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# Determinación de los coeficientes biocinéticos de un sistema aerobio para remoción de almidón de papa

SANITARY AND ENVIRONMENTAL ENGINEERING

## Determination of biokinetic coefficients of an aerobic system for potato starch removal

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#### Abstract

This study presents the performance of a laboratory scale completely mixed aerobic reactor for starch wastewater treatment. The reactor was operated for 119 days, divided into 5 operational phases which included the start-up and the variation of cell-residence time. In order to determine the biokinetic coefficients for starch removal, the cell-residence time was varied between 20 to 4 days, and the system operation was set to a hydraulic retention time of 24 hours with a completely mixed reactor setup. The chemical oxygen demand concentration was 1000mg/L in the inflow of the reactor and dissolved oxygen remained above 2mg/L.

In this experimentation time, the parameters such as pH, chemical oxygen demand (in the influent and effluent), dissolved oxygen and mixed liquor suspended solids were monitored. The calculation of biokinetic constants was done by an approximate method, using the graphical Lineweaver-Burk method. The biokinetic coefficients determined were for maximum substrate removal rate ( $k_0$ ), half saturation rate

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constant or affinity constant  $(k_m)$ , sludge yield coefficient (Y), and microbial decay or indigenous respiration rate  $(k_e)$ .

The results showed that Y,  $(k_0)$ , and  $(k_m)$ , were in the range of 0.3- 0.7 mgSSV/mgBOD<sub>5</sub>, 2- 8 day<sup>1</sup> and 40-120 mg/L (COD- BOD<sub>5</sub>), respectively. Values of the coefficients were within the range of those reported for conventional activated sludge processes.

Keywords: Biological processes; Bioremediation; Starch removal; Water treatment;

#### Resumen

Este estudio presenta el rendimiento de un reactor aerobio completamente mezclado, a escala de laboratorio, para el tratamiento de aguas residuales provenientes de la extracción de almidón de papa. El reactor se operó durante 119 días divididos en 5 fases de operación que incluyeron la puesta en marcha y la variación del tiempo de retención celular. Con el fin de determinar los coeficientes biocinéticos, el tiempo de retención celular se varió entre 20 y 4 días, y el funcionamiento del sistema se estableció con tiempo de retención hidráulica de 24 horas a una configuración hidráulica de mezcla completa. La concentración de demanda química de oxígeno fue de 1000 mg/ en la entrada del reactor y el oxígeno disuelto permaneció por encima de 2 mg/L.

En este tiempo de experimentación se monitorearon parámetros como pH, demanda química de oxígeno (en el afluente y efluente), oxígeno disuelto y sólidos suspendidos del licor de mezcla. El cálculo de las constantes biocinéticas se realizó mediante un método aproximado, utilizando el método gráfico Lineweaver-Burk. Los coeficientes biocinéticos determinados fueron: La tasa máxima de remoción de sustrato  $(k_0)$ , la tasa media de saturación o constante de afinidad  $(k_m)$ , el coeficiente de rendimiento de lodo (Y) y la tasa de respiración endógena  $(k_e)$ .

Los resultados mostraron que Y, ( $k_0$ ), y ( $k_m$ ), estuvieron en un rango de 0,3- 0,7 mgSSV/mgBOD5, 2- 8 1/día y 40-120 mg/L (COD- BOD5), respectivamente. Estos valores de los coeficientes estuvieron en el rango de los coeficientes reportados en la literatura en el tratamiento convencional de lodos activados empleados para el tratamiento de residual doméstica.

*Palabras clave:* Biotecnología; Bioremediación; Remoción de almidón; Tratamiento de agua.

