

## Heat Production Measurement of Activated Sludge by Calorimetry

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Microbial heat generation ( $Q_f$ ) is an important parameter to control a biological process.  $Q_f$  can be determined by various methods. In this study, a system for *online* calorimetric measurements for activated sludge is developed: a pilot scale bioreactor of 100 L volume as a calorimeter. The calorimetric measurements were calibrated using the heat production of *Saccharomyces cerevisiae* consuming glucose so that  $Q_f$  can also be measured using the relation of oxygen transfer rate OTR and the combustion enthalpy of glucose ( $\Delta_c H^0$ ) ( $Q_f = \Delta_c H^0 \text{ OTR}$ ). Next, the  $Q_f$  of activated sludge with glucose as substrate is measured using the verified experimental set. A constant value for OTR to calculate  $Q_f$  could not be detected because the activated sludge comprises a mixture of organic and unknown substrates and a heterogeneous flora of microorganisms. Heat energy production decreased during the experiment, pointing to a declining of microbial activity due to endogenous oxidation of the sludge.

*Key words:*

Activated sludge, calorimetric measurement, heat balance, microbial heat generation, oxygen transfer rate, yeast

### Introduction

Continuous *on-line* techniques are important for the monitoring of biological processes. Particularly the process conditions have an influence on the quality of the product and process activity. Heat generation is a universal feature of biological processes. Although non-specific, heat measurements may provide an important indication of the microbial activity, metabolism of the process and events occurring during the process.<sup>1,2</sup> For example, calorimetric measurement was used for biokinetic characterization of nitrifying population in activated sludge.<sup>3</sup>

There are at least three ways of using quantitative heat generation measurements in order to assess the state of the culture and deciding on corrective actions ensuring proper control of the process. First, it is possible to infer a wealth of information by just qualitatively looking for abrupt changes in heat release. All kinds of metabolic events, such as shifts from one substrate to another, shifts from one type of catabolism to another, occurrence of limitations, inhibitions or overflow metabolism will cause characteristic changes in the heat curve. A more quantitative technique consists of monitoring *on-line* a “heat yield” or ratio of heat release rate to the rate of production or consumption of a given

metabolite. Another technique uses heat measurement as a substitute for the oxygen uptake rate (OUR) measurement. This is based on the “oxycaloric equivalent”, which lies within  $\pm 10\%$  of 14688 kJ kg<sup>-1</sup> for all oxygen-consuming reactions, and on the fact that reactions consuming neither oxygen nor other strong oxidants produce negligible amounts of heat as compared to the former.<sup>4</sup> A modified bench scale calorimeter was employed to determine the heat generated by various microbial strains. The ratio between the heat generated and the oxygen taken up was constant at 13750 kJ kg<sup>-1</sup> O<sub>2</sub> throughout all experiments as expected from theoretical considerations for strongly aerobic process.<sup>5</sup> The correlation between heat generation and oxygen uptake was studied. In another study, heat yield on oxygen was found to be 13750 and 13500 kJ kg<sup>-1</sup> O<sub>2</sub> for continuous and dynamic experiments, respectively. The metabolic heat production was also estimated from heats of combustion of substrate, biomass and ethanol.<sup>6,7</sup>

In biological processes, the knowledge of the generated microbial heat is important for three reasons:

1. In exothermic biochemical reactions, which have to progress at a constant temperature, it is important to determine the heat energy to be removed from the system.<sup>8,9</sup>

2. In autothermal thermophilic aerobic treatment plants for high-strength and/or high tempera-

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ture wastewater and autothermal thermophilic aerobic digestion plants for sewage sludge, it is important to know the amount of the heat supplied by the waste input to the system, in order to keep the thermophilic temperature.<sup>10,11</sup>

3. Heat measurements may constitute a valuable tool for on-line determinations of the concentration of biomass, oxygen uptake, CO<sub>2</sub> evolution and product formation in a fermentation system.<sup>12</sup> Any change in heat flux derived from the addition of a substrate or toxic material can be measured in order to assess biological parameters or toxicity effects.<sup>13</sup> For example, calorimetry provided a robust and reliable tool to study the bulk-kinetics of composting.<sup>14</sup>

