

### Microbiology

# Wastewater treatment plants – a look into a black box

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The technological processes used for biological wastewater treatment in the paper industry have been developed to maturity, and plant operators know the operational concepts their systems are based on. What they do not know well enough in many cases, however, is the importance and role of tiny protagonists – a knowledge that would enable them to identify the causes of disturbances quickly and directly.

Operating a wastewater treatment plant and understanding its internal processes are two things that do not always go hand in hand. If there are problems or disturbances, however, it would certainly help to look into the "black box" of wastewater treatment plants to find efficient solutions.

Microbial, chemical or morphological biomass changes occur practically every day and over extended periods in wastewater treatment plants. Improved and more powerful analyses like gene probe technology, microscopic studies, degradation and activity tests etc. can help plant operators detect these changes and assess their importance for the process. Finally, a brief look at methods that can be used to detect disturbances in the microbiology of wastewater treatment plants is given.

The most recent survey on the water and wastewater situation of the German pulp and paper industry conducted on behalf of VDP in 2010<sup>1</sup> also asked about the problems currently encountered in wastewater treatment plants. Many plant operators find it difficult to ensure the compliance with COD, BOD<sub>5</sub> and temperature limits. Disturbances caused by solid losses, often due to sludge degeneration, occur in both anaerobic and aerobic treatment stages. Bulking and floating sludge, fine sludge/pin flocs formed by the decomposition of greater

Inflating sludge	Floating sludge	Highly dispersed sludge	
<ul> <li>&gt; Waste water properties</li> <li>&gt; Micro biocides</li> <li>&gt; Operational mode and procedure of the aerobic step</li> </ul>	Finely dispersed gas vesicles Surface active substances hydrophobic waste water ingredients (Bacteria with hydrophobic surface involved) Stabilisation of the gas vesicles	<ul> <li>Waste water properties</li> <li>Micro biocides</li> <li>Operational mode and procedure of the aerobic step</li> </ul>	

Tab. 1: Forms of aerobic sludge degeneration and their sources

flocs and foam occur especially in aerobic wastewater treatment plants. Anaerobic sludge degeneration and solid losses are mainly caused by pellet decomposition, fine sludge and floc formation and pellet flotation.

Additives used for papermaking can have biocidal, biostatic and physical effects on the biomass of biological treatment stages. They can also pass the biological stages without undergoing (full) degradation. The latter is usually assessed as biodegradability, a property that can be defined by the ratio of BOD<sub>5</sub>/COD. This provides information about which share of the additives used can be degraded in the biological wastewater treatment plant. Pulp and paper producers use additives of very low biodegradability (BOD<sub>5</sub>/COD <0.2) in some cases. These pass the biological stages nearly inertly, thus contributing to recalcitrant COD in the final effluent. Papermaking additives with low biodegradability are, for example, wet strength agents, optical brighteners, dyestuffs and retention aids.

In view of the high number of sludge degeneration problems encountered in (aerobic and anaerobic) biological treatment stages in recent years, a physical property of additives like retention aids and flocculants is important as well – the tendency to become adsorbed in biological sludge.

Additives having no immediate and strong biocidal effects often do not influence the biological treatment unless this is facilitated by further factors. Experience has shown that operational trouble due to sludge degeneration is frequently the result of the co-occurrence of unfavourable process parameters, wastewater properties and operating conditions.

Operational trouble caused by sludge degeneration in aerobic wastewater treatment plants mainly manifests itself in the well-known phenomena of bulking, floating or fine sludge. Table 1 shows that biocidal or surfaceactive additives can cause or facilitate these forms of sludge degeneration. Floating sludge occurs only if all conditions listed are met, whereas bulking and fine sludge can already be caused by the detrimental effect of just one factor.

The term sludge degeneration has hardly been used in connection with anaerobic wastewater treatment so far, but seems to be applicable here as well because the cur-



rently encountered symptoms of sludge flotation, pellet decomposition and fine sludge are very similar to those occurring in aerobic treatment stages.

Deposit formation on anaerobic sludge pellets has occurred ever more frequently in recent years. The causes are manifold, ranging from biogenic slime production (EPS) to additive adsorption or the adherence of agglutinated sludge flocs. Most of the pellets used in anaerobic plants of the paper industry are covered by some kind of deposit (e.g. filmy, tacky, cloudy, soft, cloddy, slimy, viscous...) (Figure 1). The nature of these deposits determines, among other, whether they will influence the degradation performance by substrate limitation and/or cause pellet flotation due to impaired gas discharge or gas bubbles "adhering" to the pellet surface. Additive-induced deposits can thus have biostatic, biocidal as well as physical on the anaerobic sludge biocenosis effects.

