Does Flow Play A Role in Protein Aggregation During Bioprocessing?

References

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1. Cromwell, M. E. M.; Hilario, E.; Jacobson, F. Protein Aggregation and Bioprocessing. AAPS J 2006, 8 (3), E572-9. https://doi.org/10.1208/aapsj080366.
2. Cohen, J. R.; Brych, S. R.; Prabhu, S.; Bi, V.; Elbaradei, A.; Tokuda, J. M.; Xiang, C.; Hokom, M.; Cui, X.; Ly, C.; Amos, N.; Sun, J.; Calamba, D.; Herskovitz, J.; Capili, A.; Nourbakhsh, K.; Merlo, A.; Carreon, J.; Wypych, J.; Narhi, L. O.; Jawa, V.; Joubert, M. K. A High Threshold of Biotherapeutic Aggregate Numbers Is Needed to Induce an Immunogenic Response In Vitro, In Vivo, and in the Clinic. Pharm Res 2024. https://doi.org/10.1007/s11095-024-03678-2.
3. Eyes, T. J.; Austerberry, J. I.; Dearman, R. J.; Johannissen, L. O.; Kimber, I.; Smith, N.; Thistlethwaite, A.; Derrick, J. P. Identification of B Cell Epitopes Enhanced by Protein Unfolding and Aggregation. Mol Immunol 2019, 105 (October 2018), 181–189. https://doi.org/10.1016/j.molimm.2018.11.020.
4. Farid, S. S.; Baron, M.; Stamatis, C.; Nie, W.; Coffman, J. Benchmarking Biopharmaceutical Process Development and Manufacturing Cost Contributions to R&D. MAbs 2020, 12 (1). https://doi.org/10.1080/19420862.2020.1754999.
5. Mazzer, A. R.; Perraud, X.; Halley, J.; O'Hara, J.; Bracewell, D. G. Protein A Chromatography Increases Monoclonal Antibody Aggregation Rate during Subsequent Low PH Virus Inactivation Hold. J Chromatogr A 2015, 1415, 83–90. https://doi.org/10.1016/j.chroma.2015.08.068.
6. Menzen, T.; Friess, W. Temperature-Ramped Studies on the Aggregation, Unfolding, and Interaction of a Therapeutic Monoclonal Antibody. J Pharm Sci 2014, 103 (2), 445–455. https://doi.org/10.1002/jps.23827.
7. Bekard, I. B.; Asimakis, P.; Bertolini, J.; Dunstan, D. E. The Effects of Shear Flow on Protein Structure and Function. Biopolymers 2011, 95 (11), 733–745. https://doi.org/10.1002/bip.21646.
8. Thomas, C. R.; Geer, D. Effects of Shear on Proteins in Solution. Biotechnol Lett 2011, 33, 443–456. https://doi.org/10.1007/s10529-010-0469-4.
9. Willis, L. F. The Effects of Flow on Therapeutic Protein Aggregation, University of Leeds, 2018. https://doi.org/https://etheses.whiterose.ac.uk/21963/.
10. Ratanji, K. D.; Derrick, J. P.; Dearman, R. J.; Kimber, I. Immunogenicity of Therapeutic Proteins: Influence of Aggregation. J Immunotoxicol 2014, 11 (2), 99–109. https://doi.org/10.3109/1547691X.2013.821564.

11. Papež, P.; Merzel, F.; Praprotnik, M. Rotational Dynamics of a Protein under Shear Flow Studied by the Eckart Frame Formalism. J Phys Chem B 2023, 127 (33), 7231–7243. https://doi.org/10.1021/acs.jpcb.3c02324.
12. Simon, S.; Krause, H. J.; Weber, C.; Peukert, W. Physical Degradation of Proteins in Well-Defined Fluid Flows Studied within a Four-Roll Apparatus. Biotechnol Bioeng 2011, 108 (12), 2914–2922. https://doi.org/10.1002/bit.23257.
13. Ashton, L.; Dusting, J.; Imomoh, E.; Balabani, S.; Blanch, E. W. Susceptibility of Different Proteins to Flow-Induced Conformational Changes Monitored with Raman Spectroscopy. Biophys J 2010, 98 (4), 707–714. https://doi.org/10.1016/j.bpj.2009.10.010.
14. Jaspe, J.; Hagen, S. J. Do Protein Molecules Unfold in a Simple Shear Flow? Biophys J 2006, 91 (9), 3415–3424. https://doi.org/10.1529/biophysj.106.089367.