Generated heat can be determined by calorimetric measurements. Different types of biological reaction calorimetry systems are presented in Marison *et al.* (1998).<sup>1</sup> Biological heat generation can be evaluated with parameters such as the consumption of the chemical oxygen demand (COD), the volatile solids and OUR. Messenger *et al.* (1990) showed that the close correlation and rapidity of response between the microbial heat generation rate and the OUR make the OUR a pivotal parameter in the modelling and design of autothermal thermophilic aerobic reactors.<sup>15</sup>

The linear correlation between heat generation rate and oxygen consumption has been verified in a pure culture<sup>6,7,16,17,18</sup> and in activated sludges.<sup>13,19</sup> In this study, the relation between heat generation and oxygen transfer rate (OTR) is investigated for a pure culture (*Saccharomyces cerevisiae*) and activated sludge using also glucose as substrate.

## Materials and methods

Experiments were conducted in a steel reactor of 100 L volume. Its features are given in Table 1. An oxygen sensor (WTW Oxi 323 B) for the dissolved oxygen (DO) and a Pt-100 temperature sensor were mounted onto the reactor body in order to monitor the biochemical reactions. Furthermore,

Table 1 – Features of the reactor

Height of reactor	0.830 m
Diameter of reactor	0.408 m
Liquid volume / $V_L$	60 L
Stirrers	2 x 6-blade Rushton turbines
Stirrer diameter / $D_R$	0.200 m
4 baffles	0.5 x 0.04 m
37 holes on the air ring with diameter	0.003 m

CO<sub>2</sub> and O<sub>2</sub> concentrations of the exhaust gas were measured *on-line* by an analyzer (SERVOMEX). Values of CO<sub>2</sub> and O<sub>2</sub> concentrations were used to calculate carbon dioxide transfer rate (CTR) and OTR;<sup>20</sup> both parameters were used for monitoring the biochemical reactions. The humidity and the temperature of the air sent to the reactor were measured by a humidity sensor (VAISALA) and a Pt-100 temperature sensor, respectively. The reactor was connected to a SCADA system. In this study, SCADA software was used to record the signals coming from the sensors. The reactor was insulated on the outside entirely, so that it could be used as a calorimeter. The general heat balance around the bioreactor can be expressed as eq. (1). The contributions of each term in eq. (1) were calculated using eqs. (2–11).

The experiments to determine the heat generated by the biological reaction under aerobic conditions were carried out in two stages:

### 1. Monitoring a purely oxidative, glucose-consuming baker's yeast, *Saccharomyces cerevisiae*, fermentation for calibration of the calorimeter.

The reactor was fed with 10 L yeast cream with a dry solid content of 12.9 % and filled up to 60 L with tap water. The culture was fed with molasses solution (glucose concentration 319.5 g L<sup>-1</sup>) at flow rate of 30 mL min<sup>-1</sup>. In order to keep the pH at 5.5, the culture was controlled by feeding (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> solution, consisting 21 % N. In order to keep DO > 1 mg L<sup>-1</sup>, the reactor was aerated at a rate of 122.5 L min<sup>-1</sup> and mixed with 160 rpm under the interior pressure of 50 kPa.

### 2. Monitoring activated sludge-consuming

**glucose.** The glucose consumption of the activated sludge was monitored using the sludge obtained from the Activated Sludge Unit of the Wastewater Treatment Plant of PAKMAYA in Turkey. The sludge had the following properties: Dry mass concentration 18.05 g L<sup>-1</sup>, organic carbon concentration 5 g L<sup>-1</sup> and pH 6.52. 1.2 kg glucose was added to the activated sludge of 60 L. In order to keep DO > 1 mg L<sup>-1</sup>, the reactor was aerated at a rate of 3 L min<sup>-1</sup> and mixed with 160 rpm under interior pressure of 10 kPa.

