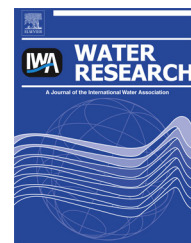


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# Breakage and growth towards a stable aerobic granule size during the treatment of wastewater

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## ABSTRACT

To better understand granule growth and breakage processes in aerobic granular sludge systems, the particle size of aerobic granules was tracked over 50 days of wastewater treatment within four sequencing batch reactors fed with abattoir wastewater. These experiments tested a novel hypothesis stating that granules equilibrate to a certain stable granule size (the critical size) which is determined by the influence of process conditions on the relative rates of granule growth and granule breakage or attrition. For granules that are larger than the critical size, granule breakage and attrition outweighs granule growth, and causes an overall reduction in granule size. For granules at the critical size, the overall growth and size reduction processes are balanced, and granule size is stable. For granules that are smaller than the critical size, granule growth outweighs granule breakage and attrition, and causes an overall increase in granule size. The experimental reactors were seeded with mature granules that were either small, medium, or large sized, these having respective median granule sizes of 425  $\mu\text{m}$ , 900  $\mu\text{m}$  and 1125  $\mu\text{m}$ . An additional reactor was seeded with a mixture of the sized granules to represent the original source of the granular sludge. The experimental results were analysed together with results of a previous granule formation study that used mixed seeding of granules and floccular sludge. The analysis supported the critical size hypothesis and showed that granules in the reactors did equilibrate towards a common critical size of around 600–800  $\mu\text{m}$ . Accordingly, it is expected that aerobic granular reactors at steady-state operation are likely to have granule size distributions around a characteristic critical size. Additionally, the results support that maintaining a quantity of granules above a particular size is important for granule formation during start-up and for process stability of aerobic granule systems. Hence, biomass washout needs to be carefully managed to optimize granule formation during the reactor start-up.

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## 1. Introduction

Aerobic granular sludge systems are an emerging technology where aerobic granules are utilised as opposed to floccular

sludge for activated sludge wastewater treatment (de Bruin et al., 2004; Liu et al., 2005a; de Kreuk et al., 2005). A major advantage of the relatively large and dense granules is that much faster sludge settling rates can be achieved in

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comparison to conventional plants, thus greatly reducing plant footprint and capital cost (de Bruin et al., 2004; Liu and Tay, 2004). Compared to floccular sludge, granular sludge achieves high biomass retention, better handles shock loads and higher loading rates (Liu and Tay, 2004) and shows improved sludge dewaterability (de Kreuk et al., 2005). Paramount to the performance of aerobic granular sludge systems is a stable, dense and reasonably large (0.5–1.5 mm) granule size (Beun and Hendrik, 1999). In reactors, average granule sizes between 0.2 and 5 mm are reported and this size is governed by a combination of growth increasing the granule size, and attrition/breakage processes decreasing its size (Liu and Tay, 2004).

The mechanisms by which granules form and grow have been under examination (Ahn et al., 2009; Barr et al., 2010; Verawaty et al., 2012; Wan et al., 2011; Ni et al., 2010). Aerobic granules are suspended biofilms and models describing their size include processes of attachment and detachment, cell growth, EPS production, cell death and lysis, and predation by higher organisms (Beun et al., 2002; Picioreanu, 1999). Granule growth is suggested to occur by single colony outgrowth or by the aggregation of multiple smaller colonies (Barr et al., 2010; Verawaty et al., 2012) and by layering of cells attaching to a surface (Hulshoff Pol et al., 2006). Seeding of aerobic granular sludge reactors with specific microbial strains (Ivanov et al., 2008) or with crushed aerobic granules and floccular sludge can accelerate granule formation (Verawaty et al., 2012; Pijuan et al., 2011). These findings suggest that seed biomass, including granule fragments, can enhance granule formation.

