The rate of denitrification using hydrodynamically disintegrated excess sludge as an organic carbon source

J. Walczak and M. Zubrowska-Sudol

ABSTRACT

This study investigates the potential of hydrodynamically disintegrated excess activated sludge when used as a supplementary carbon source for denitrification. Two objectives constituted this study: (i) to analyse the denitrification rate by using excess sludge subjected to hydrodynamic disintegration (HD), performed at different energy densities, as an organic carbon source, and (ii) to analyse the impact of hydrolysis of disintegrated sludge on the denitrification rate. Nitrate reduction tests were conducted to assess the denitrification rate for the following sources of organic carbon: thickened excess sludge disintegrated at three levels of energy density (70, 140 and 210 kJ/L), acetic acid solution and municipal wastewater after mechanical treatment. It was found that the HD of excess sludge conducted at different levels of energy density led to dissolved organic compounds characterised by various properties as donors of H^+ in the denitrification process. The susceptibility of disintegrated sludge to anaerobic hydrolysis decreased along with the increasing energy density. The obtained organic carbon contributed to a lower increase in the denitrification rate in comparison to that when disintegrated sludge not subjected to hydrolysis was applied.

Key words | carbon source, denitrification rate, disintegrated sludge, hydrodynamic disintegration, hydrolysis, nitrogen utilisation rate (NUR) test

J. Walczak (corresponding author)
M. Zubrowska-Sudol
Faculty of Building Services, Hydro and Environmental Engineering, Department of Water Supply and Wastewater Management,
Warsaw University of Technology,
ul. Nowowiejska 20, 00-653 Warsaw,
Poland
E-mail: *justyna.walczak@pw.edu.pl*

INTRODUCTION

Biological removal of nitrogen from wastewater involves the process of dissimilatory nitrate reduction (commonly called denitrification) (Tchobanoglous *et al.* 2003). This process is carried out by heterotrophic organisms. One of the factors influencing their effectiveness is the amount of easily digestible organic carbon entering the bioreactor with wastewater. A deficit of organic compounds inhibits nitrogen removal. It is a common problem in wastewater treatment plants. A typical additional carbon source, such as methanol, ethanol, acetic acid, or glucose, is used to enable denitrification to achieve the required effluent quality (Zhang *et al.* 2016b). Another solution is to use waste products from the food industry (Fernández-Nava *et al.* 2010; Zhang *et al.* 2016a) or by-products of biodiesel rich in glycerine (Latker *et al.* 2011). These are called alternative sources of organic carbon. Several studies have been conducted in recent years testing the application of disintegrated waste activated (excess) sludge (WAS) or return activated sludge to enhance nitrogen removal from wastewater (Biradar *et al.* 2010; Yan *et al.* 2013). The disintegration process is a destruction of sludge structure by added energy, leading to changes in the physico-chemical properties of the sludge. It causes a release of organic substances, which were previously extra- and intracellular bound (Kampas *et al.* 2007). In order to define energy inputs in the disintegration process, the following parameters are used: energy density (ε_L; expressed as kilojoules per litre of disintegrated sludge) and specific energy (ε_S; expressed as kilojoules per kilogram of total solids of disintegrated sludge). The results of currently available studies show that organic compounds obtained in the disintegration process are a proper substrate for denitrifying bacteria. Quite detailed research on the rate of denitrification with the application of disintegrated waste sludge as a carbon source was conducted by Kampas *et al.* (2009) and Soares *et al.* (2010). Both research teams showed that disintegrated sludges are a better source of organic carbon for denitrification bacteria than acetate. This can be related to the type of particular organic compounds occurring in the analysed media. During the disintegration of sludge from activated sludge flocs, various

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'natural' organic compounds are released, mainly belonging to easily biologically available compounds. According to the study by Kampas *et al.* (2007), the composition of dissolved organic compounds present in disintegrated sludge included proteins (30%), carbohydrates (13%) and volatile fatty acids (12%), and 45% of organic compounds was not identified. It is therefore a mixture of different compounds, which can stimulate the intensity of the denitrification process.

