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## THE DUMPING OF OSCILLATORY PHENOMENA IN THE PROCESS OF BIOETHANOL PRODUCTION BY CONTINUOUS FERMENTATION

### TŁUMIENIE ZJAWISK OSCYLACYJNYCH W PROCESIE PRODUKCJI BIOETANOLU METODĄ FERMENTACJI CIĄGŁEJ

#### Abstract

This article presents the results of numerical research on the dumping of oscillatory phenomena occurring in the continuous bioethanol production process. Proportional and proportional-integral types of controllers were tested for this purpose. Numerical analysis showed that the appropriate selection of the  $K_c$  value makes it possible to suppress the oscillations in the system. The introduction of the integral term improves the performance of control system. Using numerical calculations, it was shown that the PI controller is effective at dumping the occurring oscillations. The presence of the integral term allows the reduction of the gain coefficient value. After the proper selection of parameters, the PI controller effectively suppresses the oscillations present in the system.

**Keywords:** bioethanol production, *Saccharomyces cerevisiae*, oscillatory behaviour, process control, PI controller

#### Streszczenie

W niniejszym artykule zaprezentowano wyniki badań numerycznych, dotyczących tłumienia zjawisk oscylacyjnych występujących w procesie produkcji bioetanolu metodą ciągłą. Przebadano w tym celu regulator typu proporcjonalnego oraz proporcjonalno-calkującego. Analiza numeryczna wykazała, iż odpowiedni dobór wartości współczynnika wzmocnienia  $K_c$  umożliwia tłumienie zjawisk oscylacyjnych w układzie. Wykazano także, iż wprowadzenie członu calkującego poprawia jego działanie. Za pomocą obliczeń symulacyjnych wykazano, że regulator proporcjonalno-calkujący dobrze radzi sobie z tłumieniem występujących oscylacji. Obecność członu calkującego pozwala na zredukowanie wartości współczynnika wzmocnienia. Po odpowiednim doborze parametrów, regulator PI skutecznie tłumie obecne w układzie oscylacje.

**Słowa kluczowe:** produkcja bioetanolu, *Saccharomyces cerevisiae*, zachowania oscylacyjne, sterowanie procesem, regulator PI

## Nomenclature

$C$	– intracellular storage carbohydrate concentration (g/g biomass)
$D$	– dilution rate ( $\text{h}^{-1}$ )
$E$	– ethanol concentration ( $\text{g} \cdot \text{l}^{-1}$ )
$G$	– glucose concentration ( $\text{g} \cdot \text{l}^{-1}$ )
$G_0$	– feed glucose concentration ( $\text{g} \cdot \text{l}^{-1}$ )
$O$	– dissolved oxygen concentration ( $\text{mg} \cdot \text{l}^{-1}$ )
$O^*$	– dissolved oxygen solubility limit ( $\text{mg} \cdot \text{l}^{-1}$ )
$K_i$	– $i$ -th pathway Michealis constant ( $\text{g} \cdot \text{l}^{-1}$ )
$K_o$	– oxidative pathway oxygen saturation constant ( $\text{mg} \cdot \text{l}^{-1}$ )
$X$	– biomass concentration ( $\text{g} \cdot \text{l}^{-1}$ )
$Y_i$	– $i$ -th pathway yield coefficient ( $\text{g biomass} \cdot \text{g}^{-1}$ substrate)
$e_i$	– $i$ -th pathway intracellular enzyme concentration ( $\text{g} \cdot \text{g}^{-1}$ biomass)
$k_L a$	– oxygen mass transfer coefficient ( $\text{h}^{-1}$ )
$r_i$	– $i$ -th pathway growth rate ( $\text{h}^{-1}$ )
$r_{i,\text{max}}$	– maximum growth rate of all pathways at any instant ( $\text{h}^{-1}$ )
$u_i$	– $i$ -th pathway cybernetic variable controlling enzyme synthesis
$v_i$	– $i$ -th pathway cybernetic variable controlling enzyme activity
$\alpha$	– specific enzymatic synthesis rate ( $\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ )
$\alpha^*$	– parameter for constitutive enzyme synthesis ( $\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ )
$\beta$	– specific enzymatic degradation rate of intracellular enzymes ( $\text{g} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ )
$\gamma_i$	– stoichiometric parameters ( $\text{g} \cdot \text{g}^{-1}$ )
$\varphi_i$	– stoichiometric parameters ( $\text{g} \cdot \text{g}^{-1}$ )
$\mu_i$	– $i$ -th pathway modified specific growth rate constant ( $\text{h}^{-1}$ )
$\mu_{i,\text{max}}$	– $i$ -th pathway maximum specific growth rate constant ( $\text{h}^{-1}$ )
$S$	– controller output signal
$V$	– volume of bioreactor ( $\text{m}^3$ )
$F_{vf}$	– volumetric flow rate of feed stream ( $\text{m}^3 \cdot \text{h}^{-1}$ )
$K_p$	– gain coefficient of controller
$T_i$	– time of integration
$e(t)$	– control error