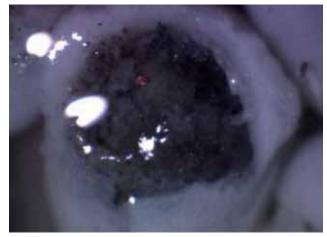


Fig. 1: Anaerobic sludge pellet with "deposit"

An anaerobic sludge biocenosis consists mainly of fermenting bacteria (hydrolysing and acidifying bacteria) and methane-forming archaebacteria. Studies into anaerobic sludge biocenosis performed in 2003/2004<sup>2</sup> and 2009/2010<sup>3</sup> revealed changes in the shares of the various bacteria (Figure 2). The share of methanogenic archaea had increased in anaerobic sludge, as had that of hydrolysing bacteria - mostly organisms belonging to the Chloroflexi and Cytophaga Flexibacter groups. The increase in hydrolysing bacteria was mainly attributable to the Chloroflexi species because the share of organisms belonging to the Cytophaga Flexibacter group had become much smaller. Chloroflexi bacteria could not be considered in the 2003/2004 study because they had not yet been discovered in anaerobic sludge back then. Because of their high share of filamentous bacteria, Chloroflexi can have a facilitating effect on sludge flotation. What is also striking in this context is the reduced shares of acidifying bacteria, especially of organisms belonging to the species of Actinobacteria, Firmicutes und Deltaproteobacteria. It is generally known that highly acidic waters are detrimental to pellet formation. The frequently separate use of hydrolysis and pre-acidifi-

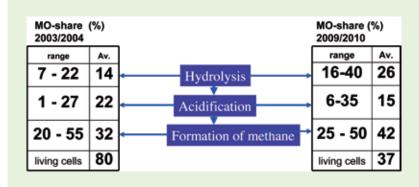


Fig. 2: Changes in anaerobic sludge biocenosis during the last 6 to 8 years<sup>2,3</sup>

cation stages in the paper industry and decreasing shares of acidifying bacteria can be assumed to lead to a high degree of acidification in paper industry wastewaters, which has a direct effect on pellet and fines formation.

As compared to 2003/2004, the average total cell count was more than six times higher in 2009/2010, whereas the share of living cells had decreased to 37 % on average (from previously 80 %). This suggests a biomass growth exceeding the removal of dead cells from the reactor during the period studied in 2009/2010. As a result, the active cells are exposed to ever higher sludge loads. This slow, unrecognised increase in sludge loading can lead to instable degradation processes over time. The described developments also indicate that most of the energy generated by degradation processes was consumed for biomass production, thus lowering the biogas yield.

Relevant influences/interactions are causes of certain forms of anaerobic sludge degeneration like pellet flotation, pellet decomposition and fine sludge formation, among others: excessive additive dosing, especially of polymers, can be a frequent cause of sludge flotation. Additives – especially adsorptive and surface-active substances – must therefore be added exactly in the amounts required to prevent sludge degeneration and sludge flotation.

Many of the additives used in the paper industry are negatively or positively charged. Anaerobic sludge is generally negatively charged, which causes the adsorption of especially positively charged polymers on anaerobic sludge particles and gradual formation of a deposit layer on the entire surface. Whether or not adhering gas bubbles or impaired gas discharge will lead to pellet/sludge flotation depends on the nature/thickness/viscosity/gas permeability of the deposits. Dosing additives only and exactly in the amounts required is therefore an imperative also from this point of view. Furthermore, the majority of cationically charged additive ingredients have an affinity to fibres and can therefore cause agglomeration with free biomass components in the inlet of anaerobic treatment stages – also with increasing solid loads (insuf-



ficient solids removal during circulation water treatment), thus promoting the formation of fines.

Pre-acidification: Paper mill wastewaters should have a degree of acidification in the range of 25 to 40 %. Higher degrees are considered to be detrimental to pellet formation or preservation. Recent studies suggest that especially the share of acidifying bacteria has significantly decreased in anaerobic pellet populations – a possible indication of causal changes in pellet stability. The mode of operation predominantly used in many anaerobic reactors of the paper industry – separate hydrolysis/pre-acidification stages and methane reactor – can have contributed to the changes observed in anaerobic sludge biocenosis.

Calcium concentrations: Calcium concentrations >800 mg/l can lead to pellet dissolution/degradation due to calcium carbonate precipitation, and to biomass being displaced from the reactor. To achieve/maintain a stable pellet structure, it is advisable to ensure Ca concentrations <200 to 300 mg/l.

Hydraulics: Operating data analyses of recently investigated plant systems and comparisons with previous studies show a tendency towards lower specific biogas production despite increased COD degradation rates. This changes the hydraulic conditions in the reactors, possibly increasing the strain on already destabilised pellets and, thus, facilitating sludge degeneration.