15. Bee, J. S.; Stevenson, J. L.; Mehta, B.; Svitel, J.; Pollastrini, J.; Platz, R.; Freund, E.; Carpenter, J. F.; Randolph, T. W. Response of a Concentrated Monoclonal Antibody Formulation to High Shear. Biotechnol Bioeng 2009, 103 (5), 936–943. https://doi.org/10.1002/bit.22336.
16. Dobson, J.; Kumar, A.; Willis, L. F.; Tuma, R.; Higazi, D. R.; Turner, R.; Lowe, D. C.; Ashcroft, A. E.; Radford, S. E.; Kapur, N.; Brockwell, D. J. Inducing Protein Aggregation by Extensional Flow. Proceedings of the National Academy of Sciences 2017, 114 (18), 4673–4678. https://doi.org/10.1073/pnas.1702724114.
17. Duerkop, M.; Berger, E.; Dürauer, A.; Jungbauer, A. Influence of Cavitation and High Shear Stress on HSA Aggregation Behavior. Eng Life Sci 2018, 18 (3), 169–178. https://doi.org/10.1002/elsc.201700079.
18. Fu, H.; Jiang, Y.; Yang, D.; Scheiflinger, F.; Wong, W. P.; Springer, T. A. Flow-Induced Elongation of von Willebrand Factor Precedes Tension-Dependent Activation. Nat Commun 2017, 8 (1). https://doi.org/10.1038/s41467-017-00230-2.
19. Bergal, H. T.; Jiang, Y.; Yang, D.; Springer, T. A.; Wong, W. P. Conformation of von Willebrand Factor in Shear Flow Revealed with Stroboscopic Single-Molecule Imaging. Blood 2022, 140 (23), 2490–2499. https://doi.org/10.1182/blood.2022016969.
20. Springer, T. Von Willebrand Factor, Jedi Knight of the Bloodstream. Blood 2016, 113 (21), 1412–1425. https://doi.org/10.1182/blood-2008-10-165621.
21. Rammensee, S.; Slotta, U.; Scheibel, T.; Bausch, A. R. Assembly Mechanism of Recombinant Spider Silk Proteins. Proc Natl Acad Sci USA 2008, 105 (18), 6590–6595. https://doi.org/10.1073/pnas.0709246105.
22. Roberts, C. J. Protein Aggregation and Its Impact on Product Quality. Curr Opin Biotechnol 2014, 30C, 211–217. https://doi.org/10.1016/j.copbio.2014.08.001.
23. Li, J.; Krause, M. E.; Chen, X.; Cheng, Y.; Dai, W.; Hill, J. J.; Huang, M.; Jordan, S.; LaCasse, D.; Narhi, L.; Shalaev, E.; Shieh, I. C.; Thomas, J. C.; Tu, R.; Zheng, S.; Zhu, L. Interfacial Stress in the Development of Biologics: Fundamental Understanding, Current Practice, and Future Perspective. AAPS Journal 2019, 21 (3). https://doi.org/10.1208/s12248-019-0312-3.
24. Kopp, M. R. G.; Grigolato, F.; Zürcher, D.; Das, T. K.; Chou, D.; Wuchner, K.; Arosio, P. Surface-Induced Protein Aggregation and Particle Formation in Biologics: Current Understanding of Mechanisms, Detection and Mitigation Strategies. J Pharm Sci 2023, 112 (2), 377–385. https://doi.org/10.1016/j.xphs.2022.10.009.

25. Rabe, M.; Verdes, D.; Seeger, S. Understanding Protein Adsorption Phenomena at Solid Surfaces. Adv Colloid Interface Sci 2011, 162 (1–2), 87–106. https://doi.org/10.1016/j.cis.2010.12.007.
26. Moino, C.; Artusio, F.; Pisano, R. Shear Stress as a Driver of Degradation for Protein-Based Therapeutics: More Accomplice than Culprit. International Journal of Pharmaceutics. Elsevier B.V. January 25, 2024. https://doi.org/10.1016/j.ijpharm.2023.123679.
27. Abaci, H. E.; Shen, Y.-I.; Tan, S.; Gerecht, S. Recapitulating Physiological and Pathological Shear Stress and Oxygen to Model Vasculature in Health and Disease. Sci Rep 2015, 4 (1), 4951. https://doi.org/10.1038/srep04951.