## On diagrams

LP	– limit point
HB	– Hopf bifurcation point

## 1. Introduction

*Saccharomyces cerevisiae* is a very important microorganism in many branches of industry and science [1]. One of its uses is in the production of bioethanol. Bioethanol is currently the most widely used biofuel in the world [2]. As a result, the improvement of its production process remains a very important challenge. A characteristic feature of continuous baker's yeast cultures is the occurrence of extracellular and intracellular parameter oscillations [3-9]. These parameters include among others the concentration of glucose, the concentration of ethanol, the pH value of the process environment, the concentration of biomass and the concentration of stored intracellular carbohydrates. In practice, the dumping of these oscillations is still a challenge which industrial companies do not always deal with [10]. For this reason, the possibility to suppress occurring oscillations is considered in this publication.

## 2. Mathematical model

The analysis involved the production of bioethanol under aerobic conditions, carried out in a continuous stirred tank bioreactor. The bioreactor is equipped with an automatic control system coupled with a valve regulating the volumetric flow rate of the feed stream. The process system is shown in Fig. 1.

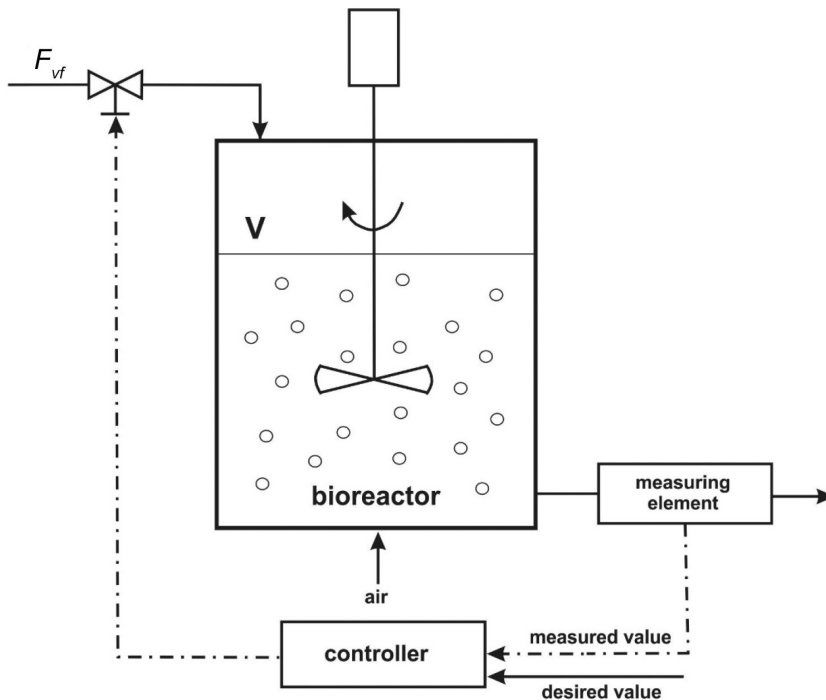


Fig. 1. Scheme of the process system with automatic bioreactor controller

## 2.1. Model of continuous stirred tank bioreactor

This biochemical process has been described by a structured and nonsegregated model of yeast growth dynamics [11]. The proposed model considers the occurrence of three biochemical reactions which are enzymatically controlled. These include the reaction of glucose fermentation ( $R_1$ ), the reaction of ethanol oxidation ( $R_2$ ) and the reaction of glucose oxidation ( $R_3$ ). Biomass growth rates according to a given biochemical reaction are modelled by equations (1-3).

$$r_1 = \mu_1 e_1 \frac{G}{K_1 + G} \quad (1)$$

$$r_2 = \mu_2 e_2 \frac{E}{K_2 + E} \cdot \frac{O}{K_{O_2} + O} \quad (2)$$

$$r_3 = \mu_3 e_3 \frac{G}{K_3 + G} \cdot \frac{O}{K_{O_3} + O} \quad (3)$$

The specific growth rate in equations (1-3) is expressed by relationship (4).