CODChemical oxygen demand (mg COD/L) $\Theta_c$ Cell-residence time (day) Maximum specific growth rate (mgVSS/mg CODday)BODBiochemical oxygen demand (mg BOD/L) $\mu_{max}$ Maximum specific growth rate (mgVSS/mgCODday)CMASCompletely mixed activated sludge $\mu$ Specific growth rate (mgVSS/mgCODday)XMixed liquor suspended solids (mgVSS/L) $V$ Reactor useful volume (m <sup>3</sup> )QFlow (L/s o m3/day) $Q_w$ Reactor outflowHRTHydraulic retention time (hours) $Q_r$ Reactor internal recirculation flowSVISludge volumetric Index $F_0$ Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L) $F$ Concentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L) $k_0$ Maximum substrate removal rate ( $k_0 = \mu_{max}/Y$ ) (mgCOD/mgVSS day)	Nomenclature						
BODBiochemical oxygen demand (mg BOD/L)μmaxMaximum specific growth rate (mgVSS/mg CODday)CMASCompletely mixed activated sludgeμSpecific growth rate (mgVSS/mgCODday)XMixed liquor suspended solids (mgVSS/L)μSpecific growth rate (mgVSS/mgCODday)QFlow (L/s o m3/day)QwReactor useful volume (m³)HRTHydraulic retention time (hours)QrReactor internal recirculation flowSVISludge volumetric IndexF0Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L)k₀Maximum substrate removal rate (k₀ = μmax /Y) (mgCOD/mgVSS day)	COD	Chemical oxygen demand (mg COD/L)	Өс	Cell-residence time (day)			
CMASCompletely mixed activated sludge $\mu$ Specific growth rate (mgVSS/ mgCODday)XMixed liquor suspended solids (mgVSS/L)VReactor useful volume (m³)QFlow (L/s o m3/day)QwReactor outflowHRTHydraulic retention time (hours)QrReactor internal recirculation flowSVISludge volumetric IndexF0Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L)k_0Maximum substrate removal rate (k_0 = $\mu_{max}$ /Y) (mgCOD/mgVSS day)k_nHalf saturation rate constant (mgCOD/L)Microbial decay or indigenous respiration rate	BOD	Biochemical oxygen demand (mg BOD/L)	$\mu_{max}$	Maximum specific growth rate (mgVSS/mg CODday)			
XMixed liquor suspended solids (mgVSS/L)VReactor useful volume (m³)QFlow (L/s o m3/day)QwReactor outflowHRTHydraulic retention time (hours)QrReactor internal recirculation flowSVISludge volumetric IndexF0Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L)k₀Maximum substrate removal rate (k₀ = $\mu_{max}/Y$ ) (mgCOD/mgVSS day)k₀Half saturation rate constant (mgCOD/L)k	CMAS	Completely mixed activated sludge	μ	Specific growth rate (mgVSS/ mgCODday)			
QFlow (L/s o m3/day)QwReactor outflowHRTHydraulic retention time (hours)QrReactor internal recirculation flowSVISludge volumetric IndexF0Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L)k_0Maximum substrate removal rate (k_0 = $\mu_{max}/Y$ ) (mgCOD/mgVSS day)k_rHalf saturation rate constant (mgCOD/L)k	Х	Mixed liquor suspended solids (mgVSS/L)	V	Reactor useful volume (m <sup>3</sup> )			
HRTHydraulic retention time (hours) $Q_r$ Reactor internal recirculation flowSVISludge volumetric IndexF0Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L) $k_0$ Maximum substrate removal rate ( $k_0 = \mu_{max}/Y$ ) (mgCOD/mgVSS day)k_rHalf saturation rate constant (mgCOD/L)k	Q	Flow (L/s o m3/day)	$Q_{\mathrm{w}}$	Reactor outflow			
SVISludge volumetric Index $F_0$ Inffluent COD concentration (mgCOD/L)VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L) $k_0$ Maximum substrate removal rate ( $k_0 = \mu_{max}/Y$ ) (mgCOD/mgVSS day)k_nHalf saturation rate constant (mgCOD/L)k_n	HRT	Hydraulic retention time (hours)	$Q_{r}$	Reactor internal recirculation flow			
VSSVolatile suspended solids (mg VSS/L)FConcentration of growth-limiting substrate (mgCOD/L)TSSTotal suspended solids (mg TSS/L) $k_0$ Maximum substrate removal rate ( $k_0 = \mu_{max}/Y$ ) (mgCOD/mgVSS day)k_rHalf saturation rate constant (mgCOD/L) $k_c$	SVI	Sludge volumetric Index	$F_0$	Inffluent COD concentration (mgCOD/L)			
TSSTotal suspended solids (mg TSS/L) $k_0$ Maximum substrate removal rate ( $k_0 = \mu_{max} / Y$ ) (mgCOD/mgVSS day)k_rHalf saturation rate constant (mgCOD/L)k	VSS	Volatile suspended solids (mg VSS/L)	F	Concentration of growth-limiting substrate (mgCOD/L)			
k Half saturation rate constant (mgCOD/L) K Microbial decay or indigenous respiration rate	TSS	Total suspended solids (mg TSS/L)	$\mathbf{k}_0$	Maximum substrate removal rate ( $k_0 = \mu_{max} / Y$ ) (mgCOD/mgVSS day)			
$k_{\rm m}$ Than subtraction rate constant ( $m_g cod/L$ ) $k_{\rm e}$ (1/day)	$\mathbf{k}_{\mathrm{m}}$	Half saturation rate constant (mgCOD/L)	k <sub>e</sub>	Microbial decay or indigenous respiration rate (1/day)			
YYield coefficient (mgVSS/mgCOD)OCROxygen consumption rate gO2/gSSVday)	Y	Yield coefficient (mgVSS/mgCOD)	OCR	Oxygen consumption rate gO2/gSSVday)			

