Available online at www.sciencedirect.com

ScienceDirect

IFAC Papers Online CONFERENCE PAPER ARCHIVE

IFAC-PapersOnLine 49-26 (2016) 258–263

Extraction of Physiological State Functions in Heterogeneous Cell Population Models ⋆

Mubashir Hussain *, **Steffen Waldherr** **,***,****

* *Institute of Multiscale Simulation, Friedrich-Alexander University, Erlangen-Nürnberg, Germany (e-mail: mubashir.hussain@fau.de)*
** *Institute of Automation Engineering, Otto von Guericke University, Magdeburg, Germany*
*** *Harvard University, School of Engineering and Applied Sciences, USA*
**** *KU Leuven, Department of Chemical Engineering, Belgium (email: steffen.waldherr@kuleuven.be)*

Abstract:
A simple yet generic approach is presented to extract the growth and breakage kinetics from the temporal data of heterogeneous cell population. The moment form of the one-dimensional population balance equation is directly solved by a MATLAB solver for linear systems to extract the kinetics. The corresponding systems of linear equations are, however, highly underdetermined and ill-conditioned. To address this, the problem is regularized by assuming a suitable upper bound of the solution. The range between minimum and maximum possible values of the solution is discretized into several sub-intervals. The system is then solved against each sub-interval, whose values are used as lower and upper bounds in a suitable MATLAB solver and a local solution is obtained in all these ranges. The final solution is then computed by taking average of all solutions having residual norms less than a particular threshold. To validate the method, the results of the inverse technique are compared and discussed against two theoretical experiments.

Keywords: Kinetics, Inverse Problem, Population Balances

1. INTRODUCTION

Extraction of underlying kinetics from the experimental data plays an important role in understanding the dynamics of a particulate process and its correlation with the process conditions and material properties. The complex nature of particulate processes, however, makes this task quite challenging, especially when the experimental data is taken from living cells.

In a cellular system, cells grow in size in the presence of favorable conditions such as adequate substrate. The cells, though being cultured in the same environment, may grow with different rates due to several reasons such as intracellular content, thus giving rise to heterogeneity in the population. Moreover, the cells start dividing upon reaching a certain size and the new born daughter cells usually also differ in sizes. This amplifies the heterogeneity in the cell population.

Mathematically, a heterogeneous cell population is usually modeled by means of population balance equations (PBEs) (Ramkrishna (2000)). A typical one-dimensional PBE with combined growth and breakage for cell size distribution (CSD) of a cell population cultured in a batch reactor with excess substrate is given by

$$\frac{\partial n(v,t)}{\partial t} + \frac{\partial}{\partial v}\left[G(v)n(v,t)\right] + \Gamma(v)n(v,t) = 2\int_{v}^{v_{max}} \Gamma(u)P(v,u)n(u,t)du. \quad (1)$$

Here, n is the cell number density, v represents the diameter/size of a cell, G represents the growth rate of a cell, Γ is the breakage rate and $P(v,u)$ is the partition probability density function i.e., the probability that a mother cell of size u will divide into daughter cells of sizes v and $u-v$ upon binary cell division.

In cellular systems, it is often assumed that cells neither divide into daughter cells of sizes less than a certain size nor grow beyond an upper threshold. Such a regularity boundary condition is usually applied to solve (1) and is given by

$$G(v_{min})n(v_{min},t) = 0 = G(v_{max})n(v_{max},t). \quad (2)$$

Furthermore, the bivariate partition probability density function $P(v,u)$ satisfies the normalization condition,

$$\int_0^u P(v,u)dv = 1. \quad (3)$$

The intrinsic physiological state (IPS) functions i.e., G, Γ and P govern the dynamics of the cell population. However, the dynamics of heterogeneous cell population

⋆ The authors of this paper gratefully acknowledge the financial support provided by the DFG (German Research Foundation) for this research work (grant WA2965/1-1).

2405-8963 © 2016, IFAC (International Federation of Automatic Control) Hosting by Elsevier Ltd. All rights reserved.
Peer review under responsibility of International Federation of Automatic Control.
10.1016/j.ifacol.2016.12.135

can only be comprehensively modeled by (1) provided that the underlying kinetics are correctly reflected by the corresponding IPS functions. This is, however, challenging as cell growth is a complex process and depends on several factors such as intracellular content, cell types, cell cycle, process conditions etc. It is, therefore, often required to extract these functions from the experimental data to better understand the underlying kinetic laws.