28. Hakala, T. A.; Yates, E. V.; Challa, P. K.; Toprakcioglu, Z.; Nadendla, K.; Matak-Vinkovic, D.; Dobson, C. M.; Martínez, R.; Corzana, F.; Knowles, T. P. J.; Bernardes, G. J. L. Accelerating Reaction Rates of Biomolecules by Using Shear Stress in Artificial Capillary Systems. J Am Chem Soc 2021, 143 (40), 16401–16410. https://doi.org/10.1021/jacs.1c03681.
29. Gikanga, B.; Maa, Y. F. A Review on Mixing-Induced Protein Particle Formation: The Puzzle of Bottom-Mounted Mixers. J Pharm Sci 2020, 1–12. https://doi.org/10.1016/j.xphs.2020.03.024.
30. Sediq, A. S.; Van Duijvenvoorde, R. B.; Jiskoot, W.; Nejadnik, M. R. No Touching! Abrasion of Adsorbed Protein Is the Root Cause of Subvisible Particle Formation during Stirring. J Pharm Sci 2016, 105 (2), 519–529. https://doi.org/10.1016/j.xphs.2015.10.003.
31. Zhan, C.; Bidkhori, G.; Schwarz, H.; Malm, M.; Mebrahtu, A.; Field, R.; Sellick, C.; Hatton, D.; Varley, P.; Mardinoglu, A.; Rockberg, J.; Chotteau, V. Low Shear Stress Increases Recombinant Protein Production and High Shear Stress Increases Apoptosis in Human Cells. iScience 2020, 23 (11). https://doi.org/10.1016/j.isci.2020.101653.
32. Dransart, B.; Wheeler, A.; Hong, T.; Tran, C.; Abalos, R.; Quezada, A.; Wang, S.; Kluck, B.; Sanaie, N. Evaluation of a Next-Generation Protein A Chromatography Resin for the Purification of Monoclonal Antibodies. Bioprocess Int 2018, 16 (12), 14–19.
33. Joseph, A.; Kenty, B.; Mollet, M.; Hwang, K.; Rose, S.; Goldrick, S.; Bender, J.; Farid, S. S.; Titchener-Hooker, N. A Scale-down Mimic for Mapping the Process Performance of Centrifugation, Depth and Sterile Filtration. Biotechnol Bioeng 2016, 113 (9), 1934–1941. https://doi.org/10.1002/bit.25967.
34. Arunkumar, A.; Singh, N.; Schutsky, E. G.; Peck, M.; Swanson, R. K.; Borys, M. C.; Li, Z. J. Effect of Channel-Induced Shear on Biologics during Ultrafiltration/Diafiltration (UF/DF). J Memb Sci 2016, 514, 671–683. https://doi.org/10.1016/j.memsci.2016.05.031.
35. ITO, T.; Wang, H.; Hwang, S. H.; Wang, B.; Wang, L.; G, S. Risk Assessment for Biopharmaceutical Single-Use Manufacturing: A Case Study of Upstream Continuous Processing. Biologicals 2023, 84. https://doi.org/10.1016/j.biologicals.2023.101713.
36. Roffi, K.; Li, L.; Pantazis, J. Adsorbed Protein Film on Pump Surfaces Leads to Particle Formation during Fill-Finish Manufacturing. Biotechnol Bioeng 2021, 118 (8), 2947–2957. https://doi.org/10.1002/bit.27801.
37. Adler, M.; Allmendinger, A. Filling Unit Operation for Biological Drug Products: Challenges and Considerations. Journal of Pharmaceutical Sciences. Elsevier B.V. 2023. https://doi.org/10.1016/j.xphs.2023.11.017.
38. Randolph, T. W.; Schiltz, E.; Sederstrom, D.; Steinmann, D.; Mozziconacci, O.; Schöneich, C.; Freund, E.; Ricci, M. S.; Carpenter, J. F.; Lengsfeld, C. S. Do Not Drop: Mechanical Shock

in Vials Causes Cavitation, Protein Aggregation, and Particle Formation. J Pharm Sci 2015, 104, 602–611. https://doi.org/10.1002/jps.24259.
39. Fleischman, M. L.; Chung, J.; Paul, E. P.; Lewus, R. A. Shipping-Induced Aggregation in Therapeutic Antibodies: Utilization of a Scale-Down Model to Assess Degradation in Monoclonal Antibodies. J Pharm Sci 2017, 106 (4), 994–1000. https://doi.org/http://dx.doi.org/10.1016/j.xphs.2016.11.021.
