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ORIGINAL PAPER

# Mixing characteristics and liquid circulation in a new multi-environment bioreactor

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**Abstract** The theoretical and experimental aspects of the hydrodynamics and mixing in a new multi-environment bioreactor that uses the air-lift design were investigated. This study focused on the mixing characteristics, residence time distribution, liquid circulation between three zones of aerobic, microaerophilic and anoxic, and liquid displacement in the bioreactor at influent flow rates of 720–1,450 L/day and air flow rates of 15–45 L/min. The theoretical analysis of liquid displacement led to the estimation of the specific rate of liquid discharge from the bioreactor at any given influent flow rate, and the number of liquid circulations between various bioreactor zones before the discharge of a given quantity of wastewater. The ratio of mean residence time to the overall hydraulic retention time ($t_m$/HRT) decreased with the increase of air flow rate at any given influent flow rate, and approached unity at higher air flow rates. Mixing was characterized in terms of the axial dispersion coefficient and Bodenstein number, demonstrating a linear relationship with the superficial gas velocity. A correlation was developed between the Bodenstein number and the Froude number. The study of liquid circulation between the zones showed that less than 1.5 % of the circulating liquid escapes circulation at each cycle and flows towards the outer clarifier, while the percentage of escaped liquid decreases with increasing air flow rate at a given influent flow rate. The specific rate of liquid discharge from the bioreactor increased from 0.19 to 0.69 $h^{-1}$ with the increase of air and influent flow rates from 15 to 45 L/min and 500 to 1,450 L/day, respectively. Under the examined operating conditions, mixed liquor circulates between 364 and 1,698 times between the aerobic, microaerophilic and anoxic zones before 99 % of its original volume is replaced by the influent wastewater.

**Keywords** Multi-environment bioreactor · Liquid circulation · Residence time distribution

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## Introduction

Multi-zone biological treatment systems have been successfully used for effective removal of carbonaceous contaminants and considerable reductions in the concentration of nitrogenous and phosphorus compounds [1, 2]. These technologies often use unconventional design and operation strategies, mostly in an effort to accommodate different environmental conditions, including aerobic, anoxic and anaerobic that are needed for the removal of organic and inorganic contaminants, while reducing the overall volume of treatment system. Many multi-zone technologies are designed based on the concept of airlift reactors and benefit from operational strategies that were shown to achieve enhanced nitrification/denitrification for nitrogen removal [3]. An example of airlift reactors used in wastewater treatment applications is the biofilm airlift suspension reactor (BAS) that has shown high ammonia conversion

rates up to 5 kg N/m$^3$ per day [4, 5]. Nitrification using biofilm nitrifying biomass in a BAS reactor coupled with denitrification by suspended biomass in a chemostat was reported by van Benthum et al. [6]. Biofilm airlift suspension reactors benefit from high oxygen transfer rates combined with a high specific area due to the use of small suspended carrier particles for biofilm formation.

The present paper reports on the mixing characteristics and liquid displacement in a multi-zone and multi-environment bioreactor that is incorporated in a new wastewater treatment technology, named BioCAST. This technology is designed for simultaneous removal of carbon, nitrogen and phosphorus along with the separation of solids from liquid while producing less sludge (biosolids) compared to the conventional technologies. The technology uses two interlinked bioreactors containing four zones with different environmental conditions of aerobic, microaerophilic, anoxic and anaerobic for biological treatment, as well as two clarification zones and a filtration unit for solid–liquid separation (Fig. 1). The different environmental conditions, defined based on their respective concentrations of dissolved oxygen (DO) and oxidation–reduction potential (ORP), support the growth and proliferation of a diversified group of microorganisms that are involved in the removal of organic contaminating compounds as well as the polluting inorganic nutrients.

The BioCAST technology owes its novelty largely to the integration of an external clarifier, isolated from the circulating flow between the aerobic, microaerophilic and anoxic zones, and the enlarged bottom section of the first bioreactor, forming an anoxic zone. The establishment of multiple zones with different environmental conditions inside the first bioreactor and the continuous circulation of mixed liquor containing the wastewater and microbial biomass between the aerobic, microaerophilic and anoxic zones are among the special features of design and operation of this technology. Liquid circulation between the aerobic and an oxygen-depleted zone is a common feature of wastewater treatment technologies that use the airlift design. However, in the BioCAST system, the presence of an additional oxygen-depleted zone (anoxic) at the bottom of aerobic and microaerophilic zones implies that the mixed liquor circulates between three zones, and the wastewater contaminants are exposed to three different environmental conditions with a wide range of oxygen concentrations during each cycle that takes only a few minutes. The presence of bottom anoxic zone also contributes to the accumulation of solid particles that escape circulation in the mixed liquor and settle to the bottom of this zone.

We previously reported on principle hydrodynamic characteristics of the first bioreactor of BioCAST technology including the overall gas holdup, gas holdup and linear liquid velocities in the riser and downcomer, mean liquid circulation time, liquid circulation velocity and overall volumetric oxygen transfer coefficient [7]. Those studies showed that liquid circulation velocity, gas holdup and overall volumetric oxygen transfer coefficient increase with the increase of superficial gas velocity, while the mean circulation time decreases with the increase of superficial gas velocity. The hydrodynamic characteristics of the first bioreactor were also investigated by computational fluid dynamics (CFD) analysis which explored the effects of varying operating conditions and geometries on circulation time, mean vertical velocities and the qualitative characteristics of flow patterns [8]. The CFD analysis showed the importance of the geometry of bioreactor around the base of the clarifier in controlling the smoothness of flow in the clarifier that has a significant impact on the settling of solids and the quality of effluent. The treatment performance of this multi-environment technology in the removal of organic and inorganic contaminants has also been evaluated [9, 10].

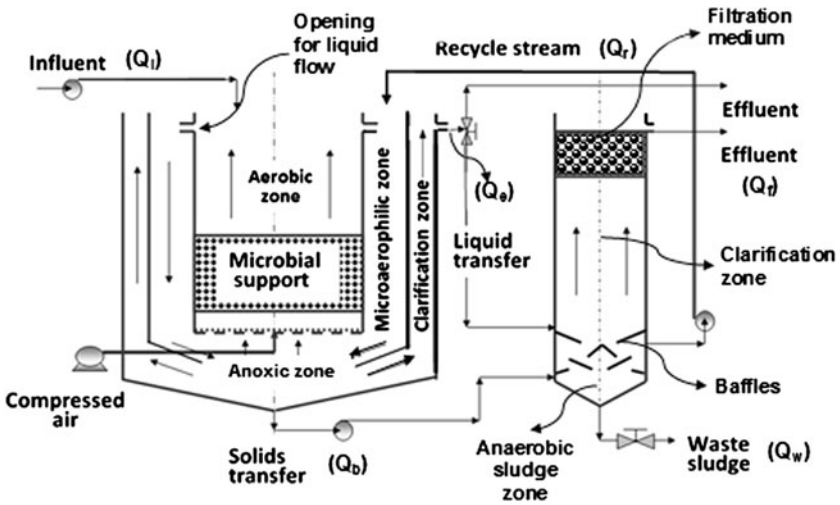


**Fig. 1** Schematic diagram of the multi-zone BioCAST technology

In the present study, liquid mixing and circulation between the three zones in the first bioreactor of the BioCAST technology, as well as residence time distribution and fractional displacement of liquid were investigated in an effort to determine the following:

- Impact of operating conditions on residence time distribution.
- Comparison of residence time with the overall hydraulic retention time under different operating conditions.
- Required time for the replacement of a given content of bioreactor by the influent wastewater.
- Number of times that a volume of wastewater added to the bioreactor circulates between various zones before a given fraction of its original volume leaves the bioreactor and flows towards the clarifier.
- Fraction of liquid that escapes circulation at every cycle and flows towards the clarifier.
- Time of a single liquid cycle between the aerobic, microaerophilic and anoxic zones, leading to the estimation of hydraulic retention time in each zone during one cycle of liquid.