#### 1 **INTRODUCTION**

Surface water pollution is a serious problem for many developing countries. The main causes are industrial and domestic wastewater discharges that contain excessive concentrations of nitrogen, phosphorous, and organic pollutants into natural waters such as rivers and lakes (1). Unfortunately, one source of this type of pollution are starch factories, which are classified as polluting industries around the world (2). In Latin America, this issue is especially problematic due to the lack or low efficiency of wastewater plants to treat general wastewater. In many cases, this raw wastewater is discharged directly into natural water.

The potato represents one of the most important crops for the food sustenance of the world population but, at the same time, it is useful for diverse industrial processes and starch production. This starch is used as an industrial

raw material in food, textile, chemical, medicines, and others(2,3). However, the starch production process requires a huge amount of water for to wash the feedstocks, perform the starch extraction process, and clean the pipelines and instruments. Most of the water that is used in the process is converted into wastewater (2). It is estimated that production each ton of starch will 10-20 m<sup>3</sup> wastewater generate between discharge(3), containing organic products such as starch, proteins, amino acids sugars, and potassium (4).

The main concern arising due to this kind of process is that wastewater discharges from starch factories usually have a high organic load, which causes a drastic decrease in the amount of oxygen dissolved in natural water resources (2). Usually, wastewater from starch production contains chemical oxygen demand (COD) concentrations between 5000 and 50000 mg/L,

biochemical oxygen demand concentration between 3000 and 30000 mg/L, and suspended solids concentration between 1000 and 5000 mg/L(3). Therefore, effective treatment of the starchy wastewater is critical.

Different kinds of processes can be used to treat wastewater from starch production. Both physicochemical and biological technologies can present good efficiencies. However, the biological process has gained a lot of attention in the last few years. Some reasons for this are that these processes present good efficiencies with low operation costs, and are sustainable and environmentally friendly technologies. The aerobic process is a biological wastewater technology that has shown good efficiencies in the removal of conventional organic pollutants as determined by values such as chemical oxygen demand, biological oxygen demand, and nutrients in the typical concentrations of domestic wastewater affluents. Therefore, this process has been a focus in the treatment of domestic wastewater. However, biokinetic coefficients vary between different types of wastewaters. Table 1 shows the typical biokinetic coefficients values for activated sludge.