One process for obtaining organic compounds for facilitating the denitrification process, extensively described in the literature m is anaerobic hydrolysis of sludge (Ucisik & Henze 2008; Guo *et al.* 2017). The idea of the process involves the transformation of long-chain organic compounds into biologically available compounds such as short-chain fatty acids or alcohols. Primary sludge is more susceptible to the hydrolysis process in comparison to excess sludge. For example, research by Ucisik & Henze (2008) shows that the efficiency of hydrolysis, referred to as the amount of obtained soluble chemical oxygen demand (SCOD), was 19.1% for primary sludge, 6.5% for activated sludge, and 21.37% for mixed sludge (for time of retaining sludge of 5 days and temperature of 37 °C). Disintegration contributed to a change of structure of activated sludge, causing among others an increase in the susceptibility of disintegrated sludge to the process of anaerobic hydrolysis (Ebenezer *et al.* 2015; Liu *el al.* 2017a). It was therefore assumed that subjecting disintegrated sludge to the process of anaerobic hydrolysis will permit an additional source of organic carbon to be obtained, which can be used for the intensification of the denitrification process. The hydrolysis process can be conducted in a container equipped with a mechanical stirrer where disintegrated sludge will be kept for a specified time. For such an operation to be profitable, disintegration combined with hydrolysis will have to lead to the release of a supplemental organic carbon load. On the other hand, the released organic compounds should be more easily digestible for denitrifying bacteria. Information on the rate of denitrification obtained during application of disintegrated sludge is still scarce in comparison with that referring to other sources of organic compounds. Moreover, no research has been conducted so far on the rate of denitrification with the application of disintegrated waste sludge subjected to anaerobic hydrolysis as a carbon source in the denitrification process. It should be stressed that knowledge of this parameter is important for designing the process. The value of this parameter determines the required volume for the anoxic chamber, or, in the case of sequencing batch reactors, the duration of the anoxic phase in the reactor.

There were two objectives in this study:

(i) To analyse the rate of denitrification by using excess sludge subjected to hydrodynamic disintegration (HD) conducted at different energy densities (ε_L) as an organic carbon source, and to compare the obtained rates of denitrification with those when acetic acid solution and municipal wastewater after mechanical treatment as a carbon source were applied.
(ii) To analyse the impact of 24 hour hydrolysis of disintegrated sludge on the denitrification rate.

METHODS

Characteristics of sludge

Activated sludge used as an inoculum for nitrogen utilisation rate (NUR) tests and excess activated sludge used for HD were collected from the secondary sedimentation tank and belt press of a wastewater treatment plant with biological nitrogen removal (LARNE II system, MUCT (modified University of Cape Town) configuration), respectively. The plant has a capacity of 53,040 population equivalent. The total suspended solids (TSS) and volatile suspended solids (VSS) concentration of thickened excess activated sludge used for HD amounted to 38.6 ± 1.6 g/L and 29.2 ± 1.2 g/L, respectively. The TSS and VSS concentrations of activated sludge used as inoculums were equal to 11.9 ± 2.2 g/L and 9.2 ± 1.8 g/L, respectively.

Characteristics of the disintegration process

A laboratory HD apparatus was used in the study. It consists of a multi-use rotor driven by a motor with a power of 2.2 kW and revolutions per minute of 2,800 (Polish patent no. 214335), installed in a 10 L tank, as described in detail in our previous study (Zubrowska-Sudol & Walczak 2014). The disintegration process was conducted at the following levels of energy density: 70, 140, and 210 kJ/L.

Degree of sludge disintegration

The direct assessment of the effects of sewage sludge disintegration employed sludge disintegration degree (DD). The index was calculated according to the following formula (Nickel & Neis 2007):

$$DD = (SCOD_{\varepsilon L} - SCOD_0)/(TCOD - SCOD_0)[\%]$$

where:

$SCOD_{\varepsilon L}$ – soluble chemical oxygen demand for disintegrated sludge with a given level of energy density, mg SCOD/L;

$SCOD_0$ – soluble chemical oxygen demand for non-disintegrated sludge, mg SCOD/L;

TCOD – total chemical oxygen demand for non-disintegrated sludge, mg COD/L.

