Selective recovery of inclusion bodies.

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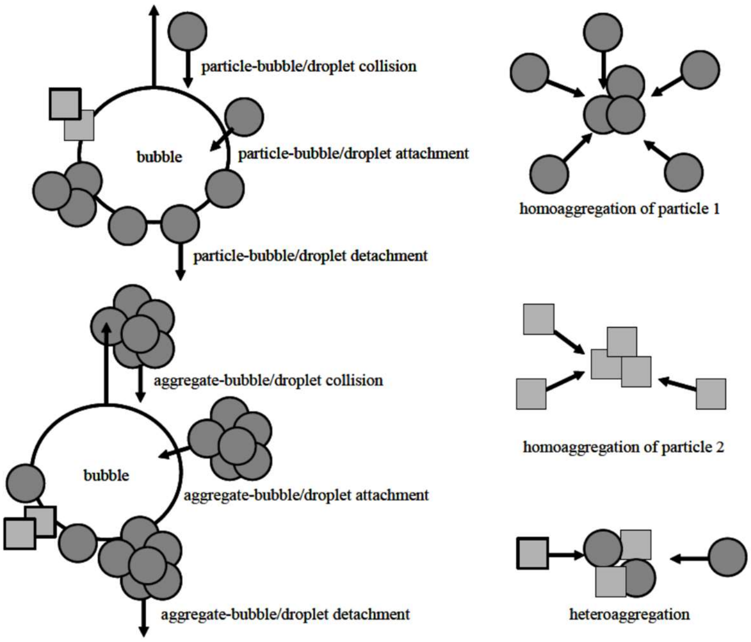
Description



Many products in biotechnological processes are produced as particles. Examples are crystals, inclusion bodies, virus-like particles and precipitates. Many of these products are recovered and purified through solubilisation in combination with extraction or selective recrystallisation. In many cases the target particles contain the product already at high purity, making selective particle recovery more efficient. It requires separation techniques that have the ability to separate the target particles from the liquid phase and in general also from other particles, such as cells, cell debris, by-products or immobilised catalysts. Centrifugation and filtration are inefficient for many of these separations due to the overlapping size distributions and similar settling velocities of the particles. Other particle separation methods are therefore desirable. Examples of driving forces that might be applied for particle-particle separation are the electric force, the magnetic force and the interfacial tension force. The application of these forces for particle-particle separation is evaluated on the basis of the typical properties of bioparticles.
In theory, the interfacial tension force has a large potential for these separations. Separation with this force is based on differences in adsorption behaviour of particles to fluid-fluid interfaces. This technique has been applied for the recovery of minerals, removal of ink from paper pulp and concentration/removal of microbial cells. However, its application for particle-particle separation in biotechnology has been scarcely reported in literature. Separation techniques that make use of fluid-fluid interfaces and aim at applying air flotation, a method that makes use of the air-water interface, for the selective recovery of bioparticles from particle mixtures was studied in detail and is advocated to be used.
Particulate products can be produced inside microbial cells. Selective recovery of these products requires cell disintegration for release of the particulate product into the fermentation medium. Cell disruption can influence the size, shape and surface properties of the product particles and the undesired particles. It therefore affects the particle-particle separation technique used. A case study on the release of medium-chain-length polyhydroxyalkanoate (mcl-PHA) inclusion bodies from *Pseudomonas putida* cells was performed. The influence of mechanical, enzymatic and combined chemical/mechanical cell disruption on cell debris particle size and inclusion body release was investigated.

Dissertation

Pim van Hee, Selective recovery of micrometer bioparticles using aggration and air flotation. PhD Thesis, Delft University of Technology, 2006. ISBN 90-9020361-3 (in TUD-repository, https://repository.tudelft.nl/islandora/object/uuid%3A5ef4e4f8-997a-4f1a-a302-ef1056f5f498?collection=research.

Publications from the dissertation

P. van Hee, Martijn A. Hoeben, Rob van der Lans, Luuk A M van der Wielen, 2007. Fluid interfaces for selective bioparticle recovery. In book: New Colloid and Surface Science Research. Chapter: 3. Publisher: Nova Science Publishers Inc. Editor: Lawrence V. Schwartz ISBN: 978-1-60021-594-0 Pages 131-136

van Hee P, Hoeben MA, van der Lans RGJM, van der Wielen, LAM, 2006
Strategy for selection of methods for separation of bioparticles from particle mixtures
Biotechnology and bioengineering 94 (4): 689-709 jul 5 2006
https://doi.org/10.1002/bit.20885

Pim van Hee, Andrea CMR Elumbaring, Rob GJM van der Lans, Luuk AM van der Wielen, 2006 Selective recovery of polyhydroxyalkanoate inclusion bodies from fermentation broth by dissolved-air flotation. Journal of Colloid and Interface Science 297 (2006) 595-606
https://doi.org/10.1016/j.jcis.2005.11.019

Van Hee P., W.K. Lin, L. Benac-Vegas, R.G.J.M. Van der Lans, G.J.M. Koper and L.A.M. Van der Wielen, 2006
Selective recovery of micrometer particles from mixtures using a combination of selective aggregation and dissolved-air flotation
Colloids and Surfaces A: Physicochemical and Engineering Aspects 280 (1-3): 216-231
https://doi.org/10.1016/j.colsurfa.2006.02.003

Hee, P van, Wielen, LAM van der, & Lans, RGJM van der (21-07-2005). Producing a fermentation product from an organism comprising lysing the organism using a lysis-promoting compound, separating the lysing-promoting agent and fermentation product, and culturing the organism with the lysis-promoting agent. Patent application. NL1025149, EP1553167, US 2005/0158817, JP2005/192561.

van Hee P, Middelberg APJ, van der Lans RGJM, van der Wielen LAM, 2004
Relation between cell disruption conditions, cell debris particle size, and inclusion body release
Biotechnology and bioengineering 88 (1): 100-110 oct 5 2004
https://doi.org/10.1002/bit.20343

van Hee P, Middelberg APJ, van der Lans RGJM, van der Wielen LAM, 2004
Quantification of solid cell material by detection of membrane-associated proteins and peptidoglycan. Journal of chromatography b-analytical technologies in the biomedical and life sciences 807 (1): 111-119 jul 25 2004
https://doi.org/10.1016/j.jchromb.2004.01.052