Table 1. Typical biokinetic coefficients values for activated sludge

Description		TT *4	Valu	Value	
	Coefficient	Units	Range	Typical	
Maximum specific rate of substrate removal	$k_0$	Day -1	2-8	4	
Endogenous respiration coefficient	ke	Day <sup>-1</sup>	0.03-0.07	0.05	
Saturation coefficient	Km	mg/L (COD- BOD <sub>5</sub> )	40-120	80	
Performance coefficient	Y	mgSSV/mgBOD5	0.3-0.7	40	

Source: Von Sperling and C. A. Chernicharo, 2005 (5), M. & E. Inc et al (6)

The biological process can be a potential tool to treat industrial wastewaters with specific characteristics, but the biokinetic coefficients (kinetic and stoichiometric coefficients) must be determined to optimize the scale applications of the process. The purpose of this study was to use an aerobic semi continuous reactor to determine the biological coefficients to treat wastewater from starch production and to evaluate the efficiencies of this process in the removal of a high concentration of potato starch.

#### 2 MATERIALS AND METHODS

#### 2.1 Experimental setup and operation

The aerobic reactor used in this study was constructed in acrylic with an effective volume of 4.0 L, overall height of 30 cm, and inner diameter of 15 cm. The system had quickclosing half-inch valves; one of these was located at the bottom of the reactor and enabled entry of the wastewater flow, while the other two facilitated the blowdown of the sludge. The incoming flow enterered using a peristaltic pump with a capacity of 6–600 RPM.

The reactor was completely mixed by a mechanical stirrer, and air was supplied by a fish tank pump through a plastic pipe in which stones

were placed to generate fine air bubbles. This facilitated the transfer of oxygen to the microorganisms. The biomass used was of the suspended type. The schematic diagram of the reactor is given in figure 1.





Source: Authors own creation.

The reactor worked in a semicontinuous configuration; therefore, the system's air supply, agitation, and peristaltic pump were activated with a digital timer for the different periods. The reactor worked for a total of 16 active hours and 8 inactive hours every day. The effluent from the system was collected in a sedimentation unit, where it was subsequently returned to the reactor. The purge was carried out directly from the aeration tank according to the age of the sludge from the experimental phase.

#### 2.2 Inoculum

The system was inoculated with an aerobic sludge from a domestic wastewater treatment plant, called Aguas Claras, located in the municipality of Bello, Antioquia. The sludge presented good sedimentation characteristics and a low sludge volumetric index.

#### 2.3 Composition of synthetic wastewater

Synthetic wastewater was prepared to simulate industrial wastewater from the potato starch extraction process. The concentrations of COD used were 500 and 1000 mg / L. The macro and micronutrients were added according to what was reported by Molina (7) for aerobic systems. The pH in the reactor was controlled by adding sodium bicarbonate into the incoming flow.

#### 2.4 Operational Strategy

The reactor was operated for 126 days through 5 phases of operation that consisted of start-up and acclimatization and four subsequent phases which evaluated the variation of the sludge retention time (sludge age). Throughout the study, the kinetic and stoichiometric coefficients were determined, varying the sludge retention time between 20, 10, 8, and 4 days. The biomass concentration varied between 3040 and 610 *.Table 2.* Operational strategy

mg/L and the F/M ratio varied between 0.36 and 1.70 1.92 gCOD / gVSS·d. Table 2.1 presents a summary of the experimental conditions used for the operation of the system

Dhogog	Period (days)	HRT	$\theta_{c}$	COD	
Fliases		(hours)	(days)	(mg/L)	
1	1-66	24	variable	500	
2	67-82	24	20	1000	
3	83-94	24	10	1000	
4	95-107	24	8	1000	
5	108-119	24	4	1000	

Source: Authors own creation.

#### 2.5 Analytical methods

Tests for COD, TSS, VSS, and SVI were done in the laboratory of the research group known as the Pollution Control and Diagnostics Group (GDCON), at the University of Antioquia, following protocols established in the Standard Methods (8). The GDCON laboratory is accredited by the Environmental Research Institute under the Ministry of Environment of Colombia (IDEAM) to carry out these analyses.