## Process models

### Biological heat production model

The heat generated by biochemical reactions can be calculated with a heat balance around the reactor (Fig. 1). The heat balance is given in eq. (1).<sup>8</sup>

$$Q_t = Q_f + Q_r - Q_v - Q_a - Q_l \quad (1)$$

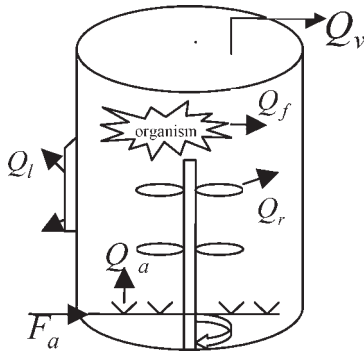


Fig. 1 – Heat balance around a bioreactor  $Q_v$ : heat loss by evaporation;  $Q_a$ : cooling by aeration;  $Q_f$ : microbial heat production;  $Q_r$ : heat generated through agitation;  $Q_i$ : loss generated from reactor;  $F_a$ : air flow rate

The total heat energy  $Q_i$  is calculated according to eq. (2).

$$Q_i = \frac{(\rho_L V_L c_{p,L} + m_{steel} c_{p,steel}) \Delta T_f}{\Delta t V_L} \quad (2)$$

Here are  $c_{p,L} = 4.18 \text{ kJ kg}^{-1} \text{ K}^{-1}$ ,  $c_{p,steel} = 0.5 \text{ kJ kg}^{-1} \text{ K}^{-1}$  and  $m_{steel} = 175 \text{ kg}$ .

It is assumed that the values of  $\rho$  and  $c_p$  for the activated sludge are almost equal to that of water.

The stirrer's effect on the homogeneity of the reactor content and on the oxygen transfer is high.  $Q_r$  can be calculated by eqs. (3) to (7).

$$Q_r = M \frac{P_R}{V_L} \quad (3)$$

$$P_R = Ne \rho_L N^3 D_R^5 \quad (4)$$

$$Ne = z(Ne_0 + 187 Q Fr^{-0.32} (D_R/D)^{1.53} - 4.6Q^{1.25} / (1 + 136Q (D_R/D)^{1.14})) \quad (5)$$

$$Q = F_a / ND_R^3 \quad (6)$$

$$Fr = N^2 D_R / g \quad (7)$$

Here are  $\rho_L = 1000 \text{ kg m}^{-3}$   $z = 2$ ,  $Ne_0 = 4.9$  and  $g$ (acceleration due to gravity) =  $9.81 \text{ m s}^{-2}$ . The constant  $M$  is determined experimentally as 0.8 for a fermentation temperature of  $30 \text{ }^\circ\text{C}$ .

The heat loss by evaporation, the fourth term of heat balance, is calculated in eqs. (8) and (9) regarding the water load in air,  $m_{ss}$  ( $\text{kg water m}^{-3}$  air), and the latent heat of water of evaporation,  $q_L$  ( $\text{kJ kg}^{-1}$ ). The absolute humidity ( $x$ ), and the density of air ( $\rho_a$ ) can be calculated with the help of the Mollier Diagram. It is assumed that the exhaust gas is saturated i.e. the relative humidity is 100 % and the temperature of the exhaust gas is equal to the interior temperature of the reactor.

$$Q_v = m_{ss} q_L F_a / V_L \quad (8)$$

$$m_{ss} = x_w \rho_{a,w} - x_a \rho_{a,a} \quad (9)$$

Because the temperature of the feed air is lower than the interior temperature of the reactor, a part of the generated heat is removed from the system with the exhaust gas. The heat removed from the system by aeration ( $Q_a$ ) can be calculated according to eq. (10) with the help of  $c_{p,a}$  ( $\text{J kg}^{-1} \text{ K}^{-1}$ ) and  $\Delta T_a$  (K).

$$Q_a = c_{p,a} \Delta T_a F_a \rho_{a,a} / V_L \quad (10)$$

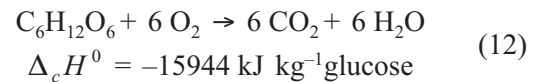
The heat energy released from non-insulated points like view glasses on the reactor body or pipes and joints can be calculated with eq. (11).

$$Q_l = 0.0643 \Delta T_f + 0.0945 \quad (11)$$

Values of  $Q_r$ ,  $Q_r$ ,  $Q_a$ ,  $Q_v$  and  $Q_l$  can be calculated with eqs. (2) to (11). The calculated values from these equations put into eq. (1) give the value of biological heat production.