## Additive effects on aerobic & anaerobic degradation

The effects of additives on aerobic and anaerobic treatment processes were investigated by batch tests and semicontinuous laboratory tests (Zahn-Wellens test DIN EN ISO 9888 and PTS Method "Studies on anaerobic degradability" PTS-WA 003/97) in the scope of several research projects. The influences identified by these tests are listed in Table 2, based on their effects and for the various concentration ranges and additive groups investigated.

Generally speaking, additives tend to have no adverse effects on aerobic treatment processes if they are used as intended and added in appropriate amounts. Effects on aerobic degradation were mainly found in connection with high biocide dosages. Unlike in aerobic treatment, anaerobic treatment processes can already be influenced by very small additive concentrations. However, these influences usually relate to the facilitation and/or triggering of pellet flotation by several additive groups. Strong pellet flotation was only observed in connection with polyacrylamides, but operational trouble due to sludge losses can also be caused by additives having only minor flotation-enhancing effects in full-scale systems. This is often associated with biogas losses - biogas adheres to floating pellets. A significant but reversible decrease in COD degradation was detected only in the presence of a volume-enhancing additive.

#### Cause analysis & tests

The causes of disturbances in wastewater treatment plants are frequently unknown because: it is difficult to analyse the substances responsible for the trouble in their complex biomass matrices and many of the effects occur with considerable delay and at irregular intervals and can therefore hardly be associated with concrete operational incidents. Plant operators are more likely to identify the causes of disturbances if they investigate all relevant influences systematically. Figure 3 shows the cause analysis of disturbances in wastewater treatment plants in schematic form.

Additives	Range of concentration in mg/l		aerobic	anaerobic		
			COD-	Pellet-	COD-	Biogas
	aerob	anaerob	degradation	flotation	degradation	production
Wet strength agents	50-1,000	50-1,500	٢	nu	٢	nu
Defoarmer/cleaner	0.1–1.0 ml/l	10–50 2.5–5.0 g/l	٢	-	٢	۲
Retention aids, flocculants, precipitation aids, dispersants	0.5-20	0.5–100	٢	+ and Z $+$	٢	🙂 and 🙁
Surface sizing/binder	0.2 – 0.5	0.5-5	٢	-	٢	۲
Softener	25-50	nu	۲	nu	nu	nu
Dry strength agent	0.1 ml/l	0.5-5	۲	z+	۲	۲
Volume increasing aids	nu	50-200	nu	z+	۲	z 🙂
Biocides	0.3-100	0.5-100	🙂 and 🙁	z+ and –	٢	۲

🙂: no influence; 🙂: weakly reduced; 😕: strongly reduced;

-: none; +: few; +: heavy; Z: occasionally; nu: not determined

Tab. 2: Effects of various additive groups on anaerobic and aerobic degradation<sup>2,3,4,5</sup>



Apart from the analyses commonly used in industry, there are various further methods which can help identify the substances or factors responsible for operational trouble. Important further analytical methods, tests and their results are:

- Microscopic and image analyses of biosludge: Changes in the floc morphology and microbial composition of activated sludge
- Gene probe tests of biosludge: Microbiological composition
- Scanning electron microscopy of biosludge: Surface properties and element distribution
- Static biodegradation tests (aerobic and anaerobic), respirometer tests:
   Degradation properties, biomass activity, additive effects on degradation
- IR spectrometry of deposits and biosludge: Organic sludge components that do not belong to the population, e.g. additives

Light-microscopy studies usually provide sufficient information about the microbial conditions of the aerobic biocenosis and floc structure of aerobic sludge, but are of limited use to the identification of organisms in anaerobic sludge. Morphologically similar bacteria can only be reliably detected by advanced gene probe tech-

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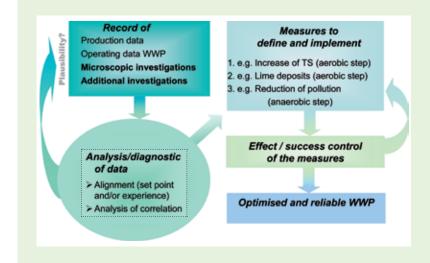


Fig. 3: Cause analysis of disturbances in wastewater treatment plants

nology<sup>6</sup>. The latter requires expert knowledge of microbiological processes; its application is highly complex and time-consuming. Gene probe technology therefore offers a tool for a hugely improved identification of biomass composition<sup>7</sup>.

#### Conclusions

Even though the technological processes used for biological wastewater treatment in the paper industry have been developed to maturity and plant operators know the operational concepts their systems are based on, disturbances continue to be a common phenomenon. Plant operators could quickly identify the causes of disturbances or avoid them in the first place if they knew more about the importance and role of the tiny protagonists involved in their treatment processes.

Microbial, chemical or morphological biomass changes occur practically every day and over extended periods in wastewater treatment plants. Improved and more powerful analyses like gene probe technology, microscopic studies, degradation and activity tests etc. can help plant operators to detect these changes and assess their importance for the process.



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