## Description of the BioCAST technology

As shown in Fig. 1, the BioCAST technology contains two interlinked bioreactors. The first bioreactor contains three zones of aerobic, microaerophilic and anoxic for biological treatment, and a clarification zone around and outside the microaerophilic zone for solid–liquid separation. The second bioreactor contains an anaerobic zone at the bottom, a sedimentation zone in middle and an upper filtration zone for additional separation of solids from liquid. The anaerobic zone contains baffles that are connected to a slow rotating shaft in order to mix the sludge (biosolids), facilitate the release of entrapped gases and restrict the upward movement of sludge. Arrows in Fig. 1 show the direction of liquid flow in various zones. The effluent can be withdrawn from either bioreactor one or two, depending on the quality of effluent. The first bioreactor, investigated in this study, is designed based on the concept of airlift reactors. The mixed liquor flows upward in the aerobic zone that serves as the riser and downward in the microaerophilic zone (downcomer) on a continuous-flow basis. The aerobic zone, located in the centre of the first bioreactor, contains air diffusers at the bottom of the zone for the introduction of air. The air bubbles mix the liquid and its content of microorganisms and provide oxygen for the aerobic biological processes that take place in this zone. Aeration also produces circulation of mixed liquor between the aerobic zone and its adjacent microaerophilic and anoxic zones that are located at the sides and under the aerobic zone, respectively. The three zones of aerobic, microaerophilic and anoxic are defined based on their respective dissolved oxygen (DO) concentration and oxidation–reduction potential. During the treatment operation, the DO concentration is commonly controlled at 4–6 mg/L in the aerobic zone, 0–2 mg/L in the microaerophilic zone and zero in the anoxic zone.

A microbial support is installed in the centre of the aerobic zone for the formation of fixed-film microbial culture. The custom-built microbial support consists of two concentric cylindrical structures made out of stainless steel and wrapped in geotextile. This microbial support was shown not to interfere with the mixing and liquid circulation, and does not affect the hydrodynamic properties of the bioreactor. The presence of biofilm in the aerobic zone enhances the growth and proliferation of slow-growing nitrifiers that are needed for nitrogen removal, while the suspended microorganisms which consist mostly of heterotrophs continue to accumulate inside the circulating mixed liquor. Most of the organic carbonaceous contaminants are removed in the aerobic zone. The nitrification process, a part of the biological nitrogen removal process that transforms ammonia nitrogen into nitrite and nitrate, also takes place in this zone. The microaerophilic and anoxic zones are employed for the denitrification process to remove nitrogen by transformation of the generated nitrite and nitrate into nitrogenous gas. The anaerobic zone in the second bioreactor is used for the digestion of biosolids transferred from the anoxic zone and production of volatile fatty acids that are sent back to the first bioreactor with the recycle stream for use during the denitrification and phosphorus removal processes. Phosphorus removal is accomplished by the enhanced phosphorus removal process, benefitting from the presence of aerobic and anaerobic conditions. The simultaneous removal of carbon, nitrogen and phosphorus in the examined multi-zone treatment system was shown before [9, 10]. Hence, the examined treatment system has the capacity to remove carbonaceous, nitrogenous and phosphorus contaminants from wastewaters originating from municipal, industrial or agricultural activities.

During the operation of this treatment system, raw wastewater is introduced into the first bioreactor from the top of aerobic or microaerophilic zone. The recycled liquid from the anaerobic zone in the second bioreactor is added to the top of microaerophilic zone. The upward flow of liquid in the aerobic zone carries the influent wastewater towards the microaerophilic zone where it flows downward towards the anoxic zone through openings (apertures) between the aerobic and microaerophilic zones. Since there is no accumulation of liquid inside the bioreactor, a volume of liquid equal to the influent volume [raw wastewater ($Q_i$) plus the recycled liquid from the second bioreactor ($Q_r$)]

leaves the system through the clarifier zone ($Q_e$) [considering negligible loss through the bottom of anoxic zone ($Q_b$)], while the rest of liquid flows towards the centre of bioreactor where it is directed towards the aerobic zone. This flow pattern, verified during the previous investigations of this treatment system, continues throughout the course of operation of first bioreactor, creating a continuously circulating liquid between the aerobic, microaerophilic and anoxic zones.

The continuous circulation of mixed liquor between the three zones is an important feature of this technology that contributes to the accumulation of microbial biomass in the circulating mixed liquor, producing a high concentration of a diversified group of microorganisms and high mean cell residence times (MCRT) or solids retention times (SRT) which have been shown to contribute to microbial adaptation as well as a low generation of sludge (biosolids) [9]. Liquid circulation between various zones also exposes the contaminants to three different environmental conditions of aerobic, microaerophilic and anoxic during each cycle, and has shown to promote denitrification of the produced nitrates and nitrites which are intermediate compounds during nitrogen removal processes, shortly after their formation, thus preventing the accumulation of these inorganic contaminants that may exert inhibitory effects on microbial metabolism. The retention time of circulating liquid in the three zones of the first bioreactor can be controlled by adjusting the number and size of the openings (apertures) between the aerobic and microaerophilic zones, and the input air flow rate. Liquid circulation between the aerobic, microaerophilic and anoxic zones is an effect caused by the difference in the fractional gas holdup (i.e. different mean densities) that exists between the zones, creating a hydrostatic pressure difference between the bottoms of the aerobic and microaerophilic zones which acts as a driving force for liquid circulation. These features of the treatment system have been established during the treatment of wastewater which produced solid retention times (SRT) of 12–138 days and an average biomass yield of 3.7 % during 225 days of operation [9]. The high SRT and biomass concentration result from the accumulation of suspended biomass in the circulating mixed liquor and the growth of attached biomass on the support material in the aerobic zone of the treatment system. During liquid circulation, the heavy bioflocs containing microorganisms, un-hydrolyzed particulate organic and inorganic matter, as well as solid inert material precipitate to the bottom of anoxic zone where they are subsequently hydrolyzed and degraded, or transferred to the anaerobic zone in the second bioreactor for further digestion. However, the dispersed microbial biomass and light bioflocs remain inside the mixed liquor as it circulates between the three zones, and continue to accumulate. Liquid circulation also controls residence time distribution which enables the characterization of mixing and flow pattern inside the bioreactor, and facilitates the comparison of bioreactor behaviour with the ideal models [11].

## Theoretical considerations

### Residence time distribution (RTD)

The residence time distribution (RTD) of a reactor is the probability distribution function that defines the amount of time a fluid element spends inside the reactor. The RTD of a reactor is expressed by the function $E(t)$ (Eq. 1). This function can be graphically illustrated from a pulse-input tracer test by dividing the effluent concentration of tracer at any given time, $C(t)$, by the area under the concentration–time curve [12]. The mean residence time inside the reactor can be calculated from the first moment of the $E(t)$ function (Eq. 2):

$$E(t) = \frac{C(t)}{\int_0^\infty C(t)dt} \tag{1}$$

$$t_m = \int_0^\infty tE(t)\mathrm{d}t. \tag{2}$$

By substituting Eq. (1) into Eq. (2), the mean residence time can be calculated from the following equation [12, 13]:

$$t_m = \frac{\int_0^\infty tC(t)\mathrm{d}t}{\int_0^\infty C(t)\mathrm{d}t}. \tag{3}$$

The variance or square of the standard deviation of RTD is represented by the second central moment (Eq. 4):

$$\sigma_t^2 = \int_0^\infty (t - t_m)^2 E(t)\mathrm{d}t. \tag{4}$$

The value of variance that has a unit of time squared is an indication of the RTD spread. The dimensionless variance can be obtained by dividing the variance ($\sigma_t^2$) by the square of mean residence time ($t_m$). This parameter measures the breadth of distribution, independent of the magnitude of $t_m$, according to the following equation:

$$\sigma^2 = \frac{\sigma_t^2}{t_m^2}. \tag{5}$$

The value of $\sigma^2$ may be used to predict the mixing behaviour of reactors. For the ideal plug flow reactor (PFR) $\sigma^2 = 0$ while for ideal continuous stirred tank reactors (CSTR) $\sigma^2 = 1$. The dimensionless variance of most reactors falls somewhere between the two limits of zero and one [11, 12].

