

MICROBIAL POPULATION DYNAMICS INVESTIGATION

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Abstract

A rigorous description of the features of the growth of microorganisms in biochemical processes was derived by general population balance modelling. It is assumed that only one property is needed to describe the state of a cell the mass m . These cells have various ages masses and so on, which need to be taken into account. This paper illustrates a complex situation where growth as well as birth and death processes must be account for. The obtained results have shown rigorous microbial population model and microbial broth state transition. This work is the first report in the literature showing population transition function determination method.

Key words: population transition, microbial kinetics, birth and death processes

Introduction

A rigorous microbial description of the kinetics and features of the growth of microorganisms in biochemical processes falls naturally into the framework of general population balance methods¹. The cells (microorganisms) have various ages, masses, and so on, which need to be taken into account. A classical description of microbial study has been given by Tsuchiya, Fredrickson and Aris². Nursevin Oztop et. al.³ were studied diffusion phenomena in hydrogels and fluids in ethyl alcohol fermentation. Modelling microbial growth kinetics has been given by Beyenal and Chen^{4,5}. Transition state population in simple protein model has been studied by Ozkan et al⁶. Savkovic-Stevanovic was studied complex models which including general population balances and probabilistic projection⁷⁻¹⁰. This paper is intended to illustrate a complex situation where growth as well as birth and death processes must be accounted for. These results illustrated the feasibility of using population modelling in microbial dynamics investigation.

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Dynamic Population Model Development

It is assumed that only one property is needed to describe the state of a cell the mass m . In well mixed and constant fermentation volume the macroscopic population balance has been derived,

$$\frac{\partial \psi}{\partial t} + \frac{\partial}{\partial \xi_1} (v_1 \psi) - (B - D) = -\frac{1}{\bar{t}} \psi \quad (1)$$

for $\xi_i = m$

$$\frac{\partial \psi}{\partial t} + \frac{\partial}{\partial m} (v_1 \psi) - (B - D) = -\frac{1}{\bar{t}} \psi \quad (2)$$

where D means death, B is birth, m means mass of cells, ψ is a distribution function and $\bar{t} = v/Q$ means holding time. The function $\psi(m,t)dm$ is representative of number of cells per volume at time t with mass between m and $m+dm$ and can be further broken down into:

$$\psi(m,t) = N(t)f(m,t)dm \quad (3)$$

where $N(t)$ total number of cells per volume at time t , and $f(m,t)$ fraction of cells at time t with mass between m and $m + dm$.

The various birth and death processes were considered. Birth occurs by division of larger cells and can be formulated as follows. Let, $X(m,c)dt$ defines fraction of cells of mass m at time t that divide in time t to time $t+dt$, where c is concentration substrate (nutrient in the vessel environment), and $p(m,m')dm$ is fraction of daughter cells of mass m to $m+dm$ obtained from a cell of mass m' at fission. Thus, the rate at which daughter cell of mass m to $m+dm$ are obtained per mass of cells at age m' is

$$\text{Population_transition_function} = 2X(m',t)p(m,m') \quad (4)$$

where the factor 2 results from the assumption that only binary fission takes place. The total number of cells of mass m obtained from fission is then found by multiplying Eq.(4) by the number of cells of age m' , $\psi(m',t)dm'$ and then integrating for all values $m' > m$.

$$B = 2 \int_m^{\infty} \psi(m',t)X(m',t)p(m,m')dm' \quad (5)$$

which daughter cells of mass m to $m+dm$ are obtained per mass of cells at age m' is multiplied by factor 2 results from the assumption that only binary fission takes place. The various death processes was determined. The rate of fission of cells of mass m to smaller cells is

$$X(m,t)\psi(m,t) \quad (6)$$

A certain number of cells die without fission (true biological death) and if we define $T(m,c)dt$ as fraction of cells of mass m at time t that die in time t to time $t+dt$ then biological death is

$$D_{\text{Biological death}} = T(m, c)\psi(m, c) \quad (7)$$

Also, the rate of increase of cells of mass m is a function of m and substrate concentration in the environment $v_1(m, c)$. Thus, substituting Eqs.(5), (6) and (7) into Eq.(2) yields:

$$\frac{\partial \psi}{\partial t} + \frac{\partial}{\partial m}(v_1 \psi) = 2 \int_m^{\infty} \psi(m', t) X(m', t) p(m, m') dm' - \left\{ \frac{1}{t} + X(m, t) + T(m, t) \right\} \psi(m, t) \quad (8)$$

If separate macroscopic population balance is made on the total number of cells present $N(t)$ it would have the form

$$\frac{\partial N}{\partial t} + D - B = -\frac{1}{t} N \quad (9)$$

where

$$N(t) = \int_0^{\infty} \psi(m, t) dm \quad (10)$$

The birth term includes all fissions and has the form

$$B = \int_0^{\infty} X(m, c) \psi(m, t) dm \quad (11)$$

Also, the death term only includes all actual biological deaths since cells fission are still somewhere in the total system:

$$D = \int_0^{\infty} T(m, c) \psi(m, t) dm \quad (12)$$

$$\frac{\partial N}{\partial t} = \int_0^{\infty} X(m, c) \psi(m, t) dm - \frac{N}{t} - \int_0^{\infty} T(m, c) \psi(m, t) dm \quad (13)$$

Any daughter cell at division will have a mass between zero and the mass of its parent cell.