$$\mu_i = \mu_{i\max} \frac{\mu_{i\max} + \beta}{\alpha + \alpha^*} \quad (4)$$

Metabolic pathways are regulated by cybernetic variables  $u_i$  and  $v_i$  (equations 5 and 6), which control the synthesis and activity of key enzymes. They are responsible for the dynamic competition between the three metabolic pathways, which is the characteristic feature of this model and it is confirmed by experimental research [11].

$$u_i = \frac{r_i}{\sum_j r_j} \quad (5)$$

$$v_i = \frac{r_i}{\max_j r_j} \quad (6)$$

The mathematical model of the process system includes eight state variables which can be classified into two different groups. The first group is comprised of extracellular process variables, such as glucose concentration ( $G$ ), ethanol concentration ( $E$ ), biomass concentration ( $X$ ) and oxygen concentration in the liquid phase ( $O$ ). The second group is formed of intracellular variables in the form of the concentration of stored carbohydrates ( $C$ ), the concentration of enzymes catalysing the glucose fermentation reaction ( $e_1$ ), the concentration of enzymes catalysing the ethanol oxidation reaction ( $e_2$ ) and the concentration of enzymes catalysing the glucose oxidation reaction ( $e_3$ ). Therefore, the aerobic continuous culture of microorganisms in a continuous tank bioreactor with perfect mixing (Fig. 1) can be described by the system of eight ordinary differential equations (7-14). Table 1 presents parameter values for the above-described model.

$$\frac{dG}{dt} = D \cdot (G_o - G) - \left( \frac{r_1 v_1}{Y_1} + \frac{r_3 v_3}{Y_3} \right) \cdot X - \varphi_4 \left( C \frac{dX}{dt} + \frac{dC}{dt} \right) \quad (7)$$

$$\frac{dX}{dt} = -D \cdot X + X \cdot \sum_i (r_i v_i) \quad (8)$$

$$\frac{dE}{dt} = -D \cdot E + \left( \varphi_1 \frac{r_1 v_1}{Y_1} - \frac{r_2 v_2}{Y_2} \right) \cdot X \quad (9)$$

$$\frac{dO}{dt} = -D \cdot O + k_L a \cdot (O^* - O) - \left( \varphi_2 \frac{r_2 v_2}{Y_2} + \varphi_3 \frac{r_3 v_3}{Y_3} \right) \cdot X \quad (10)$$

$$\frac{dC}{dt} = \gamma_3 r_3 v_3 - (\gamma_1 r_1 v_1 + \gamma_2 r_2 v_2) C - \sum_i (r_i v_i) C \quad (11)$$

$$\frac{de_1}{dt} = \alpha u_1 \frac{G}{K_1 + G} - \left( \sum_i (r_i v_i) + \beta \right) e_1 + \alpha^* \quad (12)$$

$$\frac{de_2}{dt} = \alpha u_2 \frac{G}{K_2 + E} - \left( \sum_i (r_i v_i) + \beta \right) e_2 + \alpha^* \quad (13)$$

$$\frac{de_3}{dt} = \alpha u_3 \frac{G}{K_3 + G} - \left( \sum_i (r_i v_i) + \beta \right) e_3 + \alpha^* \quad (14)$$

Table 1. Values of kinetic parameters of the cybernetic model [12]

Parameter	Unit	Value	Parameter	Unit	Value
$\mu_{1\max}$	$h^{-1}$	0.44	$K_1$	$g \cdot l^{-1}$	0.05
$\mu_{2\max}$	$h^{-1}$	0.19	$K_2$	$g \cdot l^{-1}$	0.01
$\mu_{3\max}$	$h^{-1}$	0.36	$K_3$	$g \cdot l^{-1}$	0.001
$Y_1$	$g \cdot g^{-1}$	0.16	$\varphi_1$	$g \cdot g^{-1}$	0.403
$Y_2$	$g \cdot g^{-1}$	0.75	$\varphi_2$	$g \cdot g^{-1}$	2.087
$Y_3$	$g \cdot g^{-1}$	0.60	$\varphi_3$	$g \cdot g^{-1}$	1.067
$k_L a$	$h^{-1}$	225	$\varphi_4$	$g \cdot g^{-1}$	0.95
$K_{O_2}$	$mg \cdot l^{-1}$	0.01	$\alpha$	$g \cdot g^{-1} \cdot h^{-1}$	0.3
$K_{O_3}$	$mg \cdot l^{-1}$	2.2	$\beta$	$g \cdot g^{-1} \cdot h^{-1}$	0.7
$\gamma_1$	$g \cdot g^{-1}$	10.0	$\alpha^*$	$g \cdot g^{-1} \cdot h^{-1}$	0.03
$\gamma_2$	$g \cdot g^{-1}$	10.0	$O^*$	$mg \cdot l^{-1}$	7.5
$\gamma_3$	$g \cdot g^{-1}$	0.8			