## 2.6 Aerobic biokinetic coefficients determination

The interaction between the growth of microorganisms and utilization of the growthlimiting substrate in a completely mixed activated sludge is based on the Monod model (9). This model is the most commonly used one for determining the biokinetic coefficients in biological processes, which can be expressed as (10):

$$\mu = \frac{\mu_{max} \cdot F}{K_m + F}$$

(1)

 $\mu_{max}$  is the maximum specific growth rate and  $K_m$  is the saturation constant, which is numerically equal to the substrate concentration when  $\mu = 0.5\mu_{max}$ . The coefficients were determined based on the following assumptions:

i. The reactor volume is constant and completely mixed.

Eq.

- ii. Influent COD concentration remains constant.
- iii. The synthetic influent wastewater does not contain microbial solids.
- iv. Steady-state conditions prevail throughout the system during the experimentation time.

According to figure 1, the mass balance equations of both the biomass and substrate

removal can be described by the following equations (11):

#### **3 RESULTS AND DISCUSSION**

The aerobic reactor used in the experimentation was operated under mesophilic conditions. The pH parameter (Table 2) was monitored every day to keep good environmental conditions for the microorganisms. The dissolved oxygen was always over 2 mg/L.





Source: Authors own creation.

Potato starch could be a good carbon source when used as a substrate for biomass. However, the low water solubility and high potato starch concentrations in industrial wastewater may be a limiting factor for the aerobic process. Table 2 shows the pH variation through the experimentation time. The values show a tendency to remain between 7 and 8. The pH was controlled by adding 0.5 g of sodium bicarbonate. According to the literature, the pH in this biological process can vary while remaining close to neutrality. However, the pH in wastewater can fluctuate in the range of 6.5-8.5 (16), which means that the environmental medium was suitable for the metabolic development of most microorganisms

Table 3. Reactor performance under start-up and steady-state conditions at four different  $\Theta c$ .

Doromotor	_				
r ar anieter	Start up	20	10	8	4
Operation period (days)	01-66	67-82	83-94	95-107	108-119
F/M ratio (mg CODmg/VSS day)		0.36	0.48	<u>1.11</u>	<u>1.7</u>

pH	$7.3\pm0,\!3$	$7.4\pm0.3$	$7.7\pm0.3$	$7.5\pm0.2$	$7.5\pm0.3$
Aflluent COD (mg/L)	$478\pm52$	$1081\pm71$	$1074\pm85$	$1108 \pm 115$	$1038\pm74$
Eflluent COD (mg/L)	$69\pm20$	$22 \pm 4$	$33\pm5$	$90\pm 8$	$148\pm11$
COD removal efficiencies (%)	$86\pm4$	$95\pm~1$	94 ± 1	$94\pm2$	92 ± 2

Source: Authors own creation.

The sludge retention time was used as a controlling parameter throughout the experimentation. The reactor was operated applying different  $\Theta c$  (Table 1). Therefore, the biomass concentration (mixed liquor suspended solids -MLSS) was changed for phases from 2 to 5 with average values of 3040, 2240, 1400 610 mgSS/L respectively. The biokinetic coefficients were determined at steady-state, a condition that

was assumed to exist when biomass growth and COD removal were constant.

The biokinetic coefficient values were estimated through regression analysis of experimental data using the Lineweaver-Burk method as shown in figure 3. The values obtained for  $k_0$  and  $k_m$ were 1.8 1/day and 93 mg/L COD respectively. The graphic analysis of data shows a high regression coefficient with an R<sup>2</sup> of 0.9596.

Figure 3. Lineweaver-Burk method for COD removal





In the same way, the Y and  $k_e$  coefficients were estimated through the graphical analysis of experimental data, as shown in figure 4. The Y and  $k_e$  values obtained were 0.4 mgSSV/mgCOD and 0.09 1/day respectively. In this case, the graphic analysis of data shows a good fit with a regression coefficient where the  $R^2$  was 0.9998.

#### *Figure* 4. Determination of Y and $k_e$



Source: Authors own creation.