Microorganisms Growth

In treating microbial growth kinetics as a practical matter process analysis can extend to the determination of the moments. The zeroth moment N , "segregated unstructured" models, has given in Eq.(13). The first moments are termed "distributed" models and consist of evaluating

$$\mu_c \equiv C_m = \int_0^{\infty} m \psi(m, t) dm \quad (14)$$

where m is cell mass. C_m can be found from Eq.(8) using the definition Eq.(2).

$$\frac{dC_m}{dt} = \int_0^{\infty} v_1(m, c) \psi(m) dm - \frac{C_m}{t} - \int_0^{\infty} mT(m, c) \psi(m) dm \quad (15)$$

where $T(m, c)$ is true biological death function.

The relation

$$\int_0^{m'} mp(m, m') dm = \frac{m'}{2} \quad (16)$$

where $p(m, m')$ is defined in Eq.(4), representing the fact that for binary fission the mean daughter cell size must be one half of the original, was used to derive Eq.(15). The growth function was obtained from the postulate that for single cells the rate of cell mass increase is proportional to the surface area and the rate of decrease to the cell as:

$$net_rate = \phi S - \mu_c m \quad (17)$$

where S is cell surface area/unit volume, ϕ is cell surface flux = $\left(\frac{\mu C}{K + C}\right)$, (commonly used Michaelis-Menton form) and μ_c, μ, K are constants. The surface area for rod-shaped cells (neglected ends) is $S = 2m/R\rho$ where R is radius of cell and ρ is density of cell.

Experimental Data

Experimental data for fermentation broth have been taken from Cooney and Swartz¹¹. The Hansenula polymorpha DL-1 was growing up in examined fermenter limited with methanol.

Computational Procedure

Partial differential equations solution was performed by software package PDES. Microbial growth was simulated for different contour conditions. Some of these results are shown in Figure1 -Figure5. $\psi(m, t)$ was determined by software package PDES for numerical solutions of partial differential equations¹².

Determination of the Populations Transition State

Population transition state was determined from the total number of cells present, $N(t)$ and the number of cells per volume at time t with mass m , $\psi(m, t)$ Eqs.(10) and (13).

The population transition function which was determined from Eq.(15) and (16) according to definition Eq.(4) has shown in Figure 5.

Results and Discussion

Various states of microbial media of the Hansenula polymorpha DL-1 are shown in Figure -Figure 4. Microbial distribution functions vs time for lower cells mass concentration are shown in Figure1 and Figure 2. Figure 3 shows distribution function $\psi(m, t)$ for middle cells mass concentrations. Distribution functions vs. time for higher cells mass concentrations are shown in Figure 4. Population transition function vs. mass of cells is shown in Figure 5.

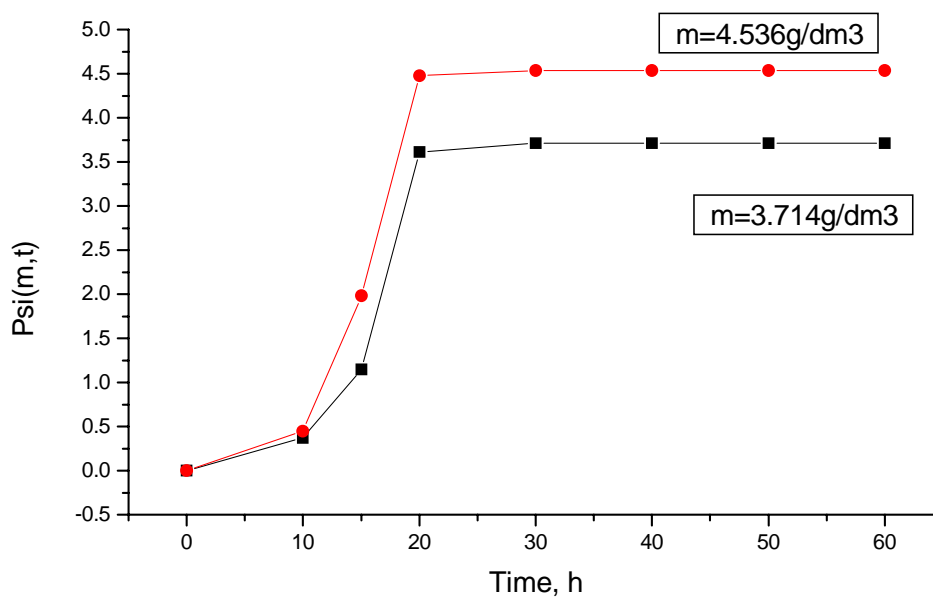


Figure 1. Cells mass distribution function with time to $m=3.714\text{g/dm}^3$ and $m=4.536\text{g/dm}^3$