40. DASNOY, S.; ILLARTIN, M.; QUEFFELEC, J.; NKUNKU, A.; PEERBOOM, C. Combined Effect of Shaking Orbit and Vial Orientation on the Agitation-Induced Aggregation of Proteins. J Pharm Sci 2023. https://doi.org/10.1016/j.xphs.2023.08.016.
41. Willis, L. F.; Toprani, V.; Wijetunge, S.; Sievers, A.; Lin, L.; Williams, J.; Crowley, T. J.; Radford, S. E.; Kapur, N.; Brockwell, D. J. Exploring a Role for Flow-Induced Aggregation Assays in Platform Formulation Optimisation for Antibody-Based Proteins. J Pharm Sci 2024, 113 (3), 625–636. https://doi.org/10.1016/j.xphs.2023.10.031.
42. Thite, N. G.; Ghazvini, S.; Wallace, N.; Feldman, N.; Calderon, C. P.; Randolph, T. W. Interfacial Adsorption Controls Particle Formation in Antibody Formulations Subjected to Extensional Flows and Hydrodynamic Shear. J Pharm Sci 2023. https://doi.org/10.1016/j.xphs.2023.07.010.
43. Thite, N. G.; Ghazvini, S.; Wallace, N.; Feldman, N.; Calderon, C. P.; Randolph, T. W. Interfacial Adsorption Controls Particle Formation in Antibody Formulations Subjected to Extensional Flows and Hydrodynamic Shear. J Pharm Sci 2023. https://doi.org/10.1016/j.xphs.2023.07.010.
44. Grabarek, A. D.; Bozic, U.; Rousel, J.; Menzen, T.; Kranz, W.; Wuchner, K.; Jiskoot, W.; Hawe, A. What Makes Polysorbate Functional? Impact of Polysorbate 80 Grade and Quality on IgG Stability During Mechanical Stress. J Pharm Sci 2020, 109 (1), 871–880. https://doi.org/10.1016/j.xphs.2019.10.015.
45. Grigolato, F.; Arosio, P. Synergistic Effects of Flow and Interfaces on Antibody Aggregation. Biotechnol Bioeng 2020, 117 (2), 417–428. https://doi.org/10.1002/bit.27212.
46. Du, Y.; Song, J.; Lu, L.; Yeung, E.; Givand, J.; Procopio, A.; Su, Y.; Hu, G. Design of a Reciprocal Injection Device for Stability Studies of Parenteral Biological Drug Products. J Pharm Sci 2023. https://doi.org/10.1016/j.xphs.2023.12.014.
47. Fardin, M. A.; Perge, C.; Taberlet, N. “The Hydrogen Atom of Fluid Dynamics” -- Introduction to the Taylor-Couette Flow for Soft Matter Scientists. Soft Matter 2014, 10 (i), 3523–3535. https://doi.org/10.1039/c3sm52828f.
48. Bekard, I. B.; Dunstan, D. E. Shear-Induced Deformation of Bovine Insulin in Couette Flow. J Phys Chem B 2009, 113 (25), 8453–8457. https://doi.org/10.1021/jp903522e.
49. Fanthom, T. B.; Wilson, C.; Gruber, D.; Bracewell, D. G. Solid-Solid Interfacial Contact of Tubing Walls Drives Therapeutic Protein Aggregation During Peristaltic Pumping. J Pharm Sci 2023, 112 (12), 3022–3034. https://doi.org/10.1016/j.xphs.2023.08.012.
50. McBride, S. A.; Sanford, S. P.; Lopez, J. M.; Hirsa, A. H. Shear-Induced Amyloid Fibrillization: The Role of Inertia. Soft Matter 2016, 12 (12), 3461–3467. https://doi.org/10.1039/c5sm02916c.
51. Ashton, L.; Dusting, J.; Imomoh, E.; Balabani, S.; Blanch, E. W. Shear-Induced Unfolding of Lysozyme Monitored in Situ. Biophys J 2009, 96 (10), 4231–4236. https://doi.org/10.1016/j.bpj.2009.02.024.

52. Dreckmann, T.; Boeuf, J.; Ludwig, I. S.; Lümkemann, J.; Huwyler, J. Low Volume Aseptic Filling: Impact of Pump Systems on Shear Stress. European Journal of Pharmaceutics and Biopharmaceutics 2020, 147 (August 2019), 10–18. https://doi.org/10.1016/j.ejpb.2019.12.006.