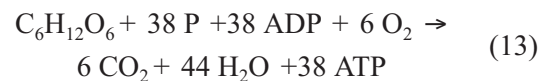
### Conceptual approach

For estimating the heat generation of bioprocesses consuming glucose the following reaction, eq. (12), is considered:



During the oxidation of glucose, chemical energy, i.e. combustion enthalpy of glucose,  $\Delta_c H^0$ , is released.  $\Delta_c H^0$  is found to have a range of values in the literature:  $15400 \text{ kJ kg}^{-1}$ ,<sup>21</sup>  $15667 \text{ kJ kg}^{-1}$ <sup>22</sup> and  $15967 \text{ kJ kg}^{-1}$ .<sup>23</sup>

This energy can then be used for ATP synthesis according to eq. (13).



As the generation of 1 mol ATP requires about 50 kJ, the free reaction enthalpy which is used for ATP formation can be calculated

$$\Delta_c H^0 = (38 \text{ mol ATP mol}^{-1} \text{ glucose}) (50 \text{ kJ mol}^{-1} \text{ ATP}) = 1900 \text{ kJ mol}^{-1} \text{ glucose} = 10555.6 \text{ kJ kg}^{-1} \text{ glucose}$$

The other part of  $\Delta_c H^0$  namely  $-15944 + 10555.6 = -5388.4 \text{ kJ kg}^{-1} \text{ glucose}$  will be considered as heat. One has therefore to conclude that in the oxidative utilization of glucose about 1/3 of the energy content (of the glucose) is deliberated in the form of heat which has to be removed from the culture media. In practice, it is not common to calculate the heat generation from energy/ATP balances. It is simpler to couple the heat generation with OUR.<sup>24</sup>

$$Q_f = \Delta_c H^0 (\text{kJ kg}^{-1} \text{O}_2) \text{OUR} (\text{kg O}_2 \text{L}^{-1} \text{min}^{-1}) \quad (14)$$

For the proportionality constant ( $\Delta_c H^0$ ), the value that is the oxygen quantity required for the complete combustion of glucose, was reported as 13750 kJ kg<sup>-1</sup> O<sub>2</sub> by Luong and Volesky (1982),<sup>25</sup> 13625–16250 kJ kg<sup>-1</sup> O<sub>2</sub> by Bailey and Ollis (1986),<sup>24</sup> 13438–15000 kJ kg<sup>-1</sup> O<sub>2</sub> by Kemp (2000),<sup>26</sup> 14125 ± 906 kJ kg<sup>-1</sup> O<sub>2</sub> by Weppen (2001),<sup>14</sup> 16438 to 17031 kJ kg<sup>-1</sup> O<sub>2</sub> by Yonsel *et al.* (2001),<sup>8</sup> 14688–14094 kJ kg<sup>-1</sup> O<sub>2</sub> by Aulenta *et al.* (2002),<sup>13</sup> and 13750–13500 kJ kg<sup>-1</sup> O<sub>2</sub> by Türker (2004).<sup>7</sup>

By Messenger *et al.* (1990), the relation (energy released per substrate oxidized ( $\Delta_c H^0$ ) with the energy released per oxygen consumed ( $Y_h$ )) for a particular substrate is expressed in eq. (15).<sup>15</sup> Eq. (15) is based on the assumption that no exothermic reactions occur without the transfer of electrons to oxygen, a reasonable assumption for a biological oxidation system.

$$Y_h = (1/Z)(\Delta_c H^0 - BEN_{ATP})(42/32) \quad (15)$$

In this study, the following model equation is used to calculate the biologically generated heat using OTR and the proportionality constant ( $\Delta_c H^0$ ).

$$Q_{f(model)} = \Delta_c H^0 \text{OTR} \quad (16)$$

There is no sink other than the microorganisms which can consume the oxygen so that the OTR is equal to the oxygen uptake rate (OUR). Eq. (16) is already verified by Yonsel *et al.* (2001)<sup>8</sup> and (2007).<sup>17</sup>