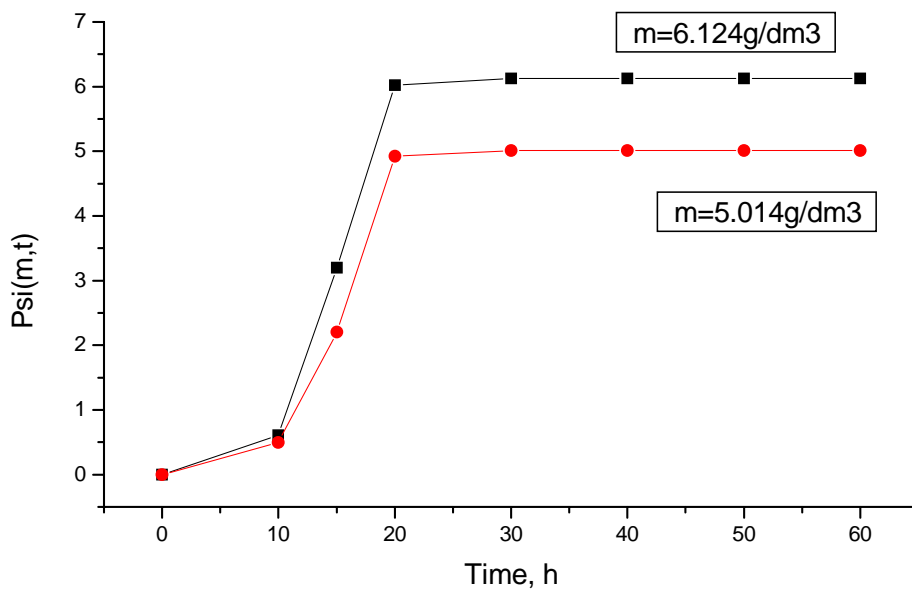


Figure 2. Cells mass distribution with time to $m= 5.014\text{g/dm}^3$ and $m=6.124\text{g/dm}^3$

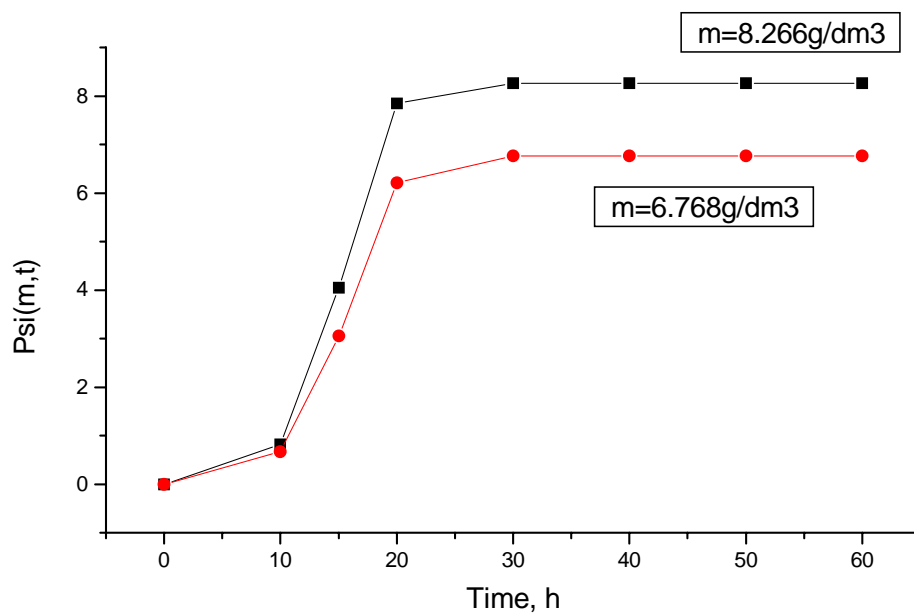


Figure 3. Cells mass distribution with time to $m=6.768\text{g/dm}^3$ and $m=8.266\text{g/dm}^3$

