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## Concept of food-chain control in the bioreactor fed with a mixture of substrates

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### ABSTRACT

Simulation of biological processes is a complex issue. This is especially true for such biological processes as the aquaculture food chain systems in the natural environment. The quality of the final product (e.g., fish and shellfish) largely depends on the conditions in the first level of the food chain. In the modern biotechnology, the biological processes are conducted in the automated continuous flow bioreactors. This is also possible for aquaculture systems, namely, the substrate-bacteria-protozoa process, which is taken under consideration in this paper. The main contribution of the authors is the idea of using a mixture of two substrates that have different properties and are fed into the continuous flow bioreactor in order to control process course and, especially, to control the oscillatory behaviour of the food chain system. The ratio of the inlet flow rates of the two substrates is proposed as a new control variable. The analysis of the system behaviour was performed based on the simulation experiments with the use of the modified mathematical model of the food chain system. The model is an extended version of the classical models defined for the single limiting substrates. The simulation results presets the possibility of using the new control variable for control of the oscillatory behaviour of the system.

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### 1. Introduction

Currently, the changes in the aquaculture environment are associated with growing influence of industrialization on the processes that take place in the food chain. In other words, the hardly controllable processes from natural environment “are transformed” to the industrial processes that are more efficient and can be easily controllable and supervised. In the case of fish or shellfish farms, the quality of the final product (e.g., fish and shellfish) largely depends on conditions in the first level of the food chain. On the other hand, the first level can be realized in a relatively simple and efficient way as a continuous biotechnological process in the bioreactor. It is expected that such processes will dominate in the industrial biotechnology in the near future (Croughan et al., 2015).

The typical aquaculture food chain is as follows: soluble nutrient (substrate) – bacteria – protozoa. The final link in the food chain, i.e., the protozoa, can be a good source of food for fish in the fish farming. In general approach, the bacteria are preys and the protozoa are predators. In the case of non-limiting substrates (e.g., in excess of substrates), there is the classical predator-prey relationship between bacteria and protozoa.

Mathematical description of the predator-prey relationship were first presented in paper by Lotka (1924) and then in paper by Volterra

(1926). The mathematical model describes the relationships between predators and preys with the assumption that the growth of the prey population is not limited in the absence of predators and its decrease results from the encounter frequency between the predators and preys. In the following years, the original model was successfully used in the analysis of various dynamical systems.

However, due to the limitations of the classical Lotka-Volterra model, its structure was constantly extended and modified. For instance, in (Bungay and Bungay, 1968) the authors presented the predator-prey model with input and output flow rates of bacteria and protozoa without considering their mortality rate. A particularly important contribution was presented in (Monod, 1942), where the growth of microorganisms was assumed to be limited on the concentration of available substrate. The function describing the limited growth of microorganisms is known in the literature as the Monod relation. By introducing a similar function, Proper and Garver (1966) presented a model where the growth of protozoa (predators) was limited by the concentration of bacteria (preys). This resulted in a new mathematical model of the food-chain for protozoa and bacteria with the growth limited by the substrate, which has been described in detail by Drake et al. (1966); Bungay and Bungay (1968), and Canale (1968, 1969, 1970). The results presented in this paper are based on the food-chain model with a single limiting substrate considered in (Canale, 1970), which has been extended to the case of two limiting substrates. It should be emphasized that the general model for the substrate-bacteria- protozoa system proposed by Canale and can also be used in the analysis of activated sludge systems (Canale, 1969). In turn, the identified

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parameter values for food-chain models can be found in papers presenting the specific problems for various species (see, e.g., Dominguez et al., 2005; Figueiredo and Narciso, 2006; Souto et al., 2008).

Based on the set of mathematical models for the substrate-bacteria-protoczoa systems, it is possible to analyze the dynamical behaviour of the food-chain system for various environmental conditions and for various inputs to the system. Most of the papers discuss the predator-prey and food-chain systems that exhibit the self-sustained oscillations (the oscillatory behaviour). For example, a three trophic level food chain model with ratio dependent functional response was studied by Hsu et al. (2003) for application to biological control. The role of alternative food as disease controller in a disease induced predator-prey system was studied in (Sahoo and Poria, 2013). The studies were continued by the same authors in (Sahoo and Poria, 2014) by analyzing the dynamical properties of the predator-prey system at the presence of additional food source for the predators. The bifurcation analysis (including oscillatory and chaotic behaviour) for the food-chain in chemostat systems with constant and pulse feeding of substrate were analyzed in the series of papers (Wang et al., 2007; Pang et al., 2008; Pang and Liang, 2008; Wang et al., 2008). The problem of modelling the role of constant and time varying recycling delay in nutrient-autotroph-herbivore system was presented in (Mukhopadhyay and Bhattacharyya, 2010). The same authors presented investigations (Bhattacharyya and Mukhopadhyay, 2010) on the role and influence of different system delays on the ecological food chain dynamical properties, including modelling and analysis of oscillations. Similar results have been presented by Cai et al. (2012).