Determination of the denitrification rate with different carbon sources

The assessment of whether the organic compounds released in the process of disintegration of thickened excess sludge at different ε_L are a proper substrate for denitrification bacteria employed an NUR test – the tests were labelled NUR-A. The assessment of the rate of the denitrification process with the application of disintegrated sludge subjected to anaerobic hydrolysis as a carbon source was also performed based on results of the NUR tests. These types of tests were labelled NUR-B. In NUR-A tests disintegrated sludge was applied immediately after performing the disintegration process at three levels of ε_L (labelled, respectively, D-70 kJ/L, D-140 kJ/L and D-210 kJ/L). Moreover, on the same day NUR tests were performed with acetic acid solution (AcOH), which is considered an easily digestible source of organic carbon for denitrification bacteria, and with municipal wastewater after mechanical treatment (W) obtained from the same wastewater treatment plant as the inoculum. An NUR test was simultaneously performed for a sample with no addition of a carbon source (the so-called blank sample) (see Table 1).

The NUR test was performed in a beaker with a working volume of 1 L at an inoculum concentration of approximately 3 g MLSS/L (MLSS: mixed liquor suspended solids). Gaseous nitrogen was applied over the surface of the liquid for the duration of the test. This permitted the elimination of the possibility of atmospheric oxygen diffusion. Before starting the test, a KNO_3 solution and examined sources of organic carbon were added to the batch reactor. The initial concentration of nitrate nitrogen and SCOD amounted to 30 mg NO_3^--N/L and 120 mg SCOD/L, respectively. The samples were collected every 30 min from the continuously stirred reactor, and immediately filtered with 0.45 µm syringe filters. Then the nitrites (NO_2^--N) and nitrates (NO_3^--N) were measured. Observation of changes in the concentration of NO_x-N (NO_3^--N + NO_2^--N) was conducted for 3 hours. The denitrification rate (V_{DN}; mg NO_x-N/gVSS·h) was calculated by the decreasing slope of the NO_x-N concentration with time and divided by grams VSS.

NUR-B tests were performed on the following day with the application of disintegrated sludge subjected to an additional 24 hours of anaerobic hydrolysis as a carbon source. The hydrolysis was performed in test reactors with a working volume of 1 L, ensuring constant stirring of the samples. The process was performed at a temperature of 20 °C (the experiment was conducted in an air-conditioned room). The NUR-B tests were repeated according to the method described for NUR-A except that the test reactor was fed with the same volume of the examined carbon source as in tests performed before hydrolysis. SCOD was additionally determined in samples after 24 hours of hydrolysis.

Four repetitions of the NUR test were performed for each of the analysed sources of organic carbon, in 1-week intervals, and subsequent tests were labelled as NUR-A.R1

Table 1 | Sources of organic carbon used for the NUR tests

	Blank	Acetic acid solution	Municipal wastewater after mechanical treatment	Thickened excess sludge disintegrated at different energy densities (ε_L)			Remark
				70 kJ/L	140 kJ/L	210 kJ/L	
NUR-A	● Blank	● AcOH	● W	● D-70 kJ/L	● D-140 kJ/L	● D-210 kJ/L	Immediately after disintegration
NUR-B	–	–	–	● D-70 kJ/L+H	● D-140 kJ/L+H	● D-210 kJ/L+H	After 24 h hydrolysis of disintegrated sludge
SCOD of organic carbon sources (mg/L)	–	10,000	678 ± 92	described in Tables 2 and 3			

where:

D-70 kJ/L (140 kJ/L, 210 kJ/L) – thickened excess sludge disintegrated at energy density equal to 70 kJ/L (140 kJ/L, 210 kJ/L);

D-70 kJ/L + H (140 kJ/L + H, 210 kJ/L + H) – thickened excess sludge disintegrated at energy density equal to 70 kJ/L (140 kJ/L, 210 kJ/L) and subjected to 24 hour hydrolysis.

and NUR-B.R1, NUR-A.R2 and NUR-B.R2, etc. An important methodological assumption was that tests for all of the analysed sources of organic carbon in a given repetition were performed for the same inoculum. Therefore the amount of denitrifying bacteria in the test reactors, constituting one of the factors determining the rate of the denitrification process, was the same.

Analytical methods

All chemical analyses, i.e. NO_3^--N, NO_2^--N, SCOD, VFA (volatile fatty acids), TSS and VSS, were measured according to *Standard Methods* (APHA/AWWA/WEF 1998). The pH was measured by means of a pH meter.