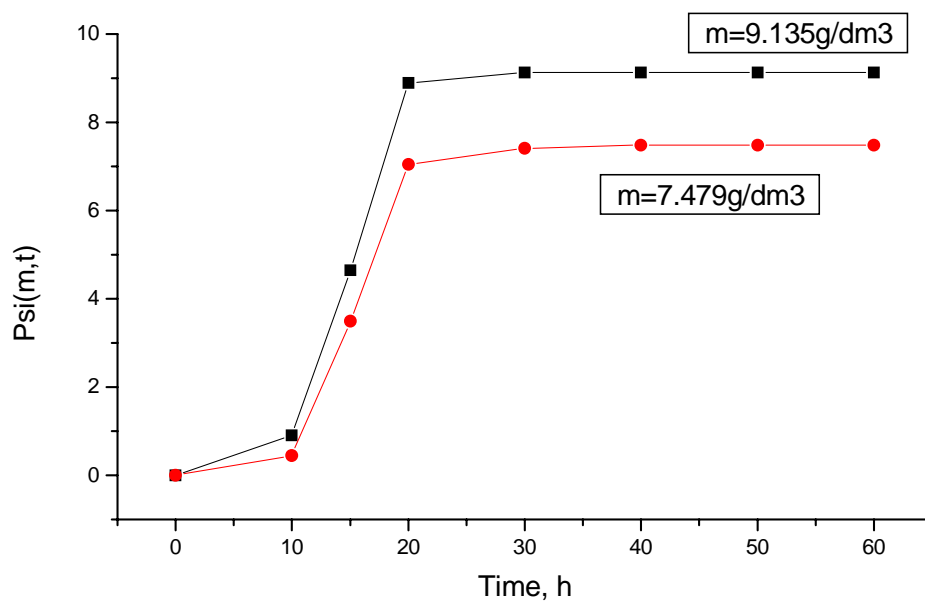


Figure 4. Cells mass distribution with time to $m=7.479\text{g/dm}^3$ and $m=9.135\text{g/dm}^3$

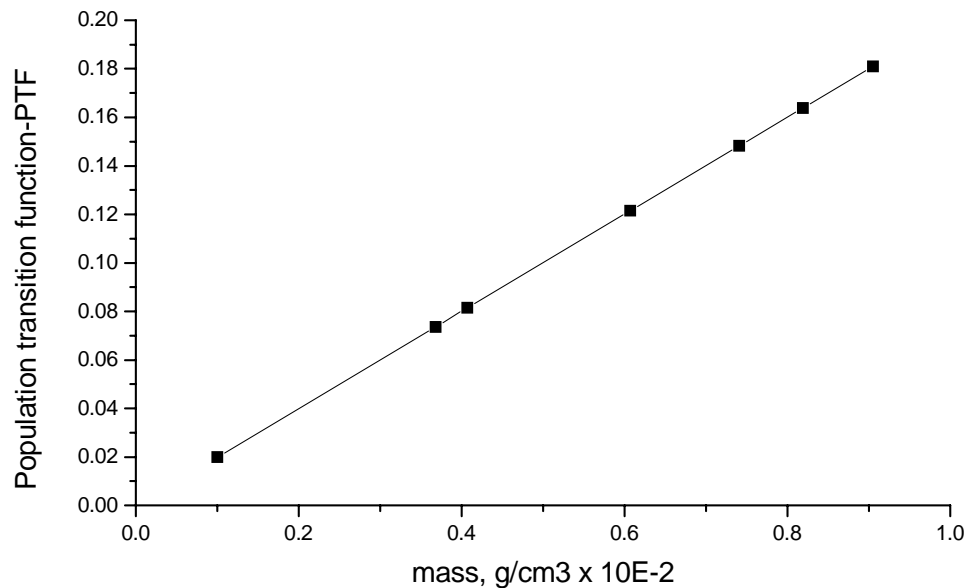


Figure 5. Population transition function vs. mass of cells

Conclusions

This study was intended to illustrate a complex microbial situation where growth as birth and death processes must be account for. The paper has shown the microbial population model for microbial processes parameters determination. The microbial population transition state was simulated for growing up the *Hanseula polymorpha* DL-1. The cells which have various ages of masses was taken into account. This work is the first report in the literature showing population transition function determination method.

Symbols

B	birth
C	substrate concentration
C_m	mass concentration
D	death
$f(m,t)$	fraction of cell mass m at time t
K	constant
m	mass of cells
N	total number of cells
$p(m,m')$	fraction of daughter cells from a cell of mass m' at fission
S	surface area/unit volume
T	time
t'	holding time

$T(m,c)$	true biological death function
v_1	rate
X	fraction

Greek Symbols

ϕ	cell surface flux
ξ	property
μ_c, μ	specific grow rate
$\psi(m,t)$	distribution function
ρ	density

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