### Axial dispersion coefficient and Bodenstein number

The mixing characteristics of liquid were further evaluated by examining axial mixing in the bioreactor, characterized by the axial dispersion coefficient and Bodenstein number which represents the ratio between the bulk movement of fluid and the mixing effects of the axial dispersion coefficient. The Bodenstein number for the liquid phase (Bo) and the Bodenstein number for the gas–liquid mixture ($Bo_{LG}$) were calculated from Eqs. 6 and 7 where Ez is the axial dispersion coefficient, $V_L$ is the averaged linear velocity of liquid in the riser and downcomer, $L_c$ is the length of the liquid circulation loop, $U_G$ is the superficial gas velocity and $d$ is mean bioreactor diameter [14, 15]:

$$\mathrm{Bo} = \frac{V_L L_c}{E_z} \tag{6}$$

$$\mathrm{Bo_{LG}} = \frac{U_G d}{E_z} \tag{7}$$

The axial dispersion coefficient (Ez) was estimated from Eq. 8 as recommended by Merchuk et al. [14]:

$$E_z = K d \left(\frac{U_G}{\varepsilon}\right)^n \tag{8}$$

where $K$ and $n$ are constants and $\varepsilon$ is the gas hold up. Merchuk et al. [14] suggested mean values of 0.89 for $K$ and 1.18 for $n$.

The relationship between the Bodenstein number for the gas–liquid mixture ($Bo_{LG}$) and the Froude number (Fr) was investigated using the equation suggested by Rice et al. [16] and used by Sanchez Miron et al. [15] to correlate the data for airlift reactors, as follows:

$$\mathrm{Bo_{LG}} = \beta\,(\mathrm{Fr}^{1/3})^{\lambda} \tag{9}$$

where $\beta$ and $\lambda$ are constants that can be determined from the experimental results using non-linear regression analysis. The Froude number was calculated from the following equation:

$$\mathrm{Fr} = \frac{U_G^2}{g\,d} \tag{10}$$

where $g$ is the gravitational acceleration.

### Liquid displacement

During the operation of the examined treatment system, the volume of wastewater inside the first bioreactor at any given time is defined as $Y_w$, while the volume of original mixed liquor at any given time is defined as $Y_r$. It is understood that in reality, the influent wastewater and mixed liquor are thoroughly mixed and cannot be considered as two separate streams. However, for the purpose of flow analysis and the estimation of liquid displacement, the influent wastewater that mixes with the original mixed liquor is considered as a separate entity inside the bioreactor. In addition, this analysis does not address the biological treatment of wastewater and does not distinguish between the treated and untreated wastewater. Assuming that the bioreactor volume ($V_R$) and the influent wastewater flow rate are constant, a constant percentage of the bioreactor liquid content, i.e. mixture of wastewater and original mixed liquor, leaves the bioreactor during any given period of time since there is no liquid accumulation inside the system. Therefore, the change of mixed liquor volume with respect to time ($dY_r$/dt) is directly proportional to its volume in the bioreactor. This relationship can be mathematically expressed as follows:

$$\frac{dY_r}{\mathrm{dt}} = -kY_r \tag{11}$$

or

$$Y_r = Y_i e^{-kt}. \tag{12}$$

Where:

$Y_r$ = volume of original mixed liquor at time ($t$), gradually replaced by the influent wastewater.
$Y_i$ = volume of original mixed liquor at time zero, equal to the overall bioreactor volume ($V_R$).
$k$ = specific rate of liquid discharge from the bioreactor.

The fraction of original mixed liquor volume in the bioreactor at any given time ($t$) can be expressed by the following equation:

$$\%Y_r = \frac{Y_r 100}{Y_i} = e^{-kt} \times 100. \tag{13}$$

Since $Y_r + Y_w = Y_i$, the volume of wastewater inside the bioreactor at any given time ($Y_w$) is expressed as follows:

$$Y_w = Y_i(1 - e^{-kt}). \tag{14}$$

The fraction of wastewater liquid based on the total content of bioreactor is as follows:

$$\%Y_W = \frac{Y_W 100}{Y_i} = (1 - e^{-kt}) \times 100. \tag{15}$$

Equations 11–15 demonstrate the following:

$t = 0 \Rightarrow Y_r = Y_i$, volume of original mixed liquor at time zero is equal to the bioreactor overall volume.
$t = 0 \Rightarrow Y_w = 0$, there is no wastewater inside the bioreactor at time zero and the original mixed liquor is the only liquid that occupies the bioreactor.

Similarly, the time-dependent changes in the volume of a given quantity of wastewater entering the bioreactor can

be estimated by knowing that a fraction of this wastewater continuously exits the bioreactor as part of the departing liquid, as follows:

$$Y_W = Y_{Wi}e^{-kt} \tag{16}$$

Where $Y_{wi}$ is an arbitrary volume of wastewater entering the bioreactor and the $k$ variable is the same $k$ as above because $Y_w$ and $Y_r$ are removed by the same process. The percentage of wastewater that has left the bioreactor at any given time ($t$) is expressed as follows:

$$\%\ \text{wastewater} = \frac{(Y_{Wi} - Y_W) \times 100}{Y_{Wi}} \tag{17}$$

The average time required for the proportion $Y_w/Y_{wi}$ of wastewater to be evacuated from the reactor ($t_{evac}$) is estimated by rearranging Eq. (16), as follows:

$$t = -\frac{1}{k}\ln\left(\frac{Y_W}{Y_{Wi}}\right) \tag{18}$$

$$t_{evac} = -\frac{1}{k}\int \ln(Y_w)dY_w = [Y_w \ln(Y_w) - Y_w]_{(0-1)} \tag{19}$$

$$t_{evac} = -1/k[-1 - 0] = 1/k. \tag{20}$$

The average number of liquid circulations (NOC) between the aerobic, microaerophilic and anoxic zones for a given quantity of wastewater before leaving the bioreactor is equal to:

$$\text{NOC} = \frac{1/k}{\text{HRT}_a + \text{HRT}_m + \text{HRT}_x}. \tag{21}$$

The percentage of liquid that escapes circulation between the three zones at each cycle (% $\text{liquid}_{esc}$) is estimated by making a flow balance around the first bioreactor, as follows:

$$\%\,\text{Liquid}_{esc} = \frac{Q_e + Q_b}{Q_i + Q_r + Q_a}. \tag{22}$$

Where:

$Q_a$ = circulating liquid flow rate in the aerobic zone.
$Q_i$ = flow rate of raw wastewater.
$Q_r$ = flow rate of the recycled stream from the second to the first bioreactor.
$Q_e$ = flow rate of liquid that exits the first bioreactor through the clarifier.
$Q_b$ = flow rate of liquid that exits the first bioreactor through the anoxic zone.

The theoretical value of the specific rate of liquid discharge from the reactor ($k$) at any given liquid flow rate can be obtained from the following equation:

$$k = \frac{Q_e + Q_b}{V_a + V_m + V_x}. \tag{23}$$

Where:

$V_a$ = volume of aerobic zone.
$V_m$ = volume of microaerophilic zone.
$V_x$ = volume of anoxic zone.