RESULTS AND DISCUSSION

Characteristics of sludge after mechanical disintegration of excess activated sludge

The obtained results show an increase in the dissolved organic compound concentration and VFA along with an increase in the amount of energy used in the HD process (expressed as ε_L). HD of thickened excess sludge conducted at 70 kJ/L resulted in obtaining a 5.5 ± 0.9-fold increase in the SCOD concentration and a 7.8 ± 2.9-fold increase in VFA in comparison to the condition before disintegration (see Table 2). For $\varepsilon_L = 210$ kJ/L, even a 26.8-fold and 66.2-fold increase in SCOD and VFA, respectively, was possible. The increase in SCOD resulting from HD shows a level similar to that in the case of other mechanical disintegration methods (Biradar *et al.* 2010; Soares *et al.* 2010; Zhang *et al.* 2007). DD reached values from $4.5 \pm 0.7\%$ for $\varepsilon_L = 70$ kJ/L to $23.4 \pm 3.0\%$ for $\varepsilon_L = 210$ kJ/L.

The increase in SCOD results from the fracturing of the floc structure, breakage of extracellular polymeric substances (EPS), lysis of cells, and metabolic disactivation of microorganisms (Wang *et al.* 2006; Kampas *et al.* 2007; Zhang *et al.* 2007). It is still unknown, however, whether the breakage of flocs and cell lysis occur at the same time or perhaps – as some authors suggest (Wang *et al.* 2006) – gradually, first involving breaking down of the floc structure, followed by cell lysis and release of intracellular substances. This issue has still not been explained. Kampas *et al.* (2007) noticed that organic compounds obtained in the initial stage of mechanical disintegration (specific energy up to 2,500 kJ/kg total solids) mainly originated from the fragmentation of activated sludge flocs and breakage of extracellular substances. Above this energy, it was not clear to the authors whether an increase in released organic matter was due to the further disintegration of flocs or a result of cell lysis. Cho *et al.* (2012), examining the influence of ultrasound on the disintegration process, suggested measurement of the concentration of EPS and RNA as criteria of floc breakage and lysis of cells, respectively. The authors observed that in the initial stage of disintegration, the disintegration of flocs and lysis of microorganism cells occurred simultaneously, but after 10 minutes of sonication the main hydrolytic process was cell lysis. Similar observations were made by Zhang *et al.* (2007). The researchers found that floc breakage and cell lysis occurred in a continuous way, but at the second stage (10–30 min of sonication) mostly cell lysis was observed, resulting in the disactivation of the sludge. Wang *et al.* (2006), also studying ultrasonic disintegration, observed changes in the concentration of polysaccharides, proteins, and DNA, with dominant release of protein. The authors linked an increase in the concentration of protein, critical for floc breakage, with the destruction of EPS. As a result of the loosening of sludge flocs, further shearing forces (after 5 minutes) led to the destruction of the cell walls and release to the aqueous phase of the intracellular substances, reflected in a rapid increase in the concentration of DNA. With an increase in time of sonication, an increase in protein and DNA concentration became slow, suggesting that the sample was becoming less vulnerable to the forces causing the disintegration. Liu *et al.* (2017b), conducting research on the course of disintegration by means of the hydrocyclone method, determined that the distribution of the size of particles of disintegrated sludge as a function of pressure

Table 2 | Characteristics of thickened excess activated sludge before and after disintegration depending on energy density

	ε_L (kJ/L)	pH (–)	SCOD (mg/L)	VFA (mg/L)	DD (–)
Before disintegration	–	6.4 ± 0.4	152 ± 14.3	7.8 ± 2.9	–
After hydrodynamic disintegration	70	6.6 ± 0.3	831 ± 118	38.8 ± 16.6	4.5 ± 0.7
	140	6.6 ± 0.3	$1{,}636 \pm 158$	125.3 ± 42.6	9.8 ± 1.2
	210	6.5 ± 0.3	$3{,}694 \pm 471$	345.0 ± 46.1	23.4 ± 3.0

changes in a range from 0.07 to 0.16 MPa hardly changed. The result was at variance with other methods of mechanical disintegration (Lehne *et al.* 2000; Chu *et al.* 2001), where particle size analysis showed size reduction but was not suitable for characterising the release of organic material and cell disruption.

