow path is a closed-circuit flow path, wherein the PFR comprises a tubing assembly packed in at least one chamber. In another example, the bypass line is an extension of the flow path that fluidically connects to the first and second locations within the flow path. The second location in the method can be positioned downstream from the first location, or the second location can be positioned upstream from the first location, or the second location can be positioned inside the PFR - downstream or upstream from the first location.

[0014] In another example of the method, the pump is selected from a group consisting of a centrifugal pump and/or a positive displacement pump. In one example, the method further comprises a second pump in the flow path, wherein the second pump modulates the non-uniform feed stream flow in the bypass line. Alternatively, the method includes a second pump selected from the group consisting of: a centrifugal pump and/or a positive displacement pump.

[0015] In one example, the non-uniform feed stream flow within the bypass line is co-current or counter-current to the non-uniform feed stream flow in the flow path. In one example, the bypass line diverts the non-uniform feed stream with a flow rate that is about 1% to about 99% of the fastest flow rate in the flow path.

[0016] In another aspect, the disclosure is directed to a method of manufacturing a product of interest by a device as defined above. In another aspect, the disclosure is also directed to a method of

manufacturing a protein by a device as defined above. In a preferred mode this protein is a therapeutic protein.

[0017] In another aspect, the disclosure is directed to a method of manufacturing a product of interest by in-line homogenizing a non-uniform feed stream as defined above. The disclosure is also directed to a method of manufacturing a protein by a non-uniform feed stream as defined above. In another aspect, the disclosure is directed to a method of manufacturing a protein by in-line homogenizing a non-uniform feed stream as described herein. In a preferred mode this protein is a therapeutic protein.

[0018] Additional features and advantages of various examples, aspects, will be set forth, in the description that follows, and will, be apparent from the description, or may be learned by the practice of various examples. The objectives and other advantages of various examples will be realized and attained using the elements and combinations particularly pointed out in the description herein.

BRIEF DESCRIPTION OF THE DRAWINGS

[0019] The present disclosure, its several aspects, and embodiments can be more fully understood from the detailed description, claims, and the accompanying drawings.

[0020] Fig. 1 is a graph illustrating the relative concentration of protein eluate resulting from Protein A column chromatography as represented by an asymmetric Gaussian curve based on the inherent properties of the bind and elute process. The X-axis shows the Column Volume of the elution stream, and the Y-axis shows the Relative Concentration (%) of the eluate.

[0021] Fig. 2 illustrates an exemplary plurality of plug flow reactors.

[0022] Fig. 3 illustrates the flow pattern in each of the plug flow reactors shown in Fig. 2, according to an example of the present disclosure.

[0023] Fig. 4 illustrates a relationship between Protein A chromatography columns and the flow circulation device according to an example of the present disclosure;

CLAIMS

We claim:

1. A device for in-line homogenization of a non-uniform feed stream, said device comprising a flow path comprising:
   (a) a plug flow reactor (PFR);
   (b) a bypass line for diverting a percentage of the non-uniform feed stream from a first location inside the PFR to a second location within the flow path;
   (c) a pump for circulating the non-uniform feed stream through the flow path.

2. The device of claim 1, wherein the non-uniform feed stream comprises a protein or product of interest.

3. The device of claim 1, wherein the device is in-line between a first and a second process step.

4. The device of claim 3, wherein the first process is selected from the group consisting of: bind and elute chromatography and Protein A chromatography.

5. The device of claim 3, wherein the second process step is a virus inactivation step.

6. The device of claim 1, wherein the device is for virus inactivation.

7. The device of claim 1, wherein the flow path is a closed-circuit flow path.

8. The device of claim 1, wherein the device is tank-less

9. The device of claim 1, wherein the PFR comprises a tubing assembly packed in at least one chamber.

10. The device of claim 1, wherein the bypass line is an extension of the flow path that

fluidically connects to locations within the flow path.

11. The device of claim 1, wherein the second location is downstream from the first location.

12. The device of claim 1, wherein the second location is upstream from the first location.

13. The device of claim 1, wherein the second location is inside the PFR downstream or upstream from the first location.

14. The device of claim 1, wherein the pump is a pump selected from the group consisting of: a centrifugal pump and a positive displacement pump.

15. The device of claim 1, wherein the device further comprises a second pump in the flow path.

16. The device of claim 15, wherein the second pump modulates the non-uniform feed stream flow in the bypass line.

17. The device of claim 15, wherein the second pump is a pump selected from the group consisting of a centrifugal pump and a positive displacement pump.

18. The device of claim 1, wherein the non-uniform feed stream flow in the bypass line is co-current or counter-current to the non-uniform feed stream flow in the flow path.

19. The device of claim 15, wherein the non-uniform feed stream flow in the bypass line is co-current or counter-current to the non-uniform feed stream flow in the flow path.

20. A method for in-line homogenizing a non-uniform feed stream, comprising:
    (a) Providing a non-uniform feed stream,
    (b) Flowing the non-uniform feed stream through a flow path comprising a PFR

bypass line and a pump;

(c) Diverting a percentage of the non-uniform feed stream from a first location inside the PFR to a second location within the flow path;

(d) Circulating the non-uniform feed stream through the flow path; and

(e) Homogenizing the non-uniform feed stream.

21. The method of claim 20, wherein the non-uniform feed stream comprises a protein or product of interest.

22. The method of claim 20, wherein the flow path is in-line between a first and a second process step.

23. The method of claim 22, wherein the first process step is a Protein A (ProA) column chromatograph.

24. The method of claim 22, wherein the second process step is a virus inactivation step.

25. The method of claim 20, wherein the method is for virus inactivation.

26. The method of claim 20, wherein the flow path is a closed-circuit flow path.

27. The method of claim 20, wherein the PFR comprises a tubing assembly packed in at least one chamber.

28. The method of claim 20, wherein the bypass line is an extension of the flow path that fluidically connects to the first and second locations within the flow path.

29. The method of claim 20, wherein the second location is downstream from the first location.

30. The method of claim 20, wherein the second location is upstream from the first location.

31. The method of claim 20, wherein the second location is inside the PFR downstream or upstream from the first location.

32. The method of claim 20, wherein the pump is a pump selected from the group consisting of a centrifugal pump and a positive displacement pump.

33. The method of claim 20, wherein the method further comprises a second pump in the flow path.

34. The method of claim 33, wherein the second pump modulates the non-uniform feed stream flow in the bypass line.

35. The method of claim 33, wherein the second pump is a pump selected from the group consisting of a centrifugal pump and a positive displacement pump.

36. The method of claim 20, wherein the non-uniform feed stream flow in the bypass line is co-current or counter-current to the non-uniform feed stream flow in the flow path.

37. The method of claim 36, wherein the bypass line diverts the non-uniform feed stream with a flow rate that is about 1% to 99% of the flow rate in the flow path.

38. Method of manufacturing a product of interest by a device according to claim 1.

39. Method of manufacturing a protein by a device according to claim 1.

40. Method of claim 39, wherein the protein is a therapeutic protein.

41. Method of manufacturing a product of interest by in-line homogenizing a non-uniform feed stream according to claim 20.

42. Method of manufacturing a protein by in-line homogenizing a non-uniform feed stream according to claim 20.

43. Method of claim 42, wherein the protein is a therapeutic protein.