A feature of biological granules is that they consist of mixed microbial populations imbedded in a three-dimensional glue of extracellular polymeric substances (EPS). The EPS plays a crucial role in determining granule stability (Liu and Tay, 2004) and is described as a hydrogel-like substance (Seviour et al., 2009). Biological activity occurs throughout a granule (not just on the granule surface) and is expected to impact the long-term growth (increase in size), strength and tendency to break (decrease in size). The three-dimensional structure of granules would cause stratification of microorganism types and metabolism, due to a combination of diffusive mass transfer into the granule and biological activity consuming and producing metabolites (Tay et al., 2003). Consequently, substrate limitations within the centres of large granules may alter biological activity and lead to a weakened granule structure (Liu et al., 2005a; Toh et al., 2003). Additionally, larger particles in mixed suspensions will have greater impact energy associated with collisions. As a consequence of these events, the rate at which aerobic granules wear would increase with their size.

Granule structure and size are critical for the performance of an aerobic granular sludge system. However, presently there is limited understanding of the dynamics of particle size for aerobic granule systems. Individual granules in an aerobic reactor are part of a particle population that will comprise variation in size, growth and strength characteristics. To date, there has been limited consideration of the entire aerobic granule population in the study of growth and size reduction processes.

The present study investigates the influence of granule size on breakage/fragmentation propensity and in-turn the influence of granule breakage on the resulting long-term granule

size distribution. Here it is hypothesized that within an aerobic granular sludge system, a certain “critical” granule size will be achieved at steady state under certain operational conditions (wastewater characteristics, aeration, reactor geometry, mixing, and solids concentration). Due to the consequences of particle collision energies and structural weaknesses, granules larger than the critical size are expected to break/attrite until reduced to the critical size or smaller. This rate of breakage/attrition of the large granules is exceeding the rate at which they grow. Conversely, the growth of small granules exceeds the rate at which their size is being reduced by breakage or attrition until they reach the critical size. To test this Critical Size hypothesis, the present study followed the change in the size of granules of various starting sizes over 50 days in separate sequencing batch reactors (SBR). The experimental design considers granule populations in their entirety.

## 2. Materials and methods

### 2.1. Preparation of aerobic granule seed sludge

Aerobic granules used as seed sludge in the experiments were sourced from a 5L lab-scale SBR called the parent reactor or SBRp. The SBRp was operated on an 8-h cycle consisting of stages of static fill (2 min), anaerobic (78 min), aerobic (305 min), anoxic (90 min), settling (2 min) and decanting (3 min), as previously described (Yilmaz et al., 2008). This parent reactor treated abattoir wastewater for biological nutrient removal (BNR). The abattoir wastewater feed contained soluble chemical oxygen demand (COD) at 862–1137 mg/L, volatile fatty acids (VFA) at 650–800 mg/L, ammonia nitrogen ( $\text{NH}_4^+ - \text{N}$ ) at 200–254 mg/L, and orthophosphate phosphorus ( $\text{PO}_4^{3-} - \text{P}$ ) at 31–40 mg/L. The wastewater volumetric exchange ratio (VER) was 50%, resulting in a hydraulic residence time (HRT) of 16 h. Mixed liquor suspended solids (MLSS) and volatile MLSS (MLVSS) were 20.2 g/L and 18.19 g/L respectively. The sludge retention time (SRT) was kept between 15 and 20 days by wasting sludge at the end of the aerated phase.

The entire sludge content of SBRp was fractionated into five different size fractions using standard sieves with apertures of 160  $\mu\text{m}$ , 350  $\mu\text{m}$ , 500  $\mu\text{m}$ , 700  $\mu\text{m}$ , 850  $\mu\text{m}$ , 1000  $\mu\text{m}$  and 1180  $\mu\text{m}$ . From these, three fractions were chosen, large, medium and small, to be used as the seed sludge for the operation of the experimental SBRs (Table 1).

**Table 1 – Size fractionation of mature aerobic granules from SBRp.**

Sieving processes	Seeds name	Particle size distributions ( $\mu\text{m}$ )			Biomass obtained (g VSS)
		$d$ (0.1)	$d$ (0.5)	$d$ (0.9)	
>1800	Large	700	1125	1550	48.19
<700, >500	Medium	606	900	1346	8.5
<500, >160	Small	164	425	763	2.3

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