Rate of denitrification using disintegrated sludge as a source of organic carbon (NUR-A)

In accordance with the methodology described above, NUR-As were planned with initial conditions identical in each test (inoculum concentration, NO_3^--N and SCOD). The only factor affecting the denitrification rate was the type of source of organic carbon. The rate of denitrification determined for disintegrated sludge as a carbon source was dependent on the energy density at which the HD process was carried out (as illustrated in Figure 1). In each series, the maximum value of this parameter occurred for D-210 kJ/L (taking into consideration only tests carried out for disintegrated sludge). This observation indicates that the organic compounds obtained during hydrodynamic sludge disintegration performed at 210 kJ/L were characterised by the best properties as an organic substrate for denitrifying bacteria. One of the factors determining different V_{DN} for D-70 kJ/L, D-140 kJ/L and D-210 kJ/L could be the amount of volatile fatty acids (ΔVFA) introduced into the reactors with the disintegrated sludge. According to the obtained results (see Figure 2), however, this was not a major factor determining the properties of disintegrated sludge as H^+ donors for denitrifying bacteria: (1) in NUR-A.R2, V_{DN} for sample D-140 kJ/L was comparable with V_{DN} calculated for D-70 kJ/L, despite the 2.8-times higher ΔVFA, (2) in NUR-A.R3, the maximum value of ΔVFA occurred for a sample characterised by the lowest V_{DN} (D-140 kJ/L). This suggests that dissolved organic compounds other than VFA contained in D-210 kJ/L stimulated a higher level of V_{DN} than V_{DN} selected for D-140 kJ/L and D-70 kJ/L. Similar conclusions were drawn by Kampas *et al.* (2009) and Soares *et al.* (2010).

The comparison of V_{DN} obtained for disintegrated sludge with V_{DN} obtained for municipal wastewater shows that V_{DN} determined for D-210 kJ/L was 1.3–1.6 times higher, and V_{DN} determined for D-70 kJ/L and D-140 kJ/L