## Results and discussion

### Evaluation of biological heat generated by *S. cerevisiae*

In this study, the purely oxidative consumption of glucose by *S. cerevisiae* under aerobic conditions is used here to calibrate the calorimetric measurements made with activated sludge. The respiratory quotient (RQ) defined as the ratio CTR to OTR ( $\text{RQ} = (\text{CTR}/\text{OTR})(32/44)$ ) is an important parameter for monitoring the decomposition of glucose. RQ variations are usually explained by the fact that the relative amounts of CO<sub>2</sub> generated and O<sub>2</sub> consumed are dependent on the degree of oxidation of the substrate and the pathways by which it is metabolized.<sup>27,28</sup> For carbohydrates, lipids, and proteins the RQ has values of 1.0, about 0.72, and about 0.84 (meat protein), respectively.<sup>18</sup> The theoretical RQ when hexoses are aerobically metabolized is 1.07,<sup>8</sup> but this value changes to 0.8 or 1.33 if fat or organic acids are the principal substrates.<sup>29</sup> High RQ values can also be obtained if ox-

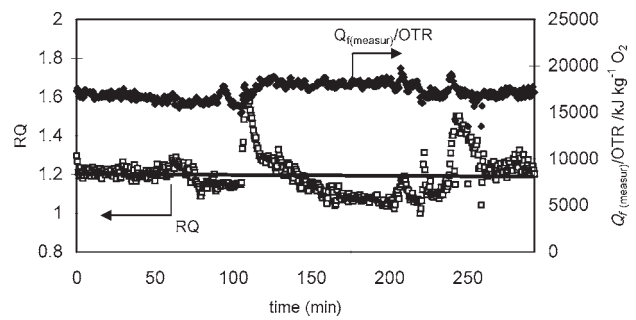


Fig. 2 – RQ and  $Q_{f(measur)}/\text{OTR} = \Delta_c H^0$  versus time in purely oxidative consumption of glucose by *S. cerevisiae*

xygen drops below a critical value, leading to the onset of fermentation. Fig. 2 shows that measured RQ values are between 1.09 and 1.29 during the decomposition of glucose by *S. cerevisiae*. These values correspond well with the literature.

If the theoretical model approach and the calorimetric measurements are accurate,  $Q_{f(measur)}$  must be equal to  $Q_{f(model)}$ . That is

$$Q_{f(measur)} = Q_{f(model)} = \Delta_c H^0 \text{OTR} \quad (17)$$

Yonsel and co-workers used a 1500 L pilot reactor as a mega-calorimeter and showed that the value of microbial heat generation  $Q_{f(model)}$  calculated via model eq. (17) is equal to measured value  $Q_{f(measur)}$  via calorimetry.<sup>8,17</sup>

Fig. 2 shows the measured values of  $Q_{f(measur)}/\text{OTR} = \Delta_c H^0$  during an experiment of 300 min. According to Fig. 2,  $\Delta_c H^0$  has values between 16331 and 18077 kJ kg<sup>-1</sup> O<sub>2</sub>. As mentioned before, the values in the literature are between 16426 and 17031 kJ kg<sup>-1</sup> O<sub>2</sub>. A parity plot gives the linear relation of  $Q_{f(model)}$  with  $Q_{f(measur)}$  as nearly 1 (Fig. 3).

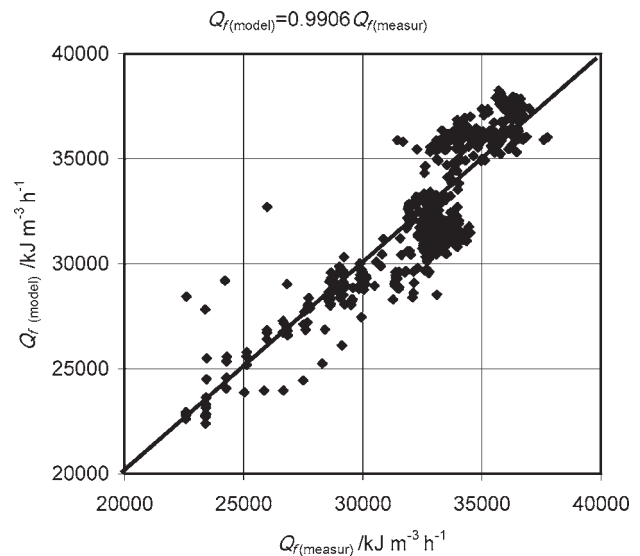


Fig. 3 – Relation between  $Q_{f(model)}$  and  $Q_{f(measur)}$  in purely oxidative consumption of glucose by *S. cerevisiae*

These results are proof that the calorimetric measurements and the model equation are accurate.