Assuming ideal mixing, the average time length of a single liquid cycle between the aerobic, microaerophilic and anoxic zones ($\text{HRT}_{cycle}$) is equal to the sum of hydraulic retention times in the three respective zones, estimated as follows:

$$\text{HRT}_{cycle} = \text{HRT}_a + \text{HRT}_m + \text{HRT}_x. \tag{24}$$

Where:

$$\text{HRT}_a = \frac{V_a}{Q_a} \tag{25}$$

$$\text{HRT}_m = \frac{V_m}{Q_m} \tag{26}$$

$$\text{HRT}_x = \frac{V_x}{Q_x} \tag{27}$$

and:

$$Q_i + Q_r = Q_e + Q_b \tag{28}$$

$$Q_m = Q_a + Q_i + Q_r. \tag{29}$$

Where:

$Q_m$ = circulating liquid flow rate in the microaerophilic zone.
$Q_x$ = circulating liquid flow rate in the anoxic zone.

The overall hydraulic retention time (HRT) of the bioreactor is calculated from the following equation:

$$\text{HRT} = \frac{V_R}{Q_i}. \tag{30}$$

The required time for the replacement of 90 and 99 % of the bioreactor content by the influent wastewater is calculated by rearranging Eq. 13:

$$t_{LD90\,\%} = \frac{\ln(0.1)}{-k} \tag{31}$$

$$t_{LD\,99\,\%} = \frac{\ln(0.01)}{-k} \tag{32}$$

Where $t_{(LD90\ \%)}$ is the required time for 90 % liquid displacement and $t_{(LD99\ \%)}$ is the required time for 99 % liquid displacement. The number of liquid circulations (NOC) for 90 and 99 % replacement of the bioreactor content is estimated by the following equations:

$$\text{NOC}_{LD\,90\,\%} = \frac{t_{LD\,90\,\%}}{\text{HRT}} \tag{33}$$

$$\text{NOC}_{LD\,99\,\%} = \frac{t_{LD\,99\,\%}}{\text{HRT}}. \tag{34}$$

## Methodology

### Fabrication of the bioreactors

The bioreactors used in this study were made from PVC and had a volume of 185 and 12 l, respectively. The volumes of aerobic, microaerophilic and anoxic zones were 17, 61 and 22 l, respectively, while the volume of clarification zone in the first bioreactor was 85 l. The reactors had a height of 1.13 m. Air was introduced in the aerobic zone through three custom-built air diffusers installed 8 cm above the bottom of this zone, each having 21 holes with the diameter of 1 mm. There were eight openings (apertures) between the aerobic and microaerophilic zones with adjustable sizes of 0.63–2.54 cm (0.25–1 in).

### Determination of residence time distribution

The residence time distribution (RTD) in the examined bioreactor was determined experimentally by the pulse-input tracer technique [13, 17, 18]. The experiments were conducted using eight openings at the size of 2.54 cm (1 in) between the aerobic and microaerophilic zones. At time zero, the bioreactor was filled with tap water and aerated through the diffusers located at bottom of aerobic zone. Tap water was added to the system at a constant flow rate from the top of aerobic zone while a flow equal to that of the added water continuously left the bioreactor (no liquid accumulation). Water-soluble Quinoline Yellow (QY, 95 %) was used as the tracer. 1.5 g QY, dissolved in 10 mL of tap water was instantaneously injected into the system from the top of aerobic zone at time zero. Samples were taken from the bioreactor exit at predetermined intervals and were analyzed for the tracer concentration using a spectrometer (HACH model DR2800). Immediately after the injection of tracer, sampling was done every 1 min for half an hour, and then every 5 min for an additional hour. After this time, the changes in the tracer concentration were less variable and sampling was carried out every half hour until the tracer concentration inside the reactor approached zero, implying that the entire volume of reactor had been replaced by the added water. The experiments were carried out at influent water flow rates ($Q_i$) of 720, 1,000 and 1,450 L/day, and air flow rates ($Q_{air}$) of 15, 30 and 45 L/min in order to determine the impact of operating conditions on the RTD in the first bioreactor.

### Measurement of gas holdup

The overall gas holdup ($\varepsilon$) was estimated using the volume expansion technique at steady-state condition. The height of liquid in the reactor was measured with and without aeration ($h_{LG}$ and $h_L$, respectively). The overall gas holdup was calculated using Eq. (35) [18–20].

$$\varepsilon = \frac{h_{LG} - h_L}{h_{LG}} \tag{35}$$

The presence of openings between the riser and downcomer in the examined bioreactor caused air dispersion to occur mainly in the riser. Therefore, an inverted U-tube manometer was used to measure the gas holdup in the riser ($\varepsilon_r$) independently. The difference between liquid levels inside the tubes ($dh_M$) in the presence of aeration was monitored. The gas holdup in the riser was estimated from the following equation [19, 21], where dz is the elevation difference of the tubes:

$$\varepsilon_r = \frac{\rho_L}{\rho_L - \rho_G}\frac{dh_M}{dz} \tag{36}$$

$\rho_G$ and $\rho_L$ are the densities of gas and liquid, respectively.

### Measurement of linear liquid velocity in the riser and downcomer

The independent values of linear liquid velocity in the riser and downcomer were estimated using the tracer technique as described before [19, 22–24]. However, in this technique, pH was monitored by two identical pH probes placed in the downcomer; the first probe was placed 15 cm below the water level where the flow was fully developed, and the second probe was placed at the bottom of downcomer where water flows toward the riser. The pH probes were connected to two separate pH meters (Oakton, Model PD650). The pH data were transmitted to a computer by a USB IrDA converter. The travel time of tracer between the two pH electrodes whose distance was almost the same as the overall length of downcomer ($L_d$), was estimated from the difference between the first moments of first peaks from the pH probes ($t_d$) [22]. The residence time in the riser was estimated from the difference between the corresponding values of $t_c$ and $t_d$. The linear liquid velocities in the riser ($V_{Lr}$) and downcomer ($V_{Ld}$) were estimated from the ratios of their corresponding lengths to their residence times.

### Measurement of liquid displacement

The reactor was filled with tap water, and 1.5 g of water-soluble QY (95 %) used as the tracer was dissolved into the entire volume of the bioreactor. The homogeneity of the solution inside the bioreactor was verified by thorough mixing of the solution, followed by sampling from various locations inside the bioreactor. These samples were analyzed by a spectrometer (HACH model DR2800) to determine their respective concentrations of QY. The

initial concentration of QY was considered as $C_{max}$. Aeration started while tap water was added to the system at a constant flow rate from the top of aerobic zone. Since the system was operating at steady state with no liquid accumulation, liquid at a flow rate equal to that of water continuously left the bioreactor. Samples were collected every 15 min from the reactor exit for the measurement of optical density, leading to the determination of relative concentration of each sample ($C/C_{max}$) and the time-dependent changes in the percentage of mixed liquor inside the reactor ($Y_r$). Sampling continued until the percentage of original mixed liquor inside the reactor approached zero. The experiments were carried out at influent water flow rates ($Q_i$) of 500, 720, 1,000 and 1,450 L/day and air flow rates of 15, 30 and 45 L/min. All experiments were carried out using eight openings of 2.54 cm (1 in) between the aerobic and microaerophilic zones. All experiments were repeated three times, and the reported results are the average of measurements from each experiment.

## Results and discussion

### Residence time distribution (RTD): experimental versus theoretical values

Figure 2a–c illustrates the residence time distribution (RTD) in the first bioreactor of the BioCAST system at different influent water flow rates and air flow rates. The sharp initial increase followed by an exponential decrease, observed under all examined conditions, is consistent with the RTD trend in completely mixed reactors. However, in the examined treatment system, the initial increase in the residence time distribution exhibited a small delay at time zero, implying a slight deviation from the profile of an ideally mixed reactor where residence time distribution increases abruptly at time zero and decreases exponentially afterwards. The results (Fig. 2) demonstrated negligible effect of the air flow rate on the RTD for a given influent flow rate. On the contrary, the RTD varied with the influent flow rate at a given air flow rate, as shown in Fig. 3a–c. This figure shows the increase in the initial peak of RTD with the increase of influent flow rate.