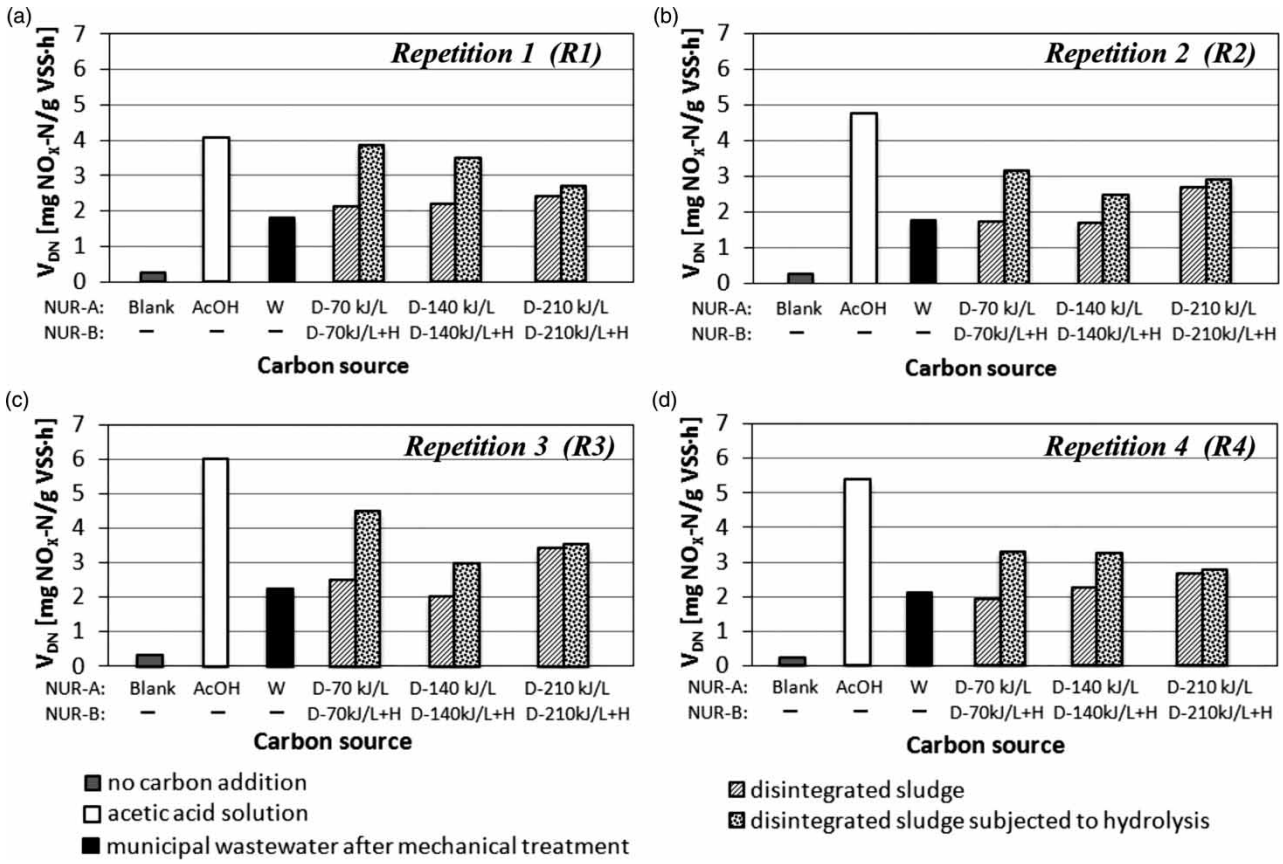

Figure 1 | Denitrification rates (V_{DN}) determined for different types of organic substrates.

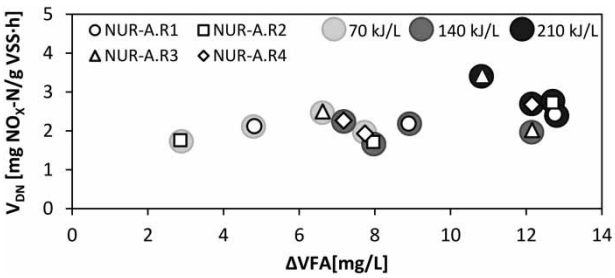

Figure 2 | Rate of denitrification (V_{DN}) as a function of the amounts of volatile fatty acids (ΔVFA) introduced with disintegrated sludge to batch reactors in NUR-A tests.

was at a comparable level. This is particularly important because the biomass was adapted to the carbon source, namely wastewater obtained from the same wastewater treatment plant as the activated sludge serving as the inoculum of the test reactors, in contrast to the application of disintegrated sludge as a source of organic compounds. It is safe to presume that, if activated sludge were subjected to adaptation to new substrates in the form of organic compounds obtained from disintegrated sludge, the V_{DN} determined for such carbon sources would be even higher. This was evidenced by Müller (2000) in experiments conducted with the application of mechanically disintegrated sludge. The analysis of the obtained results also shows that the best substrate for denitrification bacteria, allowing the highest rates of dissimilating nitrate reduction to be obtained, proved to be acetic acid; Kampas *et al.* (2009) and Soares *et al.* (2010) present different findings. Both research teams showed that disintegrated sludge is a better source of organic carbon for denitrification bacteria than acetates.

Possibility of releasing an additional load of dissolved organic compounds in the process of anaerobic hydrolysis of disintegrated sludge

The 24 hour hydrolysis of disintegrated sludge at energy densities of 70, 140 and 210 kJ/L always resulted in an increase in SCOD. It was, however, always dependent on the ε_L of the disintegration process (see Table 3). The highest increase in SCOD, amounting to 145.4 ± 24.8%, was observed for disintegrated sludge at the lowest ε_L (70 kJ/L), and the lowest increase was observed at the highest ε_L (210 kJ/L), for which SCOD did not increase by more than 32.9% in any of the tests. The hydrolysis of non-disintegrated raw sludge conducted in the experiment resulted in releasing 585 ± 170 mg/L of organic compounds (ΔSCOD). The susceptibility of sludge to anaerobic hydrolysis increased after disintegration. Hydrolysis of disintegrated sludge at ε_L = 70 kJ/L resulted in even a 2.2 ± 0.6-fold increase in ΔSCOD in comparison to ΔSCOD of non-disintegrated raw sludge subjected to hydrolysis. After hydrolysis of disintegrated sludge at ε_L = 210 kJ/L, it was possible to release only 1.2 ± 0.5 times more ΔSCOD than for sludge not subjected to the disintegration process. The obtained results suggest that an increase in ε_L caused a gradual reduction of the susceptibility of disintegrated sludge to anaerobic hydrolysis. This probably resulted from the fact that the amount of organic compounds available in sludge, which could be released to the sludge liquid as a result of application of the hydrolysis process, decreased after disintegration.