## 2.2. Process Control

Biochemical processes are characterised by high response time delays of steady state disturbance. This is caused by running cell processes which are enzymatically catalysed [1]. When a disturbance occurs in the system, the reaction of the biomass cells must precede the intracellular synthesis of the relevant enzymes. Thus, the responses appearing in biological systems are slow. Therefore, controlling such a process requires the selection of the appropriate controller.

The basic automatic controller used in industry is a proportional controller (P). The dependence of the output signal on the control error is described by equation (15).

$$S(t) = S(0) + K_p e(t) \quad (15)$$

This type of controller is used to control objects with small time constants and small delay times. In addition, the object should exhibit slow dynamics of occurring disturbances. A characteristic feature of this controller is the presence of a static error. This means that it is not possible to adjust the object to set the value with the P controller.

In the analysed case, the volumetric flow rate  $F_{vf}$  of the bioreactor feed stream was taken as the control variable. The dependence of the flow rate on time being the output signal of the controller is given by expression (16).

$$F_{vf}(t) = F_{vf}(0) + K_p e(t) \quad (16)$$

After taking into account equation (16), the expression for the dilution rate, present in the balance equations of the bioreactor (7-14), takes the form of equation (17).

$$D = \frac{F_{vf}(0) + K_p e(t)}{V} \quad (17)$$

The proportional-integral controller (PI) is another commonly used controller in industry. The output signal is proportional to both the control error and the integral of the control error, as shown in equation (18).

$$S(t) = S(0) + K_p \left( e(t) + \frac{1}{T_i} \int_0^t e(t) dt \right) \quad (18)$$

PI controllers are used to control objects of any time constants and high time delays. It is possible to control an object in which there are significant but slowly changing disturbances. It is worth noting that its use allows complete removal of the static regulation error [13].

In the analysed case, the dependence of the volumetric flow rate  $F_{vf}$  from time, which is the output signal of the regulator, is shown in equation (19).

$$F_{vf}(t) = F_{vf}(0) + K_p \left( e(t) + \frac{1}{T_i} \int_0^t e(t) dt \right) \quad (19)$$

After considering equation (19), the expression for the dilution rate, present in the balance equations of the bioreactor (7-14), takes the form of equation (20).

$$D = \frac{F_{vf}(0) + K_p \left( e(t) + \frac{1}{T_i} \int_0^t e(t) dt \right)}{V} \quad (20)$$

The glucose concentration was taken as the control variable ( $G$ ). Therefore, appearing in the above presented equations control error was defined by relationship (21).

$$e(t) = G_{SET} - G(t) \quad (21)$$

where:

- $G_{SET}$  – the set value of glucose concentration in system,
- $G(t)$  – the current value.

### 3. Numerical techniques and calculations

The analysed system of ordinary differential equations (equations 7-14) does not have an analytical solution. In order to obtain a solution, it is necessary to perform numerical calculations. The first part of the calculations aimed at steady state analysis and the identification of the areas of oscillatory solutions was performed in the XPPAUT 8 program [14]. This program is integrated with the commonly used AUTO package for bifurcation analysis [15]. Based on the results of the bifurcation analysis, three sets of process parameters were selected; these are characterised by a different oscillation amplitudes. The second part of the calculations was performed using the Gear's method. This method is recommended for the numerical integration of stiff systems of ordinary differential equations [16]. Numerical studies included the determination of time trajectories and the simulation of proportional and proportional-integral controller operations on the process system in which oscillations occur.



## 4. Results and discussion

Section 4.1 presents example results of the bifurcation analysis and the process parameters for three selected limit cycles. In point 4.2, the process control area with a description of the automatic controller influence on the position of the control valve is defined. In sections 4.3 and 4.4, the results of numerical simulations showing the workings of the P and PI controllers and their influence on the stabilisation of the process system are presented.

