FULL PAPER

Effect of mixing energy on various freshly developed microcarriers

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Abstract

In the development of biological wastewater cleaning moving bed biofilm reactors (MBBR) are one of the most efficient ways to remove pollutants from water. To have MBBR cleaning technology a biofilm carrying media required. Their key parameter is specific surface area (SSA), which is the available space for biofilm to develop colonies. The colonization rate and speed depend on many factors, such as microorganism type, nutrient forms and availability, temperature and flow velocity around the media. Traditional carriers have a massive, rigid construct, which is durable in almost any mixing condition applied in wastewater cleaning, however significantly smaller microcarriers are a different topic.

With laboratory scale experiments we measured different types of carriers under various mixing conditions to analyze if any makes harm to the structure of the media. We also studied hydrogels freshly after production and hydrogels that have been in use to determine if any activities change their size range. Calculations to determine the flow speeds around the carrier can predict their ability to be colonized by various types of microorganisms. In conclusion all types of media can endure heavy mixing in laboratory scale, however freshly produced media shows a small degradation in diameter, which can be caused by sticking together during production.

Keywords: MBBR, microcarrier, PVA based media, wastewater treatment, shear stress

Introduction & literature

In the wastewater treatment industry one of the most common cleaning methods is to have anaerob/anox/aerob (A2O) biological reactors after physical pretreatment. Mostly flocs used in them, which consist of cleaning bacterias, substrates, organic and other inorganic parts (Rieger et al 2012). In modern treatments moving bed biofilm reactor (MBBR) are more efficient, as they include microorganisms carrying medias, which can be made from HDPE, PE PP (Barwal & Chaudhary, 2014) but also PVA is one of the best materials for media (Al-Amshawee et al, 2020).

The size and the shape of the carriers also affect the properties of the biofilm over their surface. In Bjornberg et al's (2009) study they experienced with media that has inner and outer parts, and their results shows that in the inner circles the biofilm thickness reached up to 300 µm, while on the outer areas the biofilm were absent, but reached close to zero average thickness. However, Torresi et al (2016) suggests that biofilm thicknes should not be above 200 µm to maximize the nitrifying activity. They used a special AnoxKaldness® media that controls the biofilm thickness by its shape and structure.

Either technology is used, the biofilm or microorganisms' colonies greatly affected by the shear forces, the flow velocities in the biological reactor. To determine their affect Xinyan et al (2013) used velocity gradient to calculate the shear stress of microorganisms. Their calculations include the tank volume, solution viscosity, impellers constant at turbulent flow, rotational speed and impeller diameter. Di Iaconi et al (2005) modified the original formula of shear stress calculation to modell the carrier shape resistance (surface friction). They showed high positive correlation between shear forces and biofilm mass, which is due to the shelter behaviour (Tijhuis et al, 1995), that means greater shear stress makes the biofilm thickness, denser. Rittman (1982) showed that weak shear forces lead to weaker organic structure, Liu&Tai (2002) had similar results, however they also showed that shear forces have great effect not only on biomass density, but any metabolic activities.

Material/Methods

At the first experiment we tested the carriers with different mixing conditions, to see if anything happens with the carriers during the biological tests, that's not because of the mixing conditions. The experiment was taken in a 5-litre reactor with 180-220-330 RPM.

Secondly, we examined the shear stress' effect on different microorganisms with 6 reactors with the following setup:

Number	Culture	RPM
1	heterotroph	180
2	heterotroph	220
3	heterotroph	330
4	autotroph	180
5	autotroph	220
6	autotroph	330

1. table – experimental setup

Each reactor setup consists of 500ml PVA based microcarrier, 4l of artificial wastewater (according to the need of microorganism's type) and 500ml of sludge which were stirred to cell fractions and filtered. Each cycle lasted 6hrs with 5h 45min. of aeration and mixing, 10 min of settling and 5 min of water change. One water change was 2 each.

To determine forces affecting the particles in any system the following formula required:

$$F_{she} = \tau * A$$

where:

τ is shear stress,

A is the surface of the particle.

This equation shows the amount of force on each particle in a stirred aqueous system. The surface can be calculated from the diameter of the media. But we also need to determine the shear stress in the system, which is given by this equation:

$$\tau = \eta * \gamma$$

where:

η is the fluids viscosity,

γ is the shear rate. (autmix.com)

The experiments carried out in water, around room temperature, so the viscosity is 1mPas, and for calculating shear rate we used the formula:

$$\gamma = \frac{N}{D}$$

where:

N is the number of rounds per second of the impeller,

D is the diameter of impeller. (Huda, 2021)

$$V = \sqrt{\frac{2 * \tau}{C_d * \rho_f}}$$

MSZ ISO 6060 (COD), MSZ 1484-13:2009 (nitrate-nitrogen, nitrite-nitrogen), MSZ ISO 7150-1:1992 (Ammonium-nitrogen), MSZ 448-18:1977 (phosphate-phosphorous) standards were used during the laboratory measurements. pH, dissolved oxygen, and temperature were measured on-site using a Hach HQ40 portable multimeter.