53. Sharma, C.; Malhotra, D.; Rathore, A. S. Review of Computational Fluid Dynamics Applications in Biotechnology Processes. Biotechnol Prog 2011, 27 (6), 1497–1510. https://doi.org/10.1002/btpr.689.
54. Bai, G.; Bee, J. S.; Biddlecombe, J. G.; Chen, Q.; Leach, W. T. Computational Fluid Dynamics (CFD) Insights into Agitation Stress Methods in Biopharmaceutical Development. Int J Pharm 2012, 423 (2), 264–280. https://doi.org/10.1016/j.ijpharm.2011.11.044.
55. Willis, L. F.; Kumar, A.; Jain, T.; Caffry, I.; Xu, Y.; Radford, S. E.; Kapur, N.; Vásquez, M.; Brockwell, D. J. The Uniqueness of Flow in Probing the Aggregation Behavior of Clinically Relevant Antibodies. Engineering Reports 2020, 2 (5), 1–13. https://doi.org/10.1002/eng2.12147.
56. Willis, L. F.; Kumar, A.; Dobson, J.; Bond, N. J.; Lowe, D.; Turner, R.; Radford, S. E.; Kapur, N.; Brockwell, D. J. Using Extensional Flow to Reveal Diverse Aggregation Landscapes for Three IgG1 Molecules. Biotechnol Bioeng 2018, 115 (5), 1216–1225. https://doi.org/10.1002/bit.26543.
57. Bekard, I. B.; Asimakis, P.; Teoh, C. L.; Ryan, T.; Howlett, G. J.; Bertolini, J.; Dunstan, D. E. Bovine Serum Albumin Unfolds in Couette Flow. Soft Matter 2012, 8 (ii), 385. https://doi.org/10.1039/c1sm06704d.
58. Deiringer, N.; Aleshkevich, S.; Müller, C.; Friess, W. Modification of Tubings for Peristaltic Pumping of Biopharmaceutics. J Pharm Sci 2022. https://doi.org/10.1016/j.xphs.2022.08.037.
59. Strickley, R. G.; Lambert, W. J. A Review of Formulations of Commercially Available Antibodies. J Pharm Sci 2021, 110 (7), 2590-2608.e56. https://doi.org/10.1016/j.xphs.2021.03.017.
60. Brosig, S.; Cucuzza, S.; Serno, T.; Bechtold-Peters, K.; Buecheler, J.; Zivec, M.; Germershaus, O.; Gallou, F. Not the Usual Suspects: Alternative Surfactants for Biopharmaceuticals. ACS Appl Mater Interfaces 2023, 15 (29), 34540–34553. https://doi.org/10.1021/acsami.3c05610.
61. Katz, J. S.; Chou, D. K.; Christian, T. R.; Das, T. K.; Patel, M.; Singh, S. N.; Wen, Y. Emerging Challenges and Innovations in Surfactant-Mediated Stabilization of Biologic Formulations. J Pharm Sci 2022, 111 (4), 919–932. https://doi.org/10.1016/j.xphs.2021.12.002.
62. Cloutier, T.; Sudrik, C.; Mody, N.; Sathish, H. A.; Trout, B. L. Molecular Computations of Preferential Interaction Coefficients of IgG1 Monoclonal Antibodies with Sorbitol, Sucrose, and Trehalose and the Impact of These Excipients on Aggregation and Viscosity. Mol Pharm 2019, 16 (8), 3657–3664. https://doi.org/10.1021/acs.molpharmaceut.9b00545.
63. Svilenov, H. L.; Kulakova, A.; Zalar, M.; Golovanov, A. P.; Harris, P.; Winter, G. Orthogonal Techniques to Study the Effect of PH, Sucrose, and Arginine Salts on Monoclonal Antibody Physical Stability and Aggregation During Long-Term Storage. J Pharm Sci 2020, 109 (1), 584–594. https://doi.org/10.1016/j.xphs.2019.10.065.

64. Ng, Y. K.; Konermann, L. Mechanism of Protein Aggregation Inhibition by Arginine: Blockage of Anionic Side Chains Favors Unproductive Encounter Complexes. J Am Chem Soc 2024, 146 (12), 8394–8406. https://doi.org/10.1021/jacs.3c14180.
65. Kim, N. A.; Hada, S.; Thapa, R.; Jeong, S. H. Arginine as a Protein Stabilizer and Destabilizer in Liquid Formulations. Int J Pharm 2016, 513 (1–2), 26–37. https://doi.org/10.1016/j.ijpharm.2016.09.003.