Rate of denitrification using disintegrated sludge subjected to additional hydrolysis as a source of organic carbon (NUR-B)

The present study is an attempt to answer the question whether the anaerobic hydrolysis of disintegrated sludge can contribute to obtaining an additional load of dissolved organic compounds. This in turn will allow higher rates of denitrification to be obtained. Pursuant to the adopted methodology, each NUR-B test was conducted with the same volume of disintegrated sludge as that in tests conducted before hydrolysis. This meant that the initial value of SCOD in the NUR-B tests was

Table 3 | SCOD depending on energy density

	Sludge not subjected to the disintegration process	Sludge subjected to the disintegration process at different energy densities		
		70 (kJ/L)	140 (kJ/L)	210 (kJ/L)
SCOD before hydrolysis (mg/L)	152 ± 14.3	831 ± 118	1,636 ± 159	3,694 ± 472
SCOD after hydrolysis (mg/L)	737 ± 170	2,035 ± 313	2,749 ± 589	4,458 ± 864
ΔSCOD (mg/L)	585 ± 170	1,204 ± 227	1,113 ± 430	764 ± 451

different and resulted from the efficiency of release of the additional load of organic compounds during the hydrolysis process.

The research showed that the application of disintegrated sludge subjected to 24 hour hydrolysis as a source of organic carbon resulted in an increase in the rate of nitrate reduction in all of the experiments (see Figure 1). The maximum V_{DN} was obtained for disintegrated sludge at $\varepsilon_L = 70$ kJ/L subjected to 24 hour hydrolysis (3.7 ± 0.6 mg NO_x^--N/g VSS·h). It was 1.6–2.2 times higher than that obtained for municipal wastewater after mechanical treatment. This suggests that the denitrification process applied not only to dissolved organic compounds obtained in the disintegration period as H^+ donors, but also to compounds transformed in the process of hydrolysis from the colloidal and/or suspension form to a form that is digestible for microorganisms.

The figures also show (see Figure 3) that the hydrolysis of sludge previously subjected to HD with increasing energy density contributes to a gradually decreasing growth of V_{DN}. For D-70 kJ/L + H, D-140 kJ/L + H and D-210 kJ/L + H, ΔV_{DN} was equal to 71.9–82.6%, 44.3–60.4% and only 3.0–11.4%, respectively. Sludge subjected to disintegration at an increasing ε_L was characterised by reduced susceptibility to hydrolysis, as evidenced by a smaller increase in SCOD, as described in the previous section. This, in turn, resulted in a decrease in ΔV_{DN}.

Comparison of results with different carbon sources

Table 4 compares results of the authors' own research with those obtained by other research teams. The denitrification rates obtained using the HD of excess sludge as a carbon source were found to be at a similar level as those obtained using glucose (Zhang *et al.* 2016b), WAS thermal hydrolysis liquid, and WAS acidogenic liquid as a carbon source (Guo *et al.* 2017). They were lower than those obtained using methanol as a carbon source (Zhang *et al.* 2016a) and those obtained by other researchers using different carbon sources: organic waste (Fernández-Nava *et al.* 2010), sodium acetate, and fermentation liquid from food waste (Zhang *et al.* 2016b). Interestingly, V_{DN} with the application of disintegrated sludge at $\varepsilon_L =$ 70 kJ/L additionally subjected to hydrolysis as a carbon source was higher than that obtained using glucose, WAS thermal hydrolysis liquid, and WAS acidogenic liquid as a carbon source (Zhang *et al.* 2016b; Guo *et al.* 2017). A great advantage of the use of disintegrated sludge in comparison to conventional sources of organic carbon, however, is the recycling of waste produced in the wastewater treatment plant. Owing to the disintegration process leading to the release of organic compounds from activated sludge flocs, integrated sludge becomes a potential substrate for microorganisms responsible for the removal of biogenic compounds from wastewater. It can therefore be assumed that disintegration permits the transformation of cumbersome waste into a 'product', namely organic compounds. This is in accordance with current trends concerning the development of the water, sewage, and energy sectors, showing a need for the search for and implementation of technologies based on the rules of sustainable use of resources and on rules of a circular economy. Costs related to obtaining such a carbon source include exclusively the costs of electricity needed for the operation of the disintegrator. Costs of obtaining organic compounds in the process of HD are presented in the article by Walczak *et al.* (2017). In the case of disintegration combined with hydrolysis, costs of electricity used for mixing sludge should be added.