### Evaluation of the biological heat generated by activated sludge

After the accuracy of the calorimetric measurements, using the known system *Saccharomyces cerevisiae* with a defined pathway is verified, the heat production of activated sludge is examined.

Fig. 4 shows the courses of produced heat energy,  $Q_{f(\text{measur})}$ , OTR and RQ during the experiment of 1100 min.  $Q_{f(\text{measur})}$  is decreasing during the experiment from 4300 to 1900  $\text{kJ m}^{-3} \text{h}^{-1}$ . OTR slightly decreases from 0.22 to 0.18  $\text{kg m}^{-3} \text{h}^{-1}$ , CTR slightly increases from 0.22 to 0.28  $\text{kg m}^{-3} \text{h}^{-1}$  (not shown here) so that the RQ value also increases from 0.7 to 1.15.

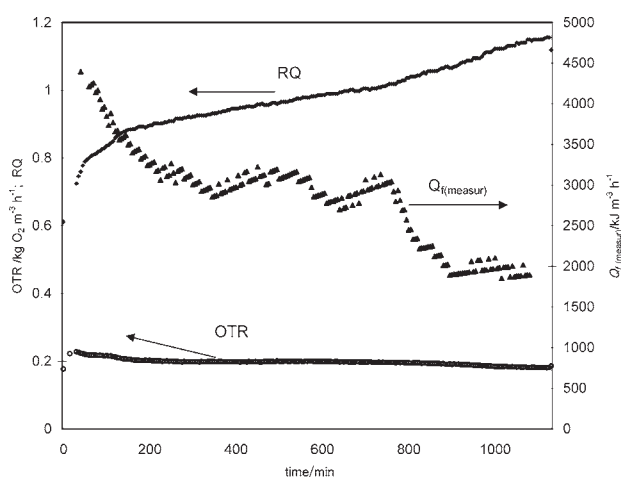


Fig. 4 – RQ, OTR and measured heat energy  $Q_f$  of activated sludge, in presence of glucose

The rate of  $Q_{f(\text{measur})}$  to OTR gives  $\Delta_c H^0$  as mentioned in eq. (17). Fig. 5 shows that this relation is not linear, i.e.  $\Delta_c H^0$  has not a constant value, but  $\Delta_c H^0$  value decreases from 20000  $\text{kJ kg}^{-1} \text{O}_2$  to 10000  $\text{kJ kg}^{-1} \text{O}_2$  during the experiment.

These values are outside the range of  $\Delta_c H^0 = 16426$  to 17031  $\text{kJ kg}^{-1} \text{O}_2$  as given in Fig. 2, hence  $Q_{f(\text{model})} = \Delta_c H^0 \text{ OTR}$  equation is not applicable for the activated sludge with mixed substrate.

A trend relation between OTR and  $Q_f$  can be detected, the results of the experiment can be described using the eq. (18) (Fig. 6)

$$Q_{f(\text{measur})} = 78567 \text{ OTR} - 12557 \quad (18)$$

$$R^2 = 0.9244$$

The experiment with *S. cerevisiae* and glucose was a single organism, single substrate and single

pathway reaction. Here it was possible to verify the combustion enthalpy of glucose by calorimetric measurements. However, in the experiment with activated sludge we have a mixture of microorganisms. Glucose is not the only substrate, but several organic materials in the sludge, and the microorganisms themselves (endogenous oxidation) serve as substrates. The major requirement for oxygen in aerobic metabolism is as the terminal electron acceptor in the electron transport chain, and therefore its consumption is intimately related to the generation of ATP. It is known that no uniformity of components in the ATP synthesizing apparatus exists among bacterial species and that a bacterium has the ability of using alternate pathways of electron transport, switching from one pathway to another depending on conditions of the growing medium such as the substrate composition or DO concentration. Therefore, the number of molecules of ATP synthesized per atom of oxygen reduced is not constant in a mixed culture but in fact can vary significantly with changes in environmental conditions or changes in predominant species. The close relation between ATP production and cell synthesis is evident. This is not the case with dissolved oxygen uptake rate. Bacteria have the ability of uncoupling cell synthesis and respiration in part through the use of alternate electron transport pathways with different sites of oxidative phosphorylation and through ATP hydrolysis. Furthermore, the uptake rate is obtained by the measurement of the DO concentration in the mixed liquor, an external element of the cell, and thus is subject to interferences from the medium and dependent on the efficiency of oxygen penetration into the biological floc.<sup>30</sup>

The value of RQ during the experiment increases from 0.7 up to 1.15 (Fig. 4) remaining in the range of theoretical values for aerobically consumed hexoses.

