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Local Mixing Intensity Measurements for the Food Industry and Biotechnology

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Summary

The application of local mixing in food industry and biotechnology is demonstrated. Using a Stirring Intensity Measuring Device, SIMD-f3 and SIMD-s1, specially designed for this purpose, optimum mixing in a bioreactor, loading-flooding transition in the aeration process, and the local mixing level were controlled. The new potentialities of applying an SIMD-type unit offer its connection with a motor-driven frequence inverter.

Key words: mixing, local turbulence, SIMD, electromagnetic drive, dosage, process control

Introduction

For different food industry and biotechnological processes, it is important to know the level and quality of mixing, as it can influence the product quality. Mixing is often characterized as »good« or »insufficient«, implying the level of integral mixing which can be quantitatively described by impeller rotational speed, gas flow rate and mechanically introduced power. The above criteria characterize the average hydrodynamic situation in a technological vessel. However, not always do these criteria characterize the mixing character in a technological vessel unequivocally.

To characterize the effect of mixing in rheologically complicated and multiphase conditions most properly, we have developed a Stirring Intensity Measuring Device, SIMD *(1,2)* which ensures measurements of the kinetic energy, *E*, of flow fluctuations in various points of a technological vessel. The main advantage of the device is that it ensures precise enough measurements in multi-phase media, including fermentation media *(3)*. The data obtained thereby serve as a base for new industrial applications.

For experiments we have developed a laboratory bioreactor FAS-5.2 (produced by the Biotechnical Centre, Ltd., Latvia) with a special electromagnetic drive *(2,4)*. Its application in industry during the candy mass dosage process has been also demonstrated. Application examples show that the use of SIMD can detect the local mixing intensity. This information can be helpful to hold mixing on an optimum level in different technological processes.

The goal of the present work was the development of food and biotechnological processes control based on local mixing measurements.

Material and Methods

Stirring Intensity Measuring Device

SIMD units consist of the following basic parts:

– a set of piezoelectric transducers,
– a data processing unit,
– mathematical programs for data sampling.

A piezoelectric transducer has two versions:

1. For summary flow fluctuations energy measurements with an ellipsoid type-head (Fig. 1);

2. For one-component flow fluctuation energy measurements with a disk-shaped head part.

The data processing unit also has two versions:

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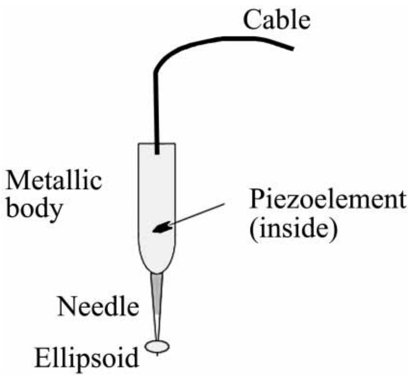

Fig. 1. Piezoelectric transducer

1. An input channel measuring unit with a possibility to adjust the calibration coefficient, integration time and RS-232 interface for data exchange (SIMD-s1);

2. An advanced version with 8 input channels with a possibility to adjust the integration time, calibration coefficients, sensitivity, frequency ranges of measured signals and data exchange parameters using RS-232 (SIMD-f3).

The transducer operates in the following way. When the turbulent multi-phase flow interacts with the flow receiver head of the transducer, mechanical oscillations are generated by the cantilever-shaped needle in the piezoelement, resulting in the generation of a proportional electrical charge signal q. To ensure the generation of identical charge signal values irrespective of the direction of the flow action, the flow interaction to the receiver is arranged in an ellipsoidal (diameter ≈ 4 mm) form. Since the interaction of identical flows in an axial direction generates the weakest signal, the ellipsoid squeezing factor is a/c = 1.5–2.5. It depends on the dimensions and mass of the transducer design elements. If measurements are carried out with a directional sensitive transducer, then a head in the form of a disk (diameter = 3–4 mm) is used. In this case, the same properties can be used in one dimensional form.