The diameter of carriers and the growth of biofilm were monitored via light microscopic analysis. The samples were TTC (triphenyl tetrazolium chloride) coloured and taken 30 pictuces for statistical analysis.

Results

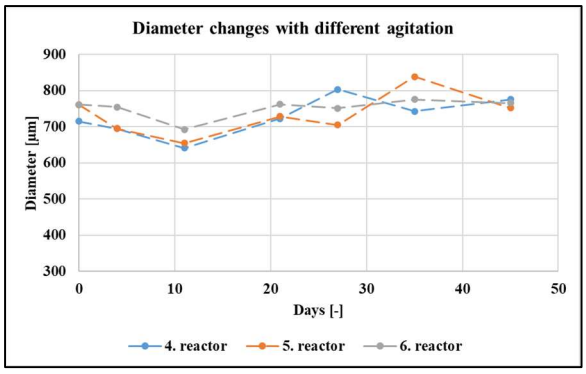
First, we calculated the velocities and shear forces in each reactor using the formulas above. The results can be seen in **Table 2**.

Reactors number	Organism type	Shear force [µN]	Flow velocity [cm/s]	Carrier producing method
1	Heterotroph	0,16	1,98	Warm polymerisation
2		0,11	1,62	
3		0,09	1,46	
4	Autotroph	0,16	1,98	
5		0,11	1,62	
6		0,09	1,46	
1	-	0,16	1,98	Cold polymerisation
2		0,11	1,62	
3		0,09	1,46	
4		0,16	1,98	Warm polymerisation
5		0,11	1,62	
6		0,09	1,46	

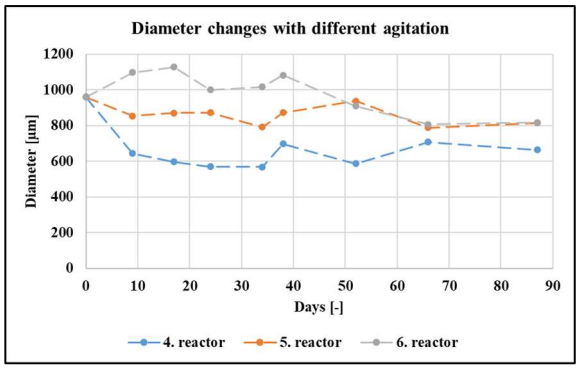
2. table – The shear forces and flow velocities in different experiments

In **Table 2**. we can see that in each different reactor the shear force can be measured in micronewton, which force represents the shear stress to each individual carrier. Flow velocity shows us how fast are the particles in the system which correlates with the shear stress.

To determine if the mentioned shear forces have effects on our freshly developed carriers we made control reactors, that has no major biological activities. It shows us the effect of different mixing conditions on the medias physical properties (size). The results can be seen in **figure 1-2.**



1. Figure – Diameter changes of warm polymerized media

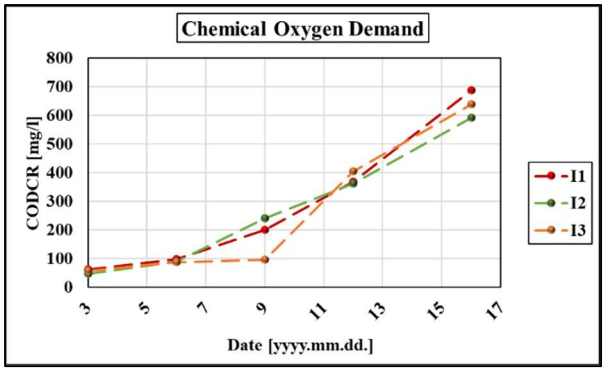


2. Figure – Diameter changes of cold polymerized media

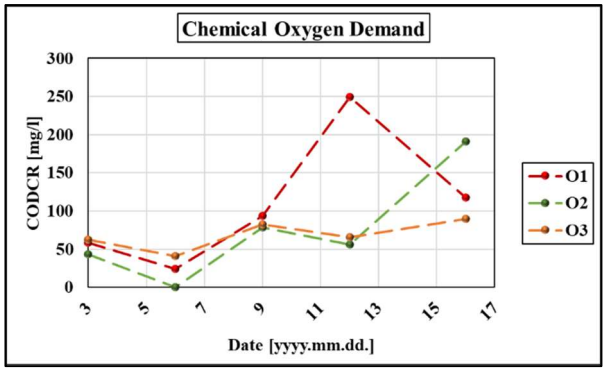
In Figure xx. the experiments with warm polymerized media can be seen. It shows no major differences between the reactors and shows no changes during our 45 days of experiment. The minor differences may be the measurements known margin of errors. However cold polymerized media show differences in the first 40 days between the reactors. The 4. reactor