### 4.1. Bifurcation analysis of steady states

An example of a bifurcation diagram obtained for the value of dilution rate  $D = 0.1000 \text{ [h}^{-1}\text{]}$ , is shown in Figure 2. The continuous line signified stable steady states and the unstable steady states are marked with a dashed line. As the bifurcation parameter, the concentration of glucose in the feed stream  $G_0$  was used. The area of oscillatory solutions is determined by the Hopf bifurcation points  $HB_1$  and  $HB_2$ . Both points are subcritical; this means that the limit cycles generated in their environment are unstable. Between points  $HB_1$  and  $LP_1$ , unstable cycles are generated. In point  $LP_1$ , the nature of the oscillatory solutions changes and between the  $LP_1$  and  $LP_2$ , stable cycles are generated. In  $LP_2$ , the system loses its stability again, which recovers at  $LP_3$ . Then, between the points  $LP_3$  and  $LP_4$ , stable limit cycles are again generated. However, in point  $LP_4$ , the system loses stability once more.

In Fig. 2, three selected values of process parameters have been marked with a vertical dashed line. These are analysed in the following sections. The generated oscillatory time series for selected values of process parameters are shown in Figure 3. Their course indicates the high dynamics of the analysed process.

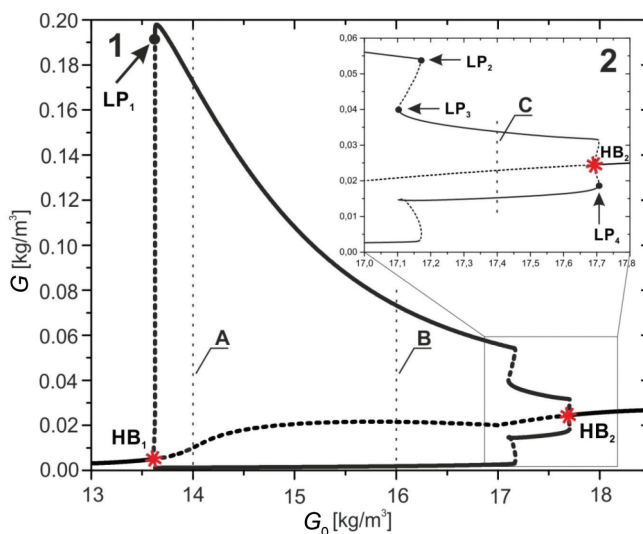


Fig. 2. Bifurcation diagram for value of dilution rate  $D = 0.1000 \text{ [h}^{-1}\text{]}$  relative to feed stream glucose concentration  $G_0$ ; A -  $G_0 = 14.0 \text{ [kg/m}^3\text{]}$ , B -  $G_0 = 16.0 \text{ [kg/m}^3\text{]}$ , C -  $G_0 = 17.4 \text{ [kg/m}^3\text{]}$



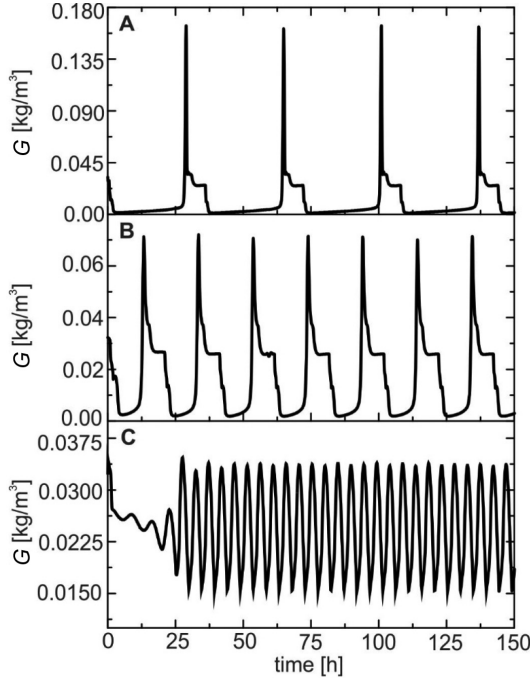


Fig. 3. Time trajectories of glucose concentrations ( $G$ ); A –  $G_0 = 14.0$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]; B –  $G_0 = 16.0$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]; C –  $G_0 = 17.4$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]

#### 4.2. Definition of process control area

The control of the process system is accomplished via a valve, which regulates the value of feed stream flow. It is therefore necessary to define the area of flow rates that it can regulate. For this purpose, the value of the critical dilution rate  $D_{kr}$  was determined. This value is the process limit, beyond which the biomass cells are washed out from the system. This situation is illustrated by the bifurcation diagram shown in Fig. 4. The diagram was made for three predefined sets of process parameter values. On the basis of this, the approximate value of  $D_{kr} = 0.4$  [h<sup>-1</sup>] was determined.