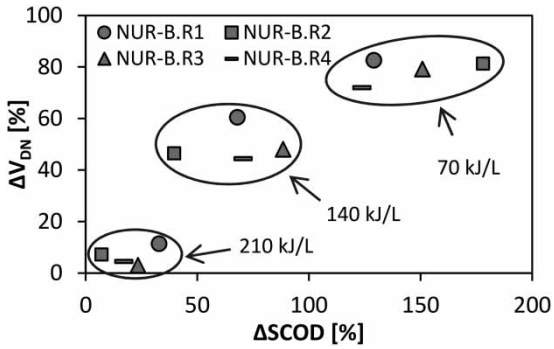

Figure 3 | Increase in the denitrification rate (ΔV_{DN}) depending on an increase in SCOD (ΔSCOD) for disintegrated sludge additionally subjected to hydrolysis.

CONCLUSIONS

The HD of excess sludge conducted at different levels of energy density (70, 140, 210 kJ/L) leads to obtaining dissolved organic compounds characterised by various properties as donors of H^+ in the denitrification process. The maximum V_{DN} with excess sludge subjected to HD at 210 kJ/L as a carbon source was equal to 2.8 ± 0.4 mg NO_x-N/g VSS·h. It was 1.3–1.6 times higher than that obtained for municipal wastewater after mechanical treatment and 2.0–1.7 times lower than that with acetic acid solution as a carbon source. The V_{DN}s for D-70 kJ/L and

Table 4 | Denitrification rates over a range of internal and external carbon sources

Carbon source	Denitrification rate[a]	Initial NO_3^--N[b]	C/N ratio[c]	Reference
Fermented primary sludge	1.4	–	10.0	Soares *et al.* (2010)
Disintegrated surplus activated sludge	1.9	–	9.0	
Sodium acetate	0.9	–	9.0	
Disintegrated surplus activated sludge	14.9	133	–	Kampas *et al.* (2009)
WAS thermal hydrolysis liquid	2.77	30	approx. 6.6	Guo *et al.* (2017)
WAS acidogenic liquid	3.23	30	approx. 6.0	
Acidogenic liquid from food waste	25.0–26.1	33–35	6.0	Zhang *et al.* (2016a)
Acetate	27.5	32.5	6.0	
Methanol	10.9	32.5	7.0	
Fermentation liquid from food waste	approx. 13.0	approx. 50	8.0	Zhang *et al.* (2016b)
Glucose	approx. 3.5	approx. 50	8.0	
Sodium acetate	approx. 16.0	approx. 50	8.0	
Organic waste from a sweets factory	30.4–41.6	560	6.5	Fernández-Nava *et al.* (2010)
Organic waste from a soft drinks factory	31.8–46.8	560	5.5	
Organic waste from a dairy factory	36.2–44.1	530	4.6	
Disintegrated sludge 70 kJ/L	1.8–2.3	30	4.0	This study
Disintegrated sludge 70 kJ/L after hydrolysis	3.2–4.5	30	9.9 ± 1.0	
Disintegrated sludge 140 kJ/L	1.7–2.3	30	4.0	
Disintegrated sludge 140 kJ/L after hydrolysis	2.5–3.5	30	6.7 ± 1.8	
Disintegrated sludge 210 kJ/L	2.4–3.4	30	4.0	
Disintegrated sludge 210 kJ/L after hydrolysis	2.7–3.5	30	4.8 ± 0.4	
Wastewater after mechanical treatment	1.8–2.5	30	4.0	
Acetic acid solution	4.1–6.0	30	4.0	

[a]Denitrification rate: mg NO_x-N/g VSS·h.
[b]Initial NO_3^--N: mg NO_3^--N/L.
[c]SCOD to NO_x-N ratio.

D-140 kJ/L were comparable to V_{DN} for wastewater. The susceptibility of disintegrated sludge to anaerobic hydrolysis decreased along with an increasing energy density. The obtained organic carbon contributed to a lower increase in the denitrification rate in comparison to the use of disintegrated sludge not subjected to the process of hydrolysis.

Further investigation will be focused on identification of organic compounds released from activated sludge flocs, and explanation of their role in the denitrification process.

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