As it has been shown earlier (*2*), the interpretation of readings is based on the fact that the charge signal q is proportional to the mean kinetic energy E of flow fluctuations:

$$q \sim E \qquad /1/$$

The data processing unit ensures:
– amplifying of signals;
– low- and high-frequency filtering;
– calibration;
– integration of measured signals;
– displaying of flow kinetic energy E;
– data exchange with the computer.

Bioreactor FAS-5.2

FAS-5.3 is a 5 L laboratory bioreactor for continuous and batch cultivation of microorganisms. Its main attraction is the introduction of a novel electromagnetic drive, which ensures convenient servicing/application as well as good aseptic conditions (Fig. 2). Interchangeable stirring systems, including a counter-flow system, provide an intensive stirring mode which prevents the damage of microorganism populations. Additionally, FAS-5.3 can be completed with a 30 L stainless steel vessel equipped with the same electromagnetic drive. This 30 L vessel is in a separate frame construction, but uses the same bioprocess controller, pneumatic part and thermostat block.

Bioprocess controller

Besides traditionally measured and controlled parameters (t, pH, pO_2, n, foam, *etc.*) the bioprocess controller ensures measurements of overpressure p, air consumption Q and local stirring energy E by SIMD. The controller is based on a Basic programmable multitasking microprocessor Tiny Tiger (Wilke Technology, GmbH, Germany) (*5*), which allows easy corrections in the controller program. The Biotechnological Centre Ltd. (*6*) has obtained software and hardware tools from Wilke Technology, GmbH for realizing flexible process control projects in biotechnology and the food industry. This is a new product which appeared on the market only in 1998. It is possible to connect other additional devices in a measuring/control circuit. Feedback is carried out with the help of the following executive elements: 3 Masterflex peristaltic pumps (for pH and foam control); temperature control which is performed through a jacket *via* a closed recirculation thermostat system; an electromagnetic drive system which ensures the rotational speed in the range 10–1000 rpm using a SIEMENS inverter. Outputs of RS-232 or RS-485 serial communications are available. The mathematical Windows program samples the process data in tabular or graphical form.

Experimental conditions in bioreactor

The local kinetic energy E was measured in two points: close to the upper turbine impeller and close to the wall in the same plane. For energy measurements, SIMD-f3 was used.

Experiments were carried out in a 5 L-vessel. The diameter of the vessel was d = 150 mm, and the height h_1 = 280 mm. The height of the liquid level used in the experiments was h = 150 mm. The vessel was equipped with three radial baffles of 20 mm in width and a sparger ring of 80 mm in diameter. A standard Rushton radial turbine mixing system of 75 mm in diameter (d) and standard parameters were used in experiments. The standard parameters were: 6 blades of 19 mm in length, 15 mm in height; the disk was 50 mm in diameter. Impeller of bottom clearance was 75 mm.

To simulate the influence of pseudoplastic liquids (*e.g.* fermentation broth and other technological broths), different carboxymethyl cellulose (CMC) mass concentrations were used, *e.g.* 0.22, 0.62 and 0.8 %. Power law coefficients of the Ostwald and de Waele rheological model were found using a rotation viscosimeter REOTEST-2 (*2*).

To ensure the automatic control depending on the value of SIMD-f3 signal, a serial output signal from SIMD-f3 was transformed in an RS-485 signal and transmitted to a bioprocess controller BIO-2 *via* the RS-485

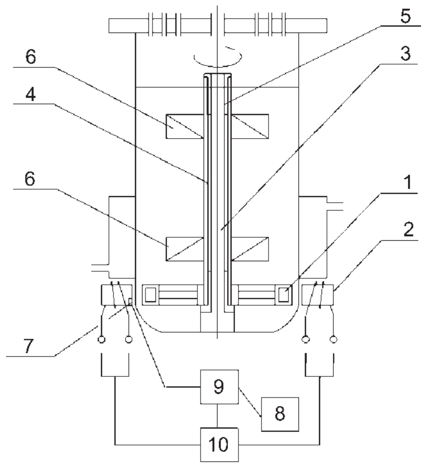

Fig. 2. Bioreactor FAS-5.2 (mixing vessel) equipped with an electromagnetic drive: (1) rotor; (2) stator poles; (3) stationary axis; (4) mobile axis; (5) bearings; (6) impellers; (7) Hall transducers; (8) adjustment panel; (9) control impulse regulator; (10) power amplifier

Bus line. The analogue output (4–20 mA) of BIO-2 was connected to the input of the SIEMENS Micromaster Junior (0.5 kW) inverter. In such a way, BIO-2 gave an output signal for changing the mixer rotational speed depending on the local energy *E* value and its time varying dynamics.