with highest shear force shows the smallest media size, and 6. reactor has the largest. After day 40 we increased the mixers RPM values up to 400 RPM (initially it was 330; 220 and 180). With increased shear forces 5. and 6. reactors average diameter size decreased to the level of 4. reactor, which did not show any size decrease. With the experiences during the microscopic we can conclude to the fact that the medias were sticked together after the polymerisation process by relatively strong but only physical bonding, that fall apart with the increasing shear forces. But with even 20% increased shear stress the media did not have any decrease in their actual structure.

After testing the medias physical properties, we tested their ability to be colonised by different types of microorganisms. In **figure 3-4.** we can observe the in and effluent of the heterotroph reactors COD values with cold polymerized media.

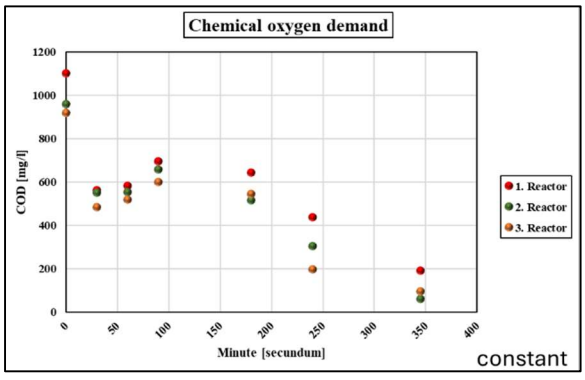


3. Figure – Influent COD values of heterotroph reactors

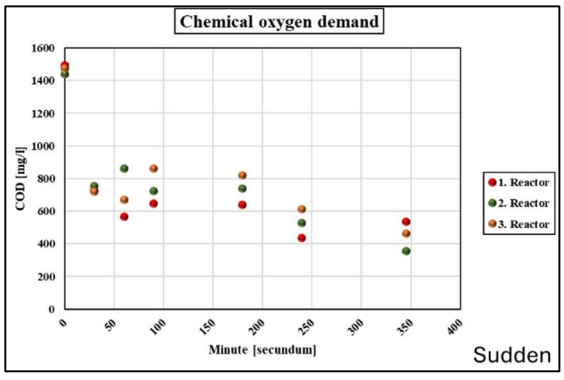


4. Figure – Effluent COD values of heterotroph reactors

We can see that during the experiments we increased the influent values from 50 mg/l up to 650 mg/l values, which is close to the COD concentration in municipal wastewater. We can conclude from Figure xx. that 1. reactor adapted a little slower to the increasing OLR (organic loading rate), but overall, the reactors show small differences in their cleaning efficiency. We also measured one full cycle to determine the metabolism speed of different cultures in the reactors (**5. Figure**). The we doubled the OLR to see how fast they can adapt to the sudden increase of COD values (**6. Figure**).