The values of mean residence time in the first bioreactor under the examined operating conditions, calculated from Eq. 3, are presented in Table 1. The ratio between the mean residence time ($t_m$) and the overall HRT is also presented in this table for each operating condition, while its variations as a function of the air flow rate at different influent flow rates are presented in Fig. 4. The resulting trend shows that the ratio $t_m$/HRT decreases with the increase of air flow rate at any given influent flow rate. For all wastewater flow rates, the ratio is closest to unity at the air flow rate of 30 L/min, implying that this is the air inflow rate that optimizes mixing.

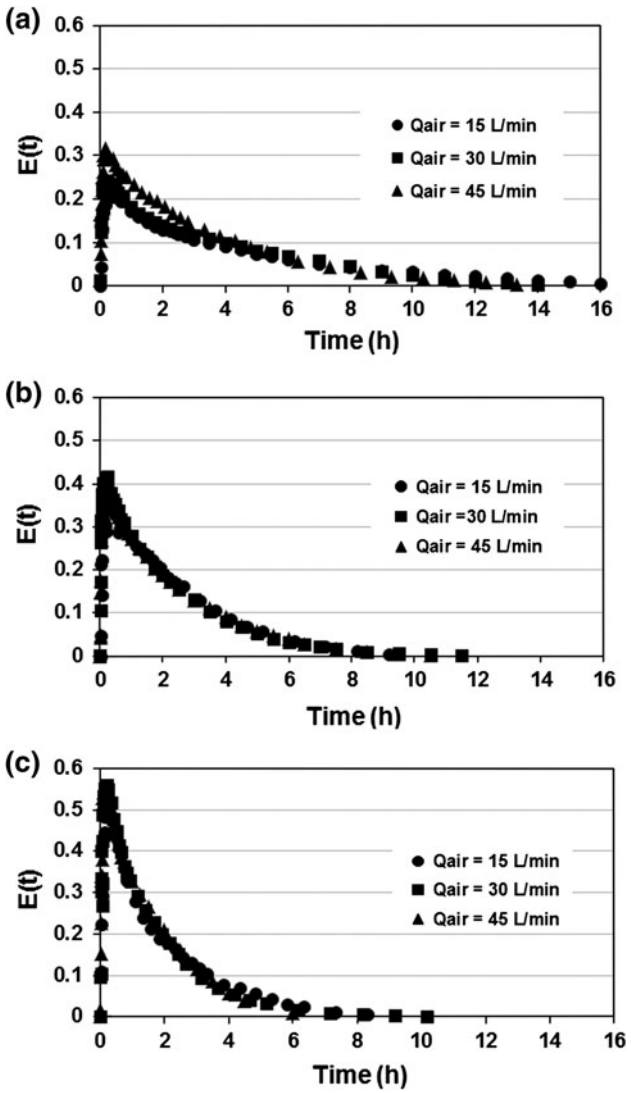


**Fig. 2** RTD in the first bioreactor at influent flow rates of **a** 720 L/day, **b** 1,000 L/day and **c** 1,450 L/day

The dependence of the dimensionless variance of RTD ($\sigma^2$) on air flow rate is illustrated in Fig. 5 for different influent flow rates. The value of this parameter ranges from 0.6 to 0.9, indicating that the RTD in the first bioreactor of the examined treatment system is close to that observed in continuous stirred tank reactors (CSTRs) which have a value equal to one, as compared to the expected value for plug flow reactors which is zero. The RTD in the examined bioreactor is particularly close to one at influent flow rate of 1,450 L/day and air flow rate of 30 L/min. Gavrilescu and Tudose [11] analyzed the residence time distribution (RTD) of liquid phase in a concentric-tube airlift reactor with liquid recirculation and showed that the liquid phase flow resembled that in the ideal plug flow with superimposed axial dispersion in the overall reactor. The flow pattern in the examined bioreactor differs from that in

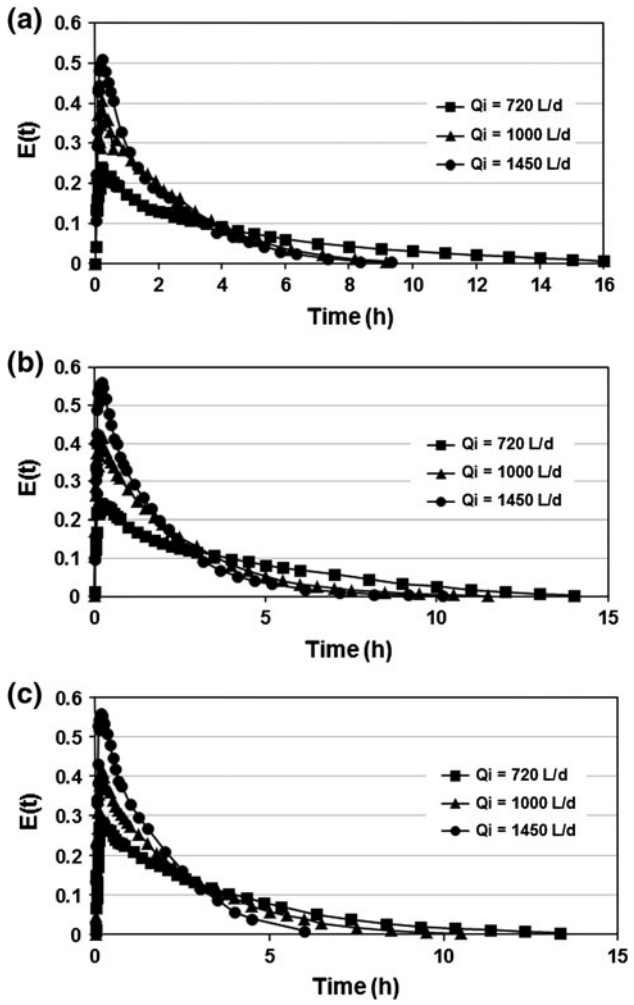


**Fig. 3** RTD in the first bioreactor at air flow rates of **a** 15 L/min, **b** 30 L/min and **c** 45 L/min

conventional airlift reactors due to the differences in the geometry of this bioreactor, including the presence of liquid flow restrictions at the top of the riser, the ratio between the diameters of riser and downcomer which is less than that commonly used in airlift reactors, and the enlarged bottom section of the bioreactor.

## Axial dispersion coefficient and Bodenstein number

The axial dispersion coefficient ($E_Z$) and Bodenstein number have been rigorously studied and found to be dependent on the mixing pattern and flow regime in the reactor [14, 15, 25]. In the examined bioreactor, the axial dispersion coefficient ($E_Z$) increased steadily with the increase of superficial gas velocity ($U_G$) up to 0.045 m/s, but remained constant with further increase of $U_G$ (Fig. 6a). A similar behaviour was reported by Merchuk et al. [14] for cylindrical spargers in concentric tube airlift bioreactors, and by Sanchez Miron et al. [15] in draft-tube airlift reactors where the axial dispersion coefficient exhibited a fast rate of increase at lower superficial gas velocities and remained practically constant at higher values of $U_G$. Figure 6b shows that the Bodenstein number for the liquid phase (Bo) and Bodenstein number for the gas–liquid mixture ($Bo_{LG}$) exhibited linear relationships with $U_G$. The values of Bo ranged from 1.3 to 5.2. It has been reported that for perfectly mixed reactors, Bo is less than 0.1 while ideal plug flow regime is established when Bo is greater than 20 [19]. Thus, the obtained values of Bodenstein number further show that the bioreactor examined in the present work exhibits a mixing behaviour that falls between the two ideal cases of perfectly mixed and plug flow regime, while being closer to perfectly mixed reactors. Weiland [26] examined the effect of the ratio of draft tube diameter to reactor diameter on hydrodynamic properties of reactors and showed that efficient mixing would require diameter ratios of 0.8 and 0.9, a range of values that are larger than that experienced in the examined bioreactor which was 0.5. The Bodenstein number for the gas–liquid mixture ($Bo_{LG}$) ranged from 0.03 to 0.15. Higher values, up to a range of 0.6–0.8, were reported by Sanchez Miron et al. [15] for a draft-tube vessel with tap water, a draft-tube vessel with sea water, a split-cylinder with tap water and a split-cylinder with sea water.