As mentioned before we here have mixed culture and multiple substrate system. Although there is glucose as a substrate, the reactions of sludge decomposition run in parallel. During the experiment, the beginning pH of 6.5 increased to pH 9 due to the alkaline sludge decomposition at the end. Considering the pH drift, the RQ curve and the practically constant OTR value during the experiment, it is very likely that the system was operated in partial oxygen limitation and the glucose degraded also under anaerobic conditions. Oxygen limitation may occur even if there is dissolved oxygen in the water phase and the system is well mixed. The explanation for this is that in activated sludge the cells are packed in flocs, so in addition to the diffusion across the cell membrane and in the cytoplasm the oxygen mass transfer is limited by the diffusion in the flocs as well. This phenomenon can be detected

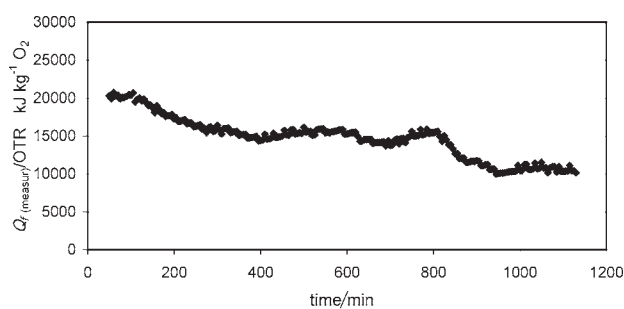


Fig. 5 –  $Q_{f(\text{measur})}/\text{OTR} = \Delta_c H^0$  versus time for activated sludge, in presence of glucose

by RQ measurements. In this case, the RQ was slightly increasing during the experiment, even if the sludge was becoming definitely alkaline (at alkaline pH values, all the carbon dioxide remains in solution). The presence of a partial oxygen limitation can explain the unanticipated relation of OTR and  $Q_f$ .

Decreasing heat energy production points to a declining of microbial activity. It is common that activated sludge lose activity in time not only because of decreasing substrate concentration, but also because the microorganisms themselves serve as substrate (endogenous oxidation). Therefore, the flora composition of the sludge changes in time and the sludge as a whole loses activity. This phenomenon can be detected also in our experiment.

## Conclusion

In this work, a system for *online* calorimetric measurements for activated sludge is developed. Using *Saccharomyces cerevisiae* as the single microorganism and glucose as the single substrate a purely oxidative single pathway reaction experiment is composed. The known value of combustion enthalpy of glucose could be measured and verified by calorimetry. It is also shown, that biological heat generation ( $Q_f$ ) can be determined also using the oxygen transfer rate (OTR). The relation of OTR with the  $Q_f$  of the system *Saccharomyces cerevisiae* consuming glucose as substrate can be calculated using the enthalpy  $\Delta_c H^0$  according to eq. (16). This experimental set, i.e. a pilot scale bioreactor of 100 L volume, is used for calorimetric measurements with activated sludge and glucose as substrate. A constant value between  $Q_f$  and OTR could not be detected. It is difficult to estimate heats of reaction for organic wastes from standard enthalpy values because the sludge we used comprised a mixture of organic and unknown composition. In this case, the standard enthalpy value of glucose was of little use, because the sludge itself was used also as substrate.