Having the  $D_{kr}$  value and the assumed volume of the bioreactor  $V = 10$  [m<sup>3</sup>], it is possible to estimate the maximum stream feed flow through the bioreactor below which the process may run. In the analysed case, it is  $F_{vfmax} = 4.0$  [m<sup>3</sup>/h]. This means that the control valve must regulate the flow rate in the range from 0 (in the closed position) to  $F_{vfmax}$  (in the fully open position). Next, using this value and the assumed diameter of the bioreactor feeding pipe  $d = 0.1$  [m], the maximum flow rate of the stream feed through this pipe was estimated to

be  $U_{max} = \frac{4F_{vfmax}}{3600\pi d^2} = 0,142$  [m/s]. This is a rational value that can occur in a real industrial

process.

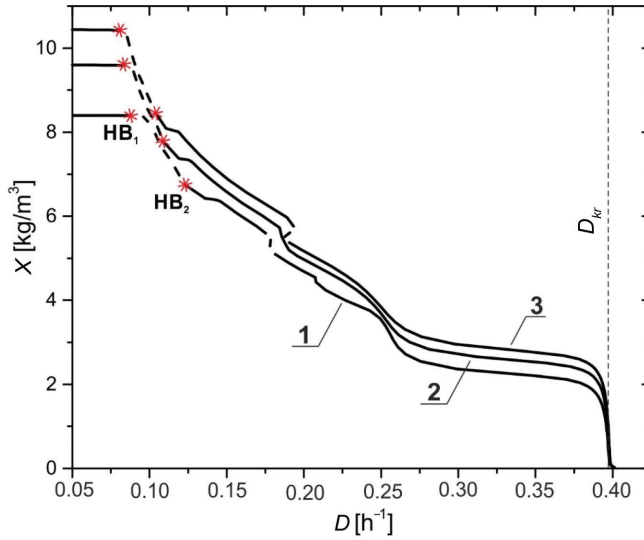


Fig. 4. Bifurcation diagram for  $G_0 = 14.0$  [kg/m<sup>3</sup>] (1),  $G_0 = 16.0$  [kg/m<sup>3</sup>] (2),  $G_0 = 17.4$  [kg/m<sup>3</sup>] (3), relative to dilution rate  $D$ , made to determine the value of critical dilution rate  $D_{cr}$

### 4.3. Evaluation of the possibility of controlling the process system using the P controller

The possibility of controlling the oscillatory behaviours using a proportional controller was investigated. For this purpose, numerical simulations were performed for all three oscillatory trajectories, assuming three different values of the gain coefficient  $K_{c1} = 25.0$ ,  $K_{c2} = 34.0$  and  $K_{c3} = 45.0$ . In the simulations, the glucose concentration in the system was set at  $G_{SET} = 0.03$  [kg/m<sup>3</sup>]; this is marked in Fig. 5 with horizontal dashed blue lines.

The conducted research has shown that for the assumed value  $K_{c1} = 25.0$ , the controller is able to suppress oscillations for stationary states B and C, as shown in Fig. 5.A2 and 5.A3. It is not able to control the steady state A, which is shown in Fig. 5.A1. In this case, the controller suppresses the occurring oscillations, the amplitude of which decreases but does not disappear. In the case of the other two values of the gain factor  $K_{c2} = 34.0$  and  $K_{c3} = 45.0$ , the controller suppresses the occurring oscillations for all three stationary states (diagrams 5.B1-3 and 5.C1-3). Furthermore, the numerical investigations showed that the setting  $K_{c2} = 34.0$  is the lowest value of the controller gain coefficient, for which the controller suppresses oscillations occurring in the system with different amplitudes for steady states A, B and C. Unfortunately, as mentioned before, this type of controller is not able to compensate the regulation error completely. As a consequence, its value stabilises at a certain level, so that the system does not reach the set point  $G_{set}$ .