The values of constants $\beta$ and $\lambda$ in Eq. 9 were calculated by plotting $Bo_{LG}$ versus $Fr^{1/3}$ (Fig. 6c) and using non-linear regression analysis. The values of these constants were

**Table 1** Hydraulic retention time and mean residence time in the first bioreactor

| Influent flow rate $Q_i$ (L/day) | Air flow rate $Q_{air}$ (L/min) | Overall HRT (h) | Mean residence time $t_m$ (h) | $t_m$/HRT |
|---|---|---|---|---|
| 720 | 15 | 3.52 | 4.52 | 1.29 |
| | 30 | 3.52 | 3.82 | 1.09 |
| | 45 | 3.52 | 3.26 | 0.93 |
| 1,000 | 15 | 2.46 | 2.31 | 0.94 |
| | 30 | 2.46 | 2.26 | 0.92 |
| | 45 | 2.46 | 2.32 | 0.94 |
| 1,450 | 15 | 1.75 | 1.99 | 1.14 |
| | 30 | 1.75 | 1.77 | 1.01 |
| | 45 | 1.75 | 1.58 | 0.90 |

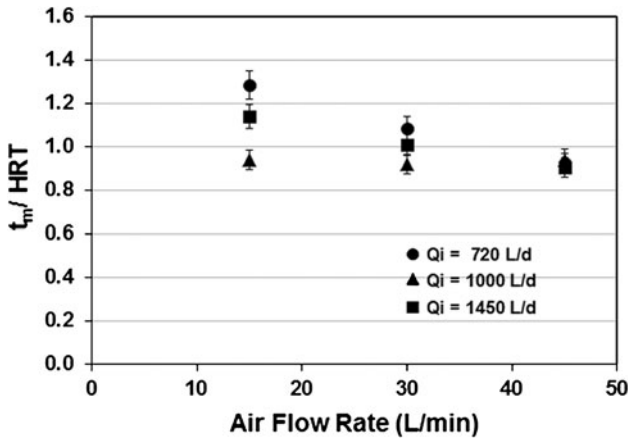


**Fig. 4** Dependence of $t_m$/(HRT) on air flow rate at different influent flow rates

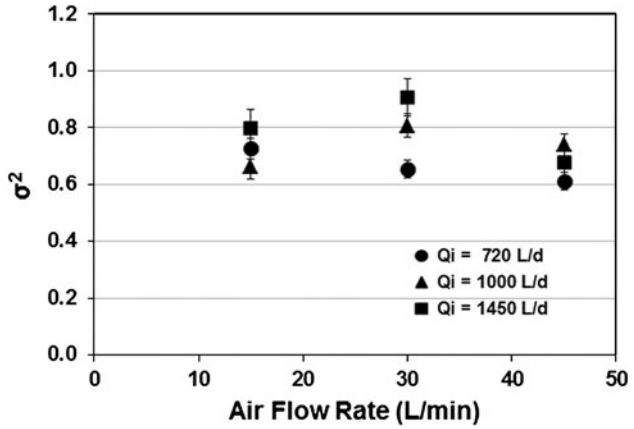


**Fig. 5** Dependence of the dimensionless variance ($\sigma^2$) of RTD on air flow rate at different influent flow rates

found to be $\beta = 1.9$ and $\lambda = 1.1$. Sanchez Miron et al. [15] reported values ranging from 3.77 to 8.28 for $\beta$ and 1.200 to 1.364 for $\lambda$ for different designs of airlift reactors and different liquid properties including draft tube vessel with tap water, draft-tube vessel with sea water, split-cylinder with tap water and split-cylinder with sea water. The different values of constants obtained for the bioreactor examined in the present work is related to the different design of this bioreactor and different mixing properties.

## Time-dependent liquid displacement and number of liquid circulations between the zones

During the treatment of wastewater, the original mixed liquor volume in the first bioreactor decreases in time as it is replaced by the wastewater that enters the bioreactor on a continuous basis. Figure 7 presents the agreement between the experimental values for the reduction of mixed liquor

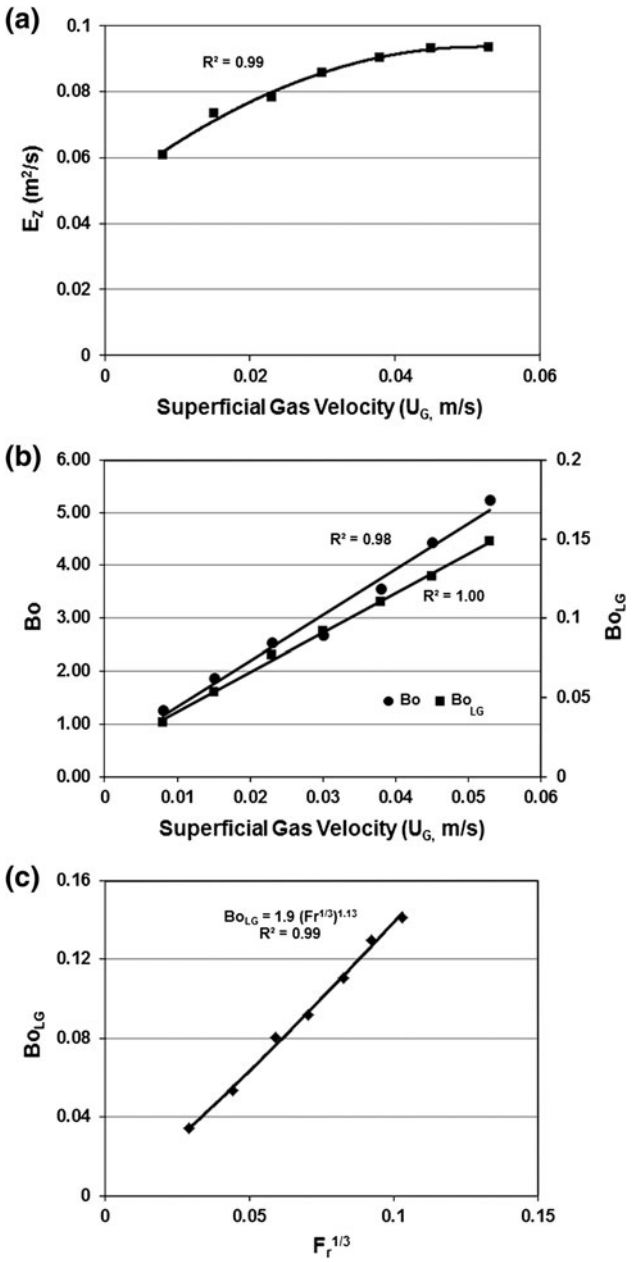


**Fig. 6** **a** Axial dispersion coefficient ($E_Z$) versus superficial gas velocity, **b** Bodenstein number in liquid phase (Bo) and Bodenstein number for the gas–liquid mixture ($Bo_{LG}$) versus superficial gas velocity, and **c** correlation between $Bo_{LG}$ and $Fr^{1/3}$ where Fr is the Froude number

volume with the theoretical values estimated from Eq. 13 at different air and influent flow rates, using a $k$ value, estimated from Eq. 23, that assumes $Q_b = 0$. The experimental values closely followed the theoretical predictions and illustrated an exponential trend for the time-dependent changes in the volume of original mixed liquor ($Y_r$). The comparison between the changes in the percentage of original mixed liquor volume ($Y_r$), estimated from Eq. 12, and the wastewater volume ($Y_w$) in the bioreactor, estimated