The inverse problem is also effortful and poses many experimental and computational challenges. Collins and Richmond (1962) derived a mathematical expression for the growth rate by assuming exponential balanced growth. However, their model can not be used to extract the growth rate from the experimental data unless the number density distribution of the daughter cells as well as the number density distribution of the mother cells are known. Both distribution are quite challenging to be determined by experiments. Similarly, Ramkrishna et al. (1968) also derived an expression for the breakage rate which also depends on the number density distribution of the mother cells.

Recently, a method to extract kinetics from experimental data of the cellular systems has been proposed by Spetsieris and Zygourakis (2012) by using the mathematical expressions of Collins and Richmond (1962) and Ramkrishna et al. (1968). They developed a new sophisticated technique based on fluorescence microscopy and image processing to determine subpopulations of mother and daughter cells (Spetsieris et al. (2009)). They used a morphological criterion based on the size of the characteristic constriction to identify the subpopulation of mother cells. The procedure, however, is quite complex and limited to rod-shaped cells. Bouaswaig and Engell (2010) also proposed an inverse technique to extract the growth kinetics from the experimental data. Their work is, however, focused on combined growth and coagulation processes. Moreover, the approach is applicable to extract growth kinetics only when the coagulation kinetics are known. Similarly, several other inverse techniques are presented in the literature see e.g., Mahoney et al. (2002); Peglow et al. (2006); Patruno et al. (2008); Chakraborty et al. (2015), however, they either may not directly be applied to cellular systems or pose significant constraints to be applicable in cell PBEs.

This work proposes a technique to extract the kinetics without any assumption regarding its structure from temporal data of a heterogeneous cell population with combined growth and breakage. The presented approach, contrary to other approaches, does not require temporal information regarding the density distributions of mother and daughter cell subpopulations.

2. METHODOLOGY

2.1 Moment Form of the PBE

We first derive the zeroth and first moment forms of (1) (Randolph and Larson (1988); Ramkrishna (2000)). For this, we integrate both sides of (1) with respect to the size and apply the boundary condition (2) to get

$$\frac{d\mu_0}{dt} + \int_0^\infty \Gamma(v)n(v,t)dv = 2\int_0^\infty \int_v^{v_{max}} \Gamma(u)P(v,u)n(u,t)dudv. \quad (4)$$

Here, μ_0 means zeroth moment i.e. the total number of cells. By reversing the order of integration in (4), we get

$$\frac{d\mu_0}{dt} + \int_0^\infty \Gamma(v)n(v,t)dv = 2\int_0^\infty \int_0^u \Gamma(u)P(v,u)n(u,t)dvdu. \quad (5)$$

Then by applying the normalization condition (3) in (5), we get the zeroth moment equation for CSD,

$$\frac{d\mu_0}{dt} = \int_0^\infty \Gamma(v)n(v,t)dv. \quad (6)$$

Similarly, for the first moment form, we multiply (1) with the size v, integrate it with respect to v, and reverse the order of integration to get

$$\frac{d\mu_1}{dt} = -\int_0^\infty v\frac{\partial}{\partial v}\left[G(v)n(v,t)\right]dv, \quad (7)$$

where, μ_1 denotes the first moment i.e. the total volume. We use integration by parts in (7) to have

$$\frac{d\mu_1}{dt} = \int_0^\infty G(v)n(v,t)dv. \quad (8)$$

Equations (6) and (8) are zeroth and first moment forms of (1) and we intend to solve them directly to get $G(v)$ and $\Gamma(v)$. This means that we intend to approximate the derivatives $d\mu_0/dt$ and $d\mu_1/dt$ from the computed/measured $n(v,t)$ and use an optimization method to find the optimal approximations of $G(v)$ and $\Gamma(v)$.

2.2 Discrete form of the forward problem

We assume that the cell size distribution over time i.e. $n(v,t)$ is available for k time points and l size classes of cells. This means that we have k number of cell size distributions where each distribution comprises of l size classes. We use this information to compute $d\mu_0/dt$ and $d\mu_1/dt$ numerically (Peglow et al. (2006)). Equations (6) and (8), can be written element-wise in discrete form as

$$m_{i,0} = \sum_{j=1}^{l} n_{ij}b_j, \quad (9)$$

$$m_{i,1} = \sum_{j=1}^{l} n_{ij}g_j. \quad (10)$$

Here, n_{ij} is the j^{th} element of the i^{th} CSD, $m_{i,0}$ and $m_{i,1}$ are the i^{th} elements in numerical approximation of $d\mu_0/dt$ and $d\mu_1/dt$, respectively, and, b_j and g_j are the j^{th} elements in breakage and growth rates, respectively. Equations (9) and (10) can be written, respectively, in matrix form as

$$\mathbf{m}_0 = \mathbf{N}\mathbf{b}, \quad (11)$$