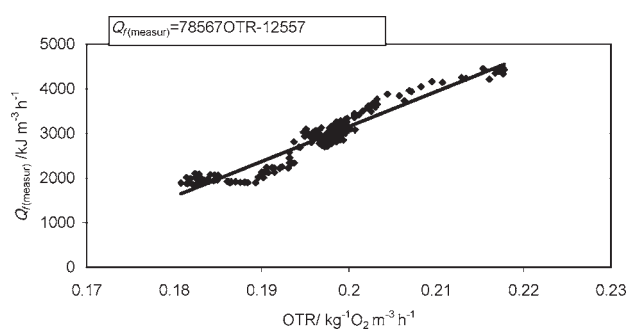


Fig. 6 – The relation between  $Q_f$  and OTR of activated sludge in presence of glucose

The calorimetric approach is undoubtedly the most accurate way to determine heats of reaction of mixtures of organics. However, calorimetric tests are not routine for most water quality laboratories and analysis by laboratories equipped for this purpose is usually required. As a result, a number of approximate formulas based on both theoretical and experimental approaches have been developed. One of the approaches requires an ultimate analysis of the waste to determine the percentages of carbon, hydrogen and oxygen.<sup>31,32</sup> Another approach requires analysis of the volatile solid content. A rule of thumb which is reasonably accurate for most organics is that about  $59547 \pm 3502$  kJ are released per g COD of the waste.<sup>31</sup> Here we found a relation given in eq. (18) for heat production and oxygen transfer rate for a mixed culture, i.e. activated sludge, and glucose as a surplus substrate.

## ACKNOWLEDGEMENTS

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## Nomenclature

- $B$  – phosphoryll bond energy,  $\text{J mol}^{-1}$  ATP
- CTR – carbon dioxide transfer rate,  $\text{kg m}^{-3} \text{h}^{-1}$
- $c_p$  – specific heat capacity,  $\text{kJ kg}^{-1} \text{K}^{-1}$
- $D$  – reactor diameter, m
- $D_R$  – stirrer diameter, m
- $\Delta T_a$  – air temperature difference at entrance and exit of reactor, K
- $\Delta T_f$  – fermentation temperature difference, K
- $\Delta_c H^0$  – combustion enthalpy,  $\text{kJ kg}^{-1}$  glucose or  $\text{kJ kg}^{-1} \text{O}_2$
- $\Delta t$  – fermentation time, h
- $E$  – fractional efficiency of energy utilization, –
- $F_a$  – air flow rate,  $\text{m}^3 \text{h}^{-1}$
- $Fr$  – Froude number, –
- $m_{ss}$  – water load of air,  $\text{kg m}^{-3}$
- $M$  – constant, 0.8
- $m$  – mass, kg

$N$  – stirrer's speed,  $s^{-1}$   
 $N_{ATP}$  – number of ATP moles generated per mole substrate oxidized,  $\text{mol ATP mol}^{-1}$   
 $Ne$  – Newton number, –  
 $Ne_0$  – Newton number for Rushton turbine, –  
 $OTR$  – oxygen transfer rate,  $\text{kg m}^{-3} \text{h}^{-1}$   
 $P_R$  – energy input via stirrer,  $\text{kJ h}^{-1}$   
 $Q$  – flow number, –  
 $q_L$  – latent heat of water of evaporation,  $\text{kJ kg}^{-1}$   
 $Q_v$  – heat loss by evaporation,  $\text{kJ m}^{-3} \text{h}^{-1}$   
 $Q_a$  – cooling by aeration,  $\text{kJ m}^{-3} \text{h}^{-1}$   
 $Q_f$  – microbial heat production,  $\text{kJ m}^{-3} \text{h}^{-1}$   
 $Q_r$  – heat generated through agitation,  $\text{kJ m}^{-3} \text{h}^{-1}$   
 $Q_l$  – loss generated from reactor,  $\text{kJ m}^{-3} \text{h}^{-1}$   
 $Q_t$  – total heat energy in system,  $\text{kJ m}^{-3} \text{h}^{-1}$   
 $RQ$  – respiratory quotient, –  
 $\rho$  – density,  $\text{kg m}^{-3}$   
 $V_L$  – liquid volume,  $\text{m}^3$   
 $x$  – absolute humidity,  $\text{kg water kg}^{-1} \text{air}$   
 $Y_h$  – specific heat yield,  $\text{MJ kg}^{-1} \text{O}_2$   
 $z$  – stirrer number, –  
 $Z$  – moles oxygen utilized per mole substrate oxidized,  $\text{mol O}_2 \text{mol}^{-1}$

### Subscripts

$a$  – air  
 $L$  – liquid  
 $w$  – exhaust gas  
 $\alpha$  – inlet gas

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