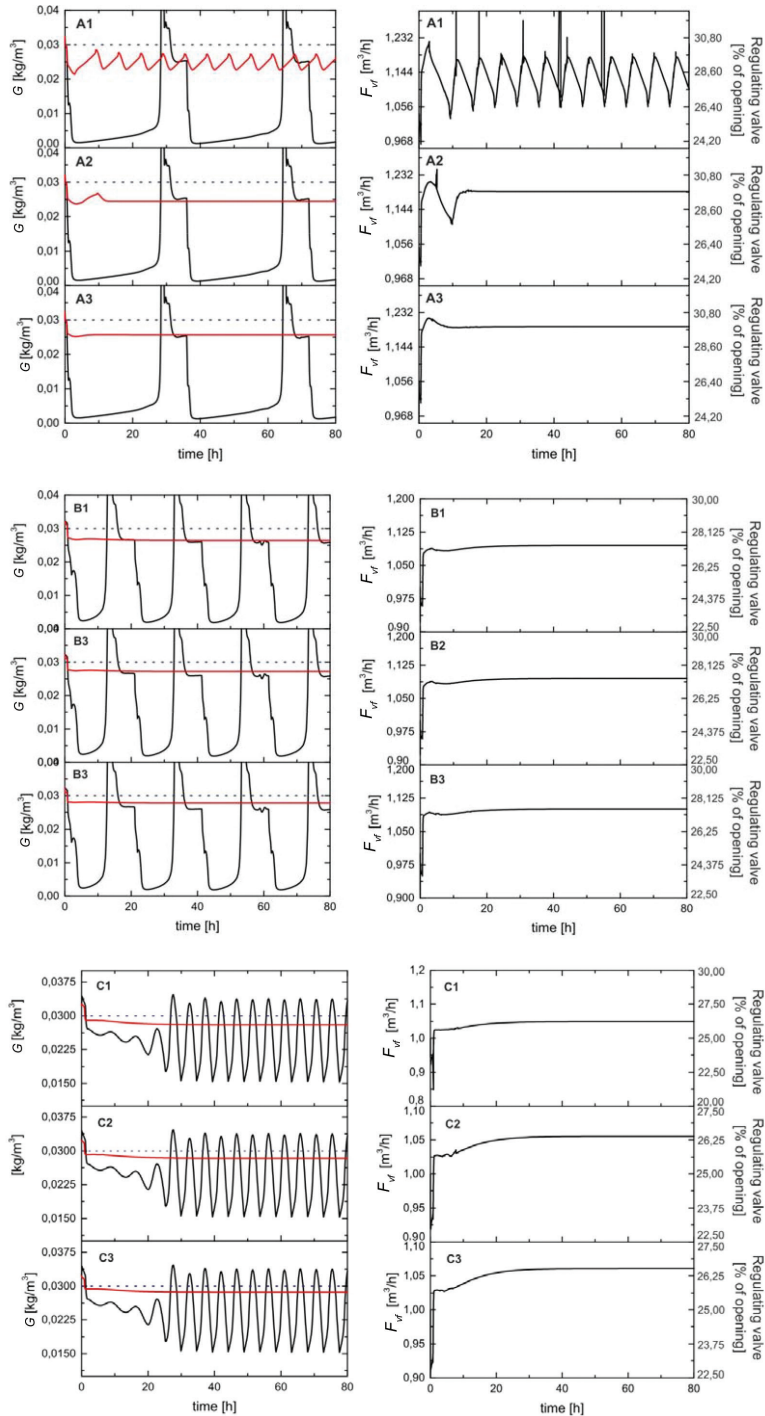


Fig. 5. Dumping of oscillations for the value of gain factor  $K_{c1} = 25.0$ , and  $K_{c2} = 45.0$ ; A -  $G_0 = 14.0$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]; B -  $G_0 = 16.0$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]; C -  $G_0 = 17.4$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]