Description of a mass dosage system for high-viscous liquids

In the technological line, a part which ensures the dosage of high-viscous pseudoplastic liquids is designed to carry out the process using a modification of SIMD-f3.

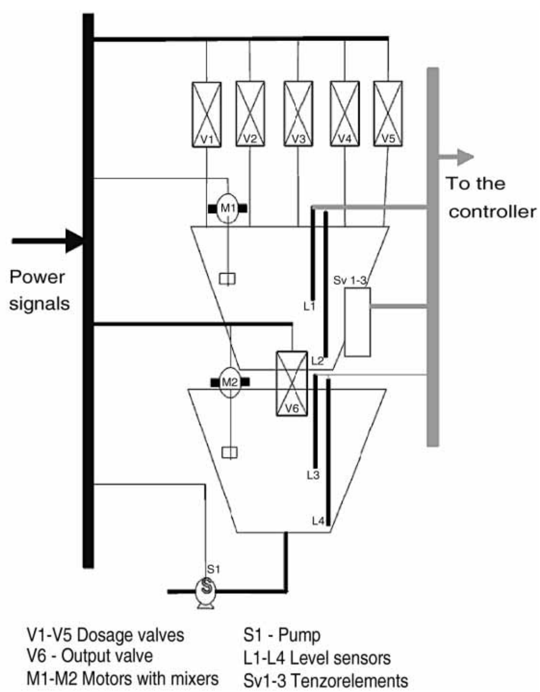

Fig. 3. Flow sheet of a caramel dosage system

The given dosage system (Fig. 3) is used for dosage, mixing and preparation of a caramel syrup. The dosage controller is based on a Basic programmable microprocessor Basic Stamp (Parallax, Inc., USA). At the factory LAIMA (Latvia's confectionery producer), 5 such controllers are installed, and all of them are connected through RS-485 to an IBM PC computer to fix each dosage cycle and summarize weight of each component in tabular form.

The dosage station PROGRESS, produced in Russia according to the license of Kloeckner (Germany), one of the world's greatest confectionery machine producers, consists of four valve pairs (each pair has one rough and one fine valve), a vibrator (for sugar dosage), the upper and lower vessels (each about 200 L). Different media (syrups, water, sugar) come from other technological stages through tubes to the valves of the dosage station. Both of the vessels have a motor with mixers. The upper and lower vessels are connected through an automatic valve (normally closed). The output of the lower vessel is connected with a pump. The upper and lower vessels are connected with tensor gauges to register the weight. Each vessel has a level meter. In addition, there are different manual valves and other accessories.

The dosage controller has the following working regimes which are selected with the key CHANGE: Calibration (weight calibration), Manual (manual switching on/off of valves), Recipe adjustment (adjustment of the mass of each component and the dosage sequence), Summary components (explanation of the realized cycles and summary mass of each component), Time (adjustment of the mixing time in the upper vessel) and Start. When the Start regime is selected, then the automatic dosage process is started according to the selected recipe. Each component (sequentially) is passed through the corresponding valve in the upper vessel. For each component, first of all, the rough valve is opened, followed, after a pause, by the fine one. To ensure better dosage precision (the valves have some inertia), the fine valve works with pauses – if the rest of the weight is smaller, then the dose is also smaller. When all the components are dosed into the upper vessel, then, at a certain moment (which can be selected from 1 to 255 s), all the components are mixed with the help of a motor M1, connected with impellers. If the lower vessel is not full (according to the readings of the level meter L2), then, after mixing, the obtained caramel mass is poured out through the output valve V6 into the lower vessel. Until the upper vessel is full, the valve V6 will not open, and the cycle will not begin. In the upper vessel, the mass is mixed by the motor M2 and then, transferred to the next technological stages by the pump. M1 and S1 are operated manually, because these operations depend on the next stages which are difficult to put under automatic control. When the mass is poured out through V6, in some time (about 2 s), the data on the last cycle are sent to the computer (*via* RS-485), and a new dosage cycle begins. If the upper vessel is full during the dosage (according to the readings of the level meter L1), then the process is interrupted. It is possible to interrupt the process manually by pressing HOLD and get out of the process by pressing RESET.