5. Figure – Full cycle of heterotrophs

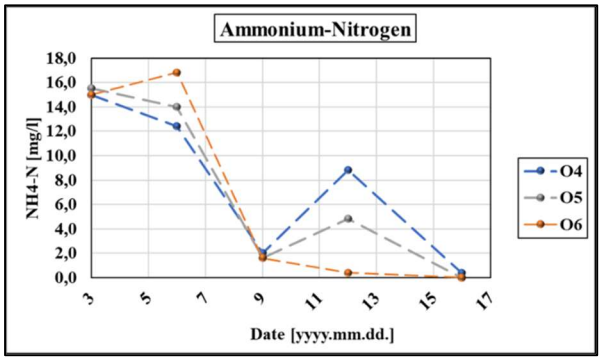


6. Figure – Full cycle of sudden OLR increase of heterotrophs

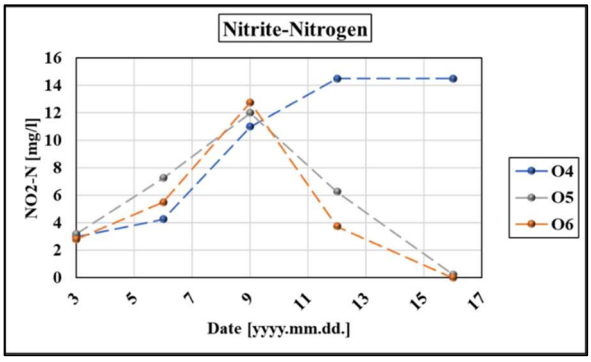
We can see that in the first 100 minutes the COD values decreased to the half of the initial amount, after that a smaller decrease occurred. With increased OLR the heterotrophs significantly increased their metabolism, as in the first 100 minutes they degraded 800 mg/l of OLR. However, after that their metabolism slowed, and at the end of the cycle they could not get lower concentrations than 400 mg/l. The experiments show no significant differences

between the reactors, all have great COD removal rate, and shows rapid adaptation to increased OLR.

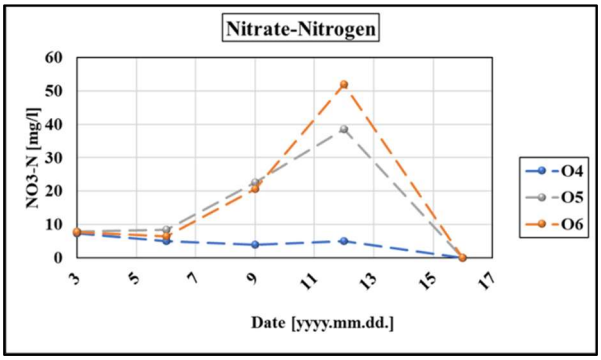
In the 4-6. reactors we experimented with autotroph cultures, both with *Nitrosomonas* and *Nitrobacters*. In **Figure 7-9**. we show the nitrogen forms in the effluent.



7. Figure – Ammonium-nitrogen values of autotrophs



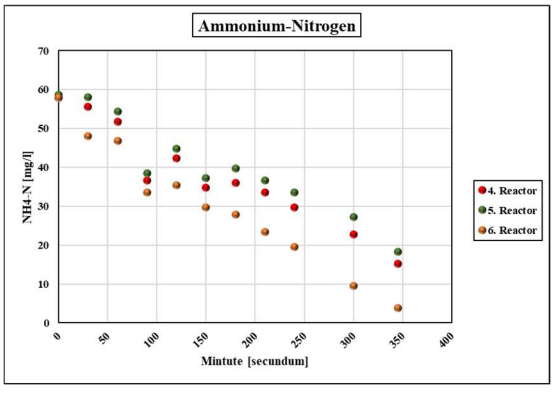
8. Figure – Nitrite-nitrogen values of autotrophs



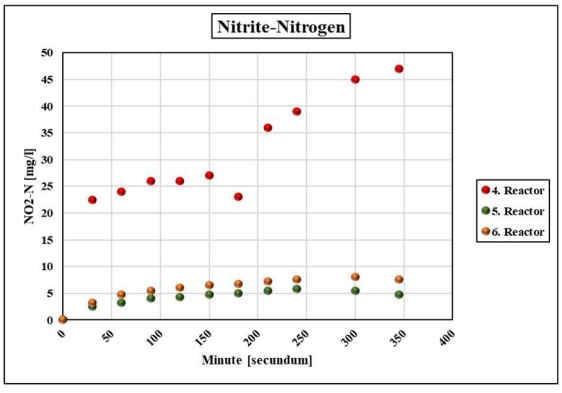
9. Figure – Nitrate values of autotrophs

We can see that ammonium-nitrogen values decreased over time as biofilm began to develop. First few days 6. reactor shows slower progress, but after that it has highest *Nitrosomonas* activity. As ammonium began to decrease nitrite-nitrogen began to increase, however from day 9 *Nitrobacter* cultures developed in 5. and 6. reactor, so nitrate-nitrogen appeared, and nitrite-nitrogen decreased. However, in 4. reactor *Nitrobacter* cultures were handicapped due to the too high shear stress.