The main goal of this paper is to discuss the idea of using a mixture of two growth limiting substrates as the input to the food chain system. Although, the problem of using a mixture of two different substrates for the chemostat system is known in the literature (see, e.g., Parulekar et al., 1986; Egli, 1995; Martegani et al., 1990; Sridhar, 2011), the proposition of using the ratio of input flow rates of both substrates as a new control variable is a novel approach. Hence, the presented extension of

the food chain model to the case of two limiting substrates is also novel. It should also be noted that one of the goals is to study the possibilities of using the mixture of substrate to stimulate the food-chain process (i.e., to induce or eliminate the oscillatory behaviour in the system).

The paper is organized as follows. The next section contains a description of the food chain process under consideration. Section 3 presents the mathematical model of the system and numerical methods used in the simulations. Finally, the main results showing the possibility of inducing and eliminating the oscillatory behaviour and concluding remarks are presented in Section 4.

## 2. Problem under consideration

The research objective presented in the paper is the theoretical analysis and simulation verification (with the use of the proposed mathematical model) of hypotheses about the use of a mixture of two various substrates for controlling the oscillatory behaviour in typical food chain. If the concentration of substrate is a limiting factor, the substrate parameters play an important role in the food chain. Because, it is possible to find two substrates of different properties, the bioreactor can be fed with a mixture of these substrates (see Fig. 1). Hence, the main contribution of the paper is the idea of using a mixture of two substrates that have different properties and are fed into the continuous flow bioreactor at the constant flow rates in order to control the oscillatory behaviour of the bioreactor.

The rate of the individual substrates to the mixture can be set by the three-way valve (3WV - represented by the parameter  $r$ ), irrespective of the dilution rate  $D$  (normalized flow) in the bioreactor. The contribution of both substrates to the mixture (i.e., the ratio of the inlet flow rates of the substrates) is proposed as a new control variable  $r$ . The new control variable has an influence on the bioprocess behaviour and the most interesting problem is the induction or elimination of the oscillatory behaviour.

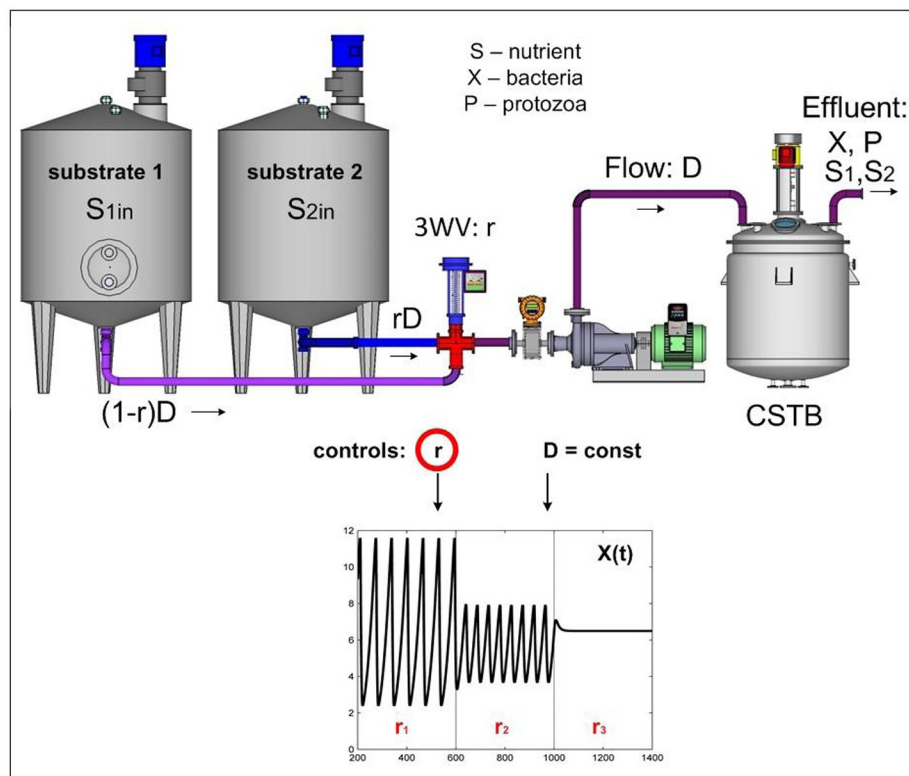


Fig. 1. The graphical presentation of the main research objective. The novelty approach, proposed by the authors, is to influence the course of bioprocess by a new control variable  $r$  (marked by the circle).

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