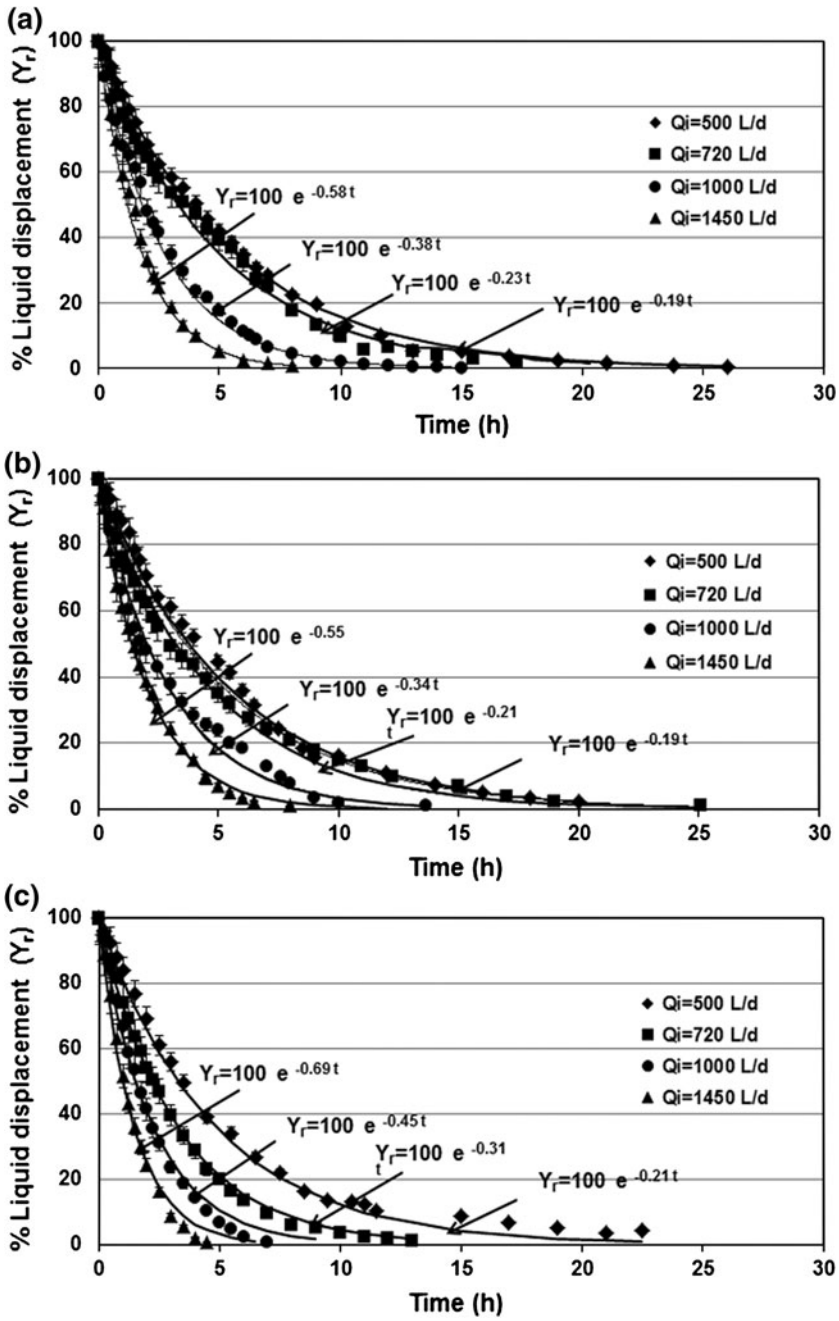


**Fig. 7** Time-dependent changes in the volume of mixed liquor ($Y_r$) at various air and influent flow rates. The experimental points are presented by markers, and the *solid lines* are drawn from Eq. 13. **a** $Q_{air} = 15$ L/min, **b** $Q_{air} = 30$ L/min, **c** $Q_{air} = 45$ L/min

from Eq. 14, with the increasing number of liquid circulations between the three zones is presented in Fig. 8 for the influent wastewater flow rate of 1,000 L/day and air flow rate of 30 L/min. It should be noted that according to the exponential function expressing the liquid displacement, it will theoretically take an infinite number of circulations for a complete (100 %) replacement of original mixed liquor with the influent wastewater in the bioreactor.

The specific rate of liquid discharge from the reactor ($k$) at each operating condition, calculated from Eq. 13 using the experimental values in Fig. 7, increased with the influent flow rate as expected (Table 2). It increased from 0.19 to 0.58 $h^{-1}$ at the air flow rate of 15 L/min, and from 0.21 to 0.69 $h^{-1}$ at the air flow rate of 45 L/min with the increase of influent flow rate from 500 to 1,450 L/day, respectively. The theoretical values of k, estimated from Eq. 23, are also presented in this table. This table shows that the percentage of liquid inside the bioreactor (mixture of mixed liquor and wastewater) that escapes circulation at each cycle and flows towards the clarification zone, calculated from Eq. 22 (assuming $Q_b = 0$), ranges from 0.25 to 1.34 %, emphasizing that less than 1.5 % of the bioreactor liquid escapes circulation between the various zones at each cycle. Moreover, the percentage of escaped liquid

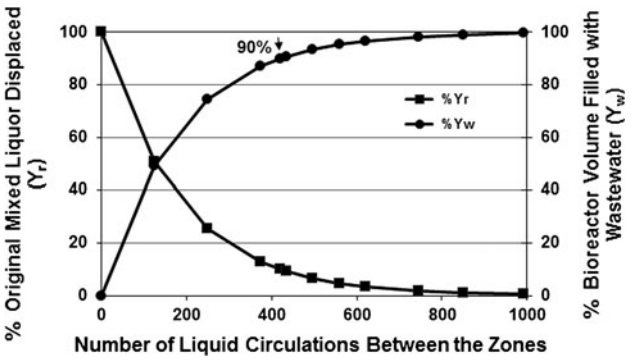


**Fig. 8** Theoretical predictions of the dependence of the percentage of original mixed liquor volume in the bioreactor ($Y_r$) and the percentage of wastewater volume in the bioreactor ($Y_w$) on the number of liquid circulations between the aerobic, microaerophilic and anoxic zones for the influent flow rate of 1,000 L/day and air flow rate of 30 L/min

increases with the increasing influent flow rate at a given air flow rate. However, at a constant influent flow rate the percentage of escaped liquid decreases with the increasing air flow rate.

The dependence of the number of liquid circulations between the aerobic, microaerophilic and anoxic zones on the influent flow rate for 90 and 99 % liquid displacement in the bioreactor were estimated from Eqs. 33 and 34 using the $k$ values from Table 2. The results are graphically presented in Fig. 9a–c for three different air flow rates of 15, 30 and 45 L/min, and demonstrate that the number of liquid circulations between the zones for the replacement of 90 and 99 % volume of the original mixed liquor decreases with the increasing flow rate of influent wastewater at a given air flow rate. At the air flow rate of 15 L/min, the number of liquid circulations between the three zones of bioreactor decreases from 521 to 182 times with the increase of influent flow rate from 500 to 1,450 L/day, before 90 % of bioreactor volume is replaced by the influent wastewater. Under this condition, mixed liquor circulations of 1,047 to 364 times are required for 99 % liquid displacement. The increase of air flow rate to 45 L/min increases mixed liquor circulations to the range of 261–859 for 90 % liquid replacement, and to 524–1,698 for 99 % liquid replacement. At a given influent flow rate, the number of liquid circulations between the zones increases with the increase of air flow rate for 90 and 99 % liquid displacement, while it decreases with the increase of influent flow rate. For example, liquid circulates between 521 and 859 times for 90 % liquid displacement at the influent flow rate of 500 L/day when the air flow rate increases from 15 to 45 L/min. These values increase to the range of 1,047–1,698 times for 99 % liquid displacement. With the increase of influent flow rate to 1,450 L/day, liquid circulations decrease to the range of 182–261 times for 90 % liquid displacement, and to the range of 364–590 times for 99 % liquid displacement when the air flow rate increases from 15 to 45 L/min. Regardless of the operating conditions, these values exhibit the extended number of exposure of mixed liquor content, including the contaminating compounds, to three different environmental conditions and a variety of microbial population during wastewater treatment operation.