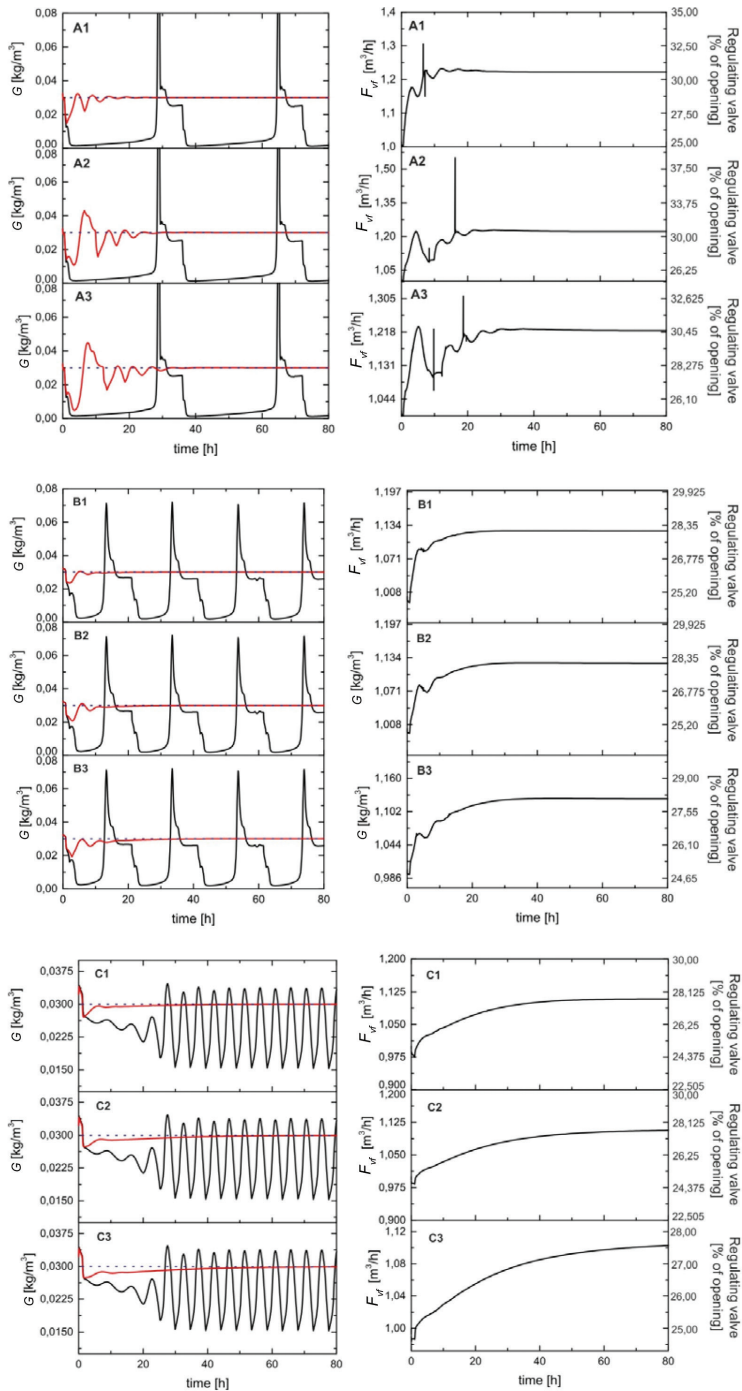


Fig. 6. Dumping of oscillations for the value of gain factor  $K_c = 3.0$  and time of integration  $T_{i1} = 0.5$ ,  $T_{i2} = 1.0$ ,  $T_{i3} = 1.5$  A -  $G_0 = 14.0$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]; B -  $G_0 = 16.0$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]; C -  $G_0 = 17.4$  [kg/m<sup>3</sup>],  $D = 0.1000$  [h<sup>-1</sup>]

#### 4.4. Evaluation of the possibility of controlling the process system using the PI controller

Due to the occurrence of a static error in the case of the P controller, as a result of which the system was not able to reach the settled value of  $G_{\text{SET}}$ , it was decided to investigate the proportional-integral controller (PI). Similarly to the previous case, a series of PI controller simulations were performed for all three selected oscillatory steady states. On the basis of these, the values of the gain coefficient  $K_c = 3.0$  and time of integration  $T_{i1} = 0.5$ ,  $T_{i2} = 1.0$ ,  $T_{i3} = 1.5$  were proposed. Also in this case, a set point of glucose concentration in the system was assumed to  $G_{\text{SET}} = 0.03$  [kg/m<sup>3</sup>]; this is marked on the diagrams in Fig. 6 with horizontal dashed blue lines.

The addition of an integral term resulted in a significant reduction of the gain coefficient value  $K_c$ . For the analysed set of gain coefficients and integration times, the controller suppressed oscillations each time and reached the set point  $G_{\text{SET}}$  (Fig. 6.A1-3, 6.B1-3 and 6.C1-3). Thus, the introduction of the integral term eliminated the occurrence of a static error. The performed research also showed that increasing the value of the integration time  $T_i$  has a negative effect on the suppression of oscillations in the system. This is manifested by the extension of the time required to reach the stable steady state.

### 5. Conclusions

The numerical tests aimed at estimating the possibility of the suppression of oscillations occurring during the production of bioethanol using a continuous method were performed. Proportional and proportional-integral types of controller were tested for this purpose. Numerical analysis showed that by appropriate selection of gain coefficient value  $K_c$ , it is possible to regulate the oscillatory phenomena in the system. Unfortunately, this type of controller is accompanied by the occurrence of a static control error. This fact may disqualify this type of regulation for industrial use. The introduction of the integral term into the control system significantly improves its operation. As a result, the proportional-integral controller is effective at dumping the occurring oscillations. In addition, a system with this type of regulation is able to compensate the regulation error completely. The presence of the integral element results in a significant reduction of the gain coefficient value. After proper selection of settings, the PI controller is able to effectively suppress the oscillating phenomena occurring in the continuous process of bioethanol production.

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