It is important to mention that the biokinetics coefficient values obtained for starch removal are similar to those for domestic wastewater reported in table 1. The constant  $k_m$  represents the affinity of the biomass for the substrate. In this case, the high values of this constant mean a high affinity of the microorganisms for the starch. This could be because starch is a highly biodegradable substrate, and the reactor had a good performance at the COD concentration of 1000 mg/L. However, the reactor was operated at a high HRT (24 hours), which means that if the COD load changes the starch removal efficiency could change as well (17). A study on starch wastewater treatment reported by Wang and collaborators (1) showed that aerobic system

efficiency is affected by HRT. They proved that at high HRT, such as 24 hours, the reactor had efficiencies up to 96.5% but when the HRT was low (HRT = 6 hours) the efficiencies decreased considerably.

Figure 5 shows the behavior of the F/M ratio vs Oc. The F/M ratio decreases when Oc is higher and vice versa. Therefore, this graph describes the way the F/M ratio changes with the Oc variation through the experimentation time. It is important to note that the reactor had a strong variation in biomass concentration (mixed liquor suspended solids) under the effect of Oc decrease and, as a consequence, the ratio of F/M rose considerably during the experimentation time, as can be seen in figure 5.



*Figure 5.* Graphic representation of a) the behavior of F/M vs the variation in the  $\Theta c$ . b) variation of system efficiency vs the variation in the  $\Theta c$ 

Source: Authors own creation.

On the other hand, the efficiency of the system is strongly affected by the F/M ratio. When the F/M ratio is lower the efficiency of the system increases. This good efficiency in the system occurs because, when the ratio F/M is lower, the biomass is in high competition for the substrate, which may not be enough for the microorganism. Therefore, there is higher efficiency in the system. However, it is important to note that the system dynamics can change if the cell residence time is too high. At a

low F/M ratio the presence of nematodes and rotifers predominates, but if the cell-residence time continues to increase, the biomass decreases to the point that is not possible to find active cells through microscopic observation (figure 6 c). In this case, system efficiency decreases dramatically. Therefore, when determining biokinetic coefficients, the system never reached this critical condition. The reactor was operated under a steady-state with active biomass.

Figure 6. Change in the microbial structure under the effect of ration F/M variation



(Source: own elaboration).

However, when the F/M ratio increases, the system efficiency decreases (figure 5b). This drop-in efficiency is associated with low concentration of the biomass, despite its greater biological activity. In this case, there is a change in the sludge microbial structure whereby dispersed bacterial populations, free ciliates, and flagellates predominate (figure 6 a-c).

These changes in the biological structure also affect the system's operation. When the F/M

ratio is lower, the SVI is good, and the sludge has good sedimentation characteristics with SVI values below 100 mL/g (7). However, when the F/M ratio is higher the SVI is also higher (>100 mL/g). Therefore, the sludge does not have good sedimentation characteristics, which is a consequence of the presence of dispersing biomass.







The oxygen consumption rate was also monitored through the experimentation phases under the  $\Theta c$  variation. Figure 7 shows the values for the OCR for the experimental test in each  $\Theta c$  and the theoretical calculation.

### 4. CONCLUSIONS AND RECOMMENDATIONS

The aerobic process can be used as an alternative to treat wastewater from starch extraction. The biokinetics coefficients showed good values for starch removal, which are close to the coefficient values of domestic wastewater treatment. This similarity arises because starch is an organic substrate with high biodegradability. However, it is important to consider the effect of the organic load when the aerobic system is going to be applied as the only process to treat this kind of wastewater. This is due to the fact that the starch wastewater has a high COD concentration; therefore, a high HRT is needed to obtain good efficiency, as was the case in this study.

Consequently, it is suggested that future research should evaluate the maximum starch load which

can be applied to the system without losing efficiency, and also, to evaluate whether there is a change in the biokinetics values when this load increases. However, the results obtained in this research are a good basis for the application of biological processes to treat wastewater from starch production.

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