The hydraulic retention time in each zone was calculated from Eqs. 25 to 27, resulting in the estimation of the time of a single liquid cycle between the three zones. This time ranged from 47 to 86 s for the air flow rates of 15–45 L/min and can be controlled by changing the air flow rate at any given influent liquid flow rate.

## Conclusions

The time-dependent changes in the volume of mixed liquor in the first bioreactor of the BioCAST technology and the dependence of the number of liquid circulations between the aerobic, anoxic and microaerophilic zones on the influent flow rate and air flow rate were theoretically analyzed and experimentally observed. This study showed that at a constant air flow rate, the number of liquid circulations between the three zones for 90 and 99 % liquid displacement decreases with the increasing influent flow rate, in accordance with the increased specific rate of liquid discharge from the reactor. The results showed the high number of mixed liquor circulations between the three

**Table 2** Specific rate of liquid discharge from the bioreactor ($k$) and the percentage of escaped liquid during each cycle at different operating conditions

| $Q_i$ (L/day) | $Q_{air}$ = 15 (L/min) | | $Q_{air}$ = 30 (L/min) | | $Q_{air}$ = 45 (L/min) | | Theoretical* |
|---|---|---|---|---|---|---|---|
| | $k$ (1/h) | % Escaped liquid | $k$ (1/h) | % Escaped liquid | $k$ (1/h) | % Escaped liquid | $k$ (1/h) |
| 500 | 0.19 | 0.47 | 0.19 | 0.32 | 0.21 | 0.25 | 0.21 |
| 720 | 0.23 | 0.67 | 0.21 | 0.46 | 0.31 | 0.37 | 0.30 |
| 1,000 | 0.38 | 0.94 | 0.34 | 0.64 | 0.45 | 0.51 | 0.42 |
| 1,450 | 0.58 | 1.34 | 0.55 | 0.92 | 0.69 | 0.74 | 0.60 |

* Estimated from Eq. 23

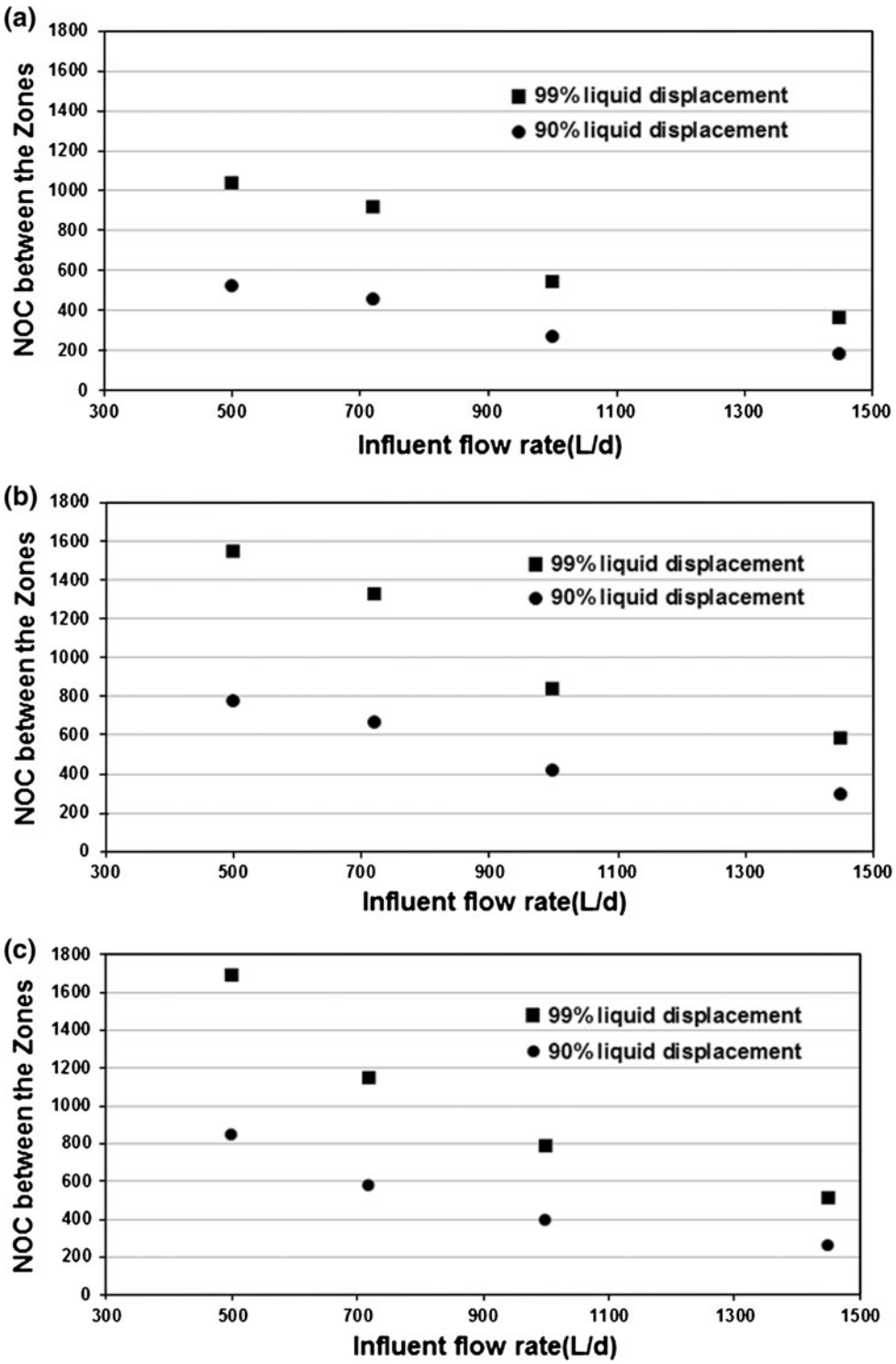


**Fig. 9** Dependence of the number of liquid circulations (NOC) between the three zones of bioreactor on the influent flow rate for 90 and 99 % liquid displacement at various air flow rates ($Q_{air}$). **a** $Q_{air} = 15$ L/min, **b** $Q_{air} = 30$ L/min, **c** $Q_{air} = 45$ L/min

zones having different environmental conditions before the displacement of 90–99 % of its original volume from the bioreactor. Under the examined operating conditions, mixed liquor circulates between 182 and 859 times between the three zones of the bioreactor before 90 % of bioreactor volume is replaced by the influent wastewater. Similarly, for the replacement of 99 % of bioreactor volume, mixed liquor circulations of 364–1,698 times between the three zones are required. Obviously, during continuous operation this trend will repeat, implying that mixed liquor continues to circulate for an extended number of times between the three zones before 99 % of the bioreactor volume is replaced by the influent wastewater. The high number of liquid circulation between the three zones

that reflects the number of wastewater exposure to three different environmental conditions demonstrates increased potential for the removal of contaminants by technologies that use similar design and operation strategies. The results also showed that the percentage of liquid that escapes circulation at each cycle and flows towards the clarifier varies between 0.25 and 1.34 %. This again shows the increased exposure of contaminating compounds in the wastewater to three environmental conditions having a diverse group of microbial biomass during the treatment. The experimental and theoretical investigations of residence time distribution (RTD) and the analysis of the dependence of the dimensionless variance of RTD on air flow rate showed that the RTD in the examined bioreactor is closer to patterns observed in continuous stirred tank reactors (CSTRs) compared to those observed at plug flow regimes. The relationships of the axial dispersion coefficient and Bodenstein number with the superficial gas velocity are similar to those observed in airlift reactors and showed that the mixing regime in the examined bioreactor falls between the patterns observed in perfectly mixed and plug flow reactors.

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