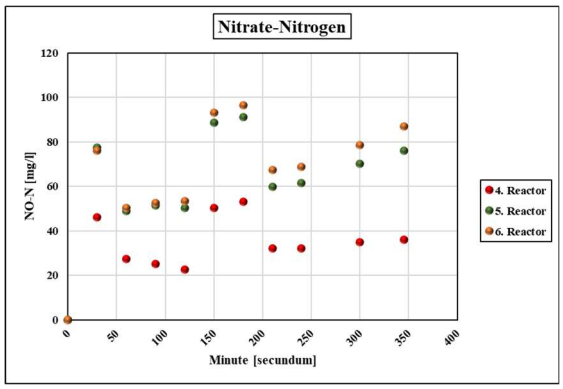
We measured the same two cycles as at heterotrophs, one with constant loading rate and after increasing the influents ammonium concentration (**Figure 10-15.**)



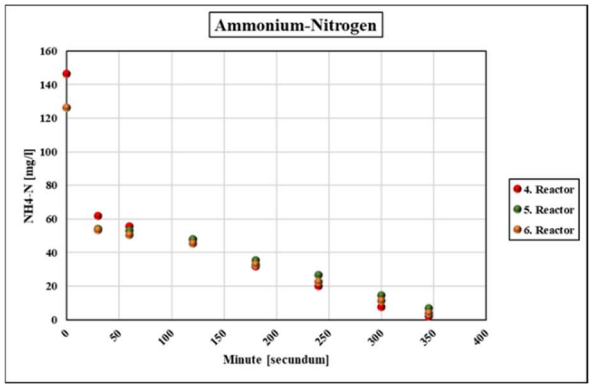
10. Figure – Ammonium-nitrogen at constant cycle



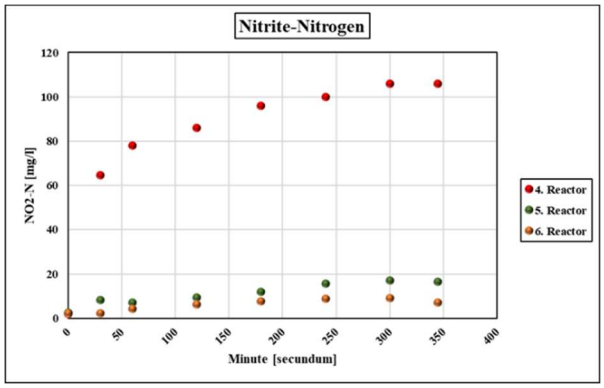
11. Figure – Nitrite-nitrogen at constant cycle



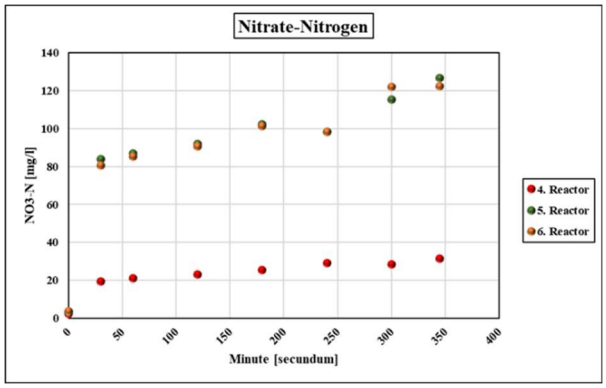
12. Figure – Nitrate-nitrogen at constant cycle



13. Figure – Ammonium-nitrogen at sudden cycle



14. Figure – Nitrite-nitrogen at sudden cycle



15. Figure – Nitrate-nitrogen at constant cycle

It is clear that 4. reactor is handy capped both cycles in terms of turning nitrite into nitrate, however this reactor has slightly better *Nitrosomonas* activity, than 5. reactor. It produces nitrate; however, it stays almost constant with small increase. When sudden ammonium load occurs, 4. reactor show even smaller *Nitrosomonas* activity, their values remain under 40 mg/l, however its *Nitrobacter* cultures are able to degrade the whole ammonium influent concentration. The other two reactor (5. and 6.) shows slight decreased *Nitrobacter* activity as in constant load. However, they also removed all of the initial ammonium concentration, which is impressive considering we doubled the loading rate from a relatively high (60 mg/l to 120 mg/l) starting point.

Conclusion

We made many different experiments to determine the capacity of our freshly developed carriers both in terms of microorganisms carrying capacity and durability and resistance against shear forces. We found that both warm and cold polymerised medias are able to stand various shear forces without any decrease in size. After the producing method cold medias are likely to stick together, for detachment relatively strong shear forces are required.

Both autotroph and heterotroph cultures are able to colonize the warm polymerised medias. Heterotrophs show almost no decrease due to any shear stress, and their COD removal rate are outstanding even with sudden OLR increases. While autotroph organisms can colonise well the medias, they shows decrease in their activity due to shear stress. With increased shear stress *Nitrobacter* activites tend to show a significant amount of decrease even with constant loading rates. *Nitrosomonas* activity however shows almost no decrease and also have an impressive ability to remove ammonium from inlet water even with doubled loading rates.

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