Results and Discussion

Control of optimum mixing in a laboratory bioreactor

For this purpose, different portions of CMC powder were added to the media; mass fractions of CMC were 0.22, 0.62 and 0.80 %. After adding these portions, the powder was dissolved during approximatelly 3 h. In these experiments, the BIO-2 controller was switched in the PID mode (*e.g.* automatic control). The value 54.0 J/m^3 was selected as the energy radial component set-point (E_r). For SIMD-f3, integration time was adjusted to be 99 s, and PID coefficients were as follows: P = 200, *I* = 85, *D* = 0. Fig. 4 shows average revolutions at a stabilized *E*, after which the experiments were carried out for another 40 min. At a constant energy *E*, with increasing CMC mass fraction, rotational speed also increased.

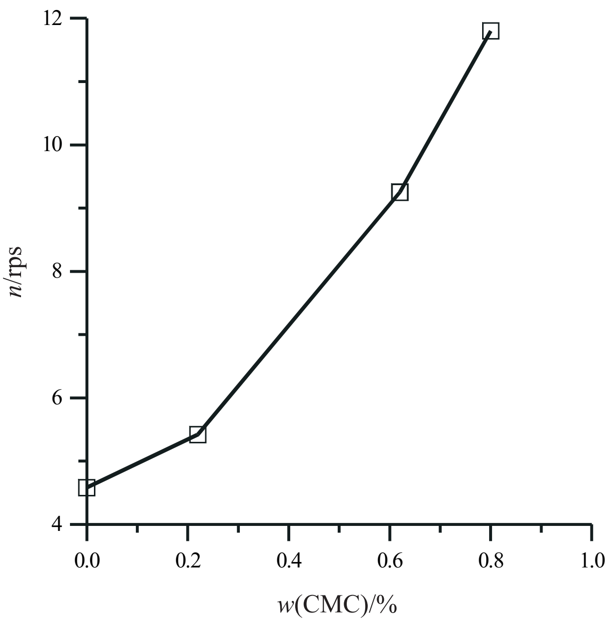

Fig. 4. Dependence of impeller rotational speed on CMC concentration at constant energy *E* = 54.0 J/m^3

The results of the earlier experiments (*2*) have shown that, at higher CMC concentrations, dispersion of control tends to increase (up to 4 %). This is explained, firstly, by the fact that impeller rotational speed values increase at higher CMC concentrations; as a result, the hydrodynamic situation in the reactor turns unstable, *i.e.* the liquid level becomes more fluctuated. Secondly, at higher CMC mass fractions and rotational speeds of the impeller, irreversible changes in the mixing medium structure can start, which affects the energy *E* distribution curve as well as the values of stabilized revolutions.

Control of the appearance of the flooding regime

The ascertaining of the flooding regime is very important in biotechnological processes of microorganism cultivation, as in this case, the air designed for aeration is supplied only to a part of the cultivation medium, *i.e.* the part located above the impeller. Another essential reason for mass exchange decrease in the flooding aeration regime is the worsening of gas bubble dispersion. To ascertain the appearance of the flooding regime, the mixer rotation rate *n* = 3 rps and the sensor location was as follows:

1. z_1 = 50 mm; r_1 = 19 mm (a point between the axis and the impeller blades tip and 50 mm above the turbine).

2. z_2 = 0 mm; r_2 = 45 mm (a point in the middle plane of the impeller relatively close to the tips of the impeller blades).

Both upper positions in an agitated batch were chosen to demonstrate a dramatic increase in energy (*E*) upon setting up the flooding flow regime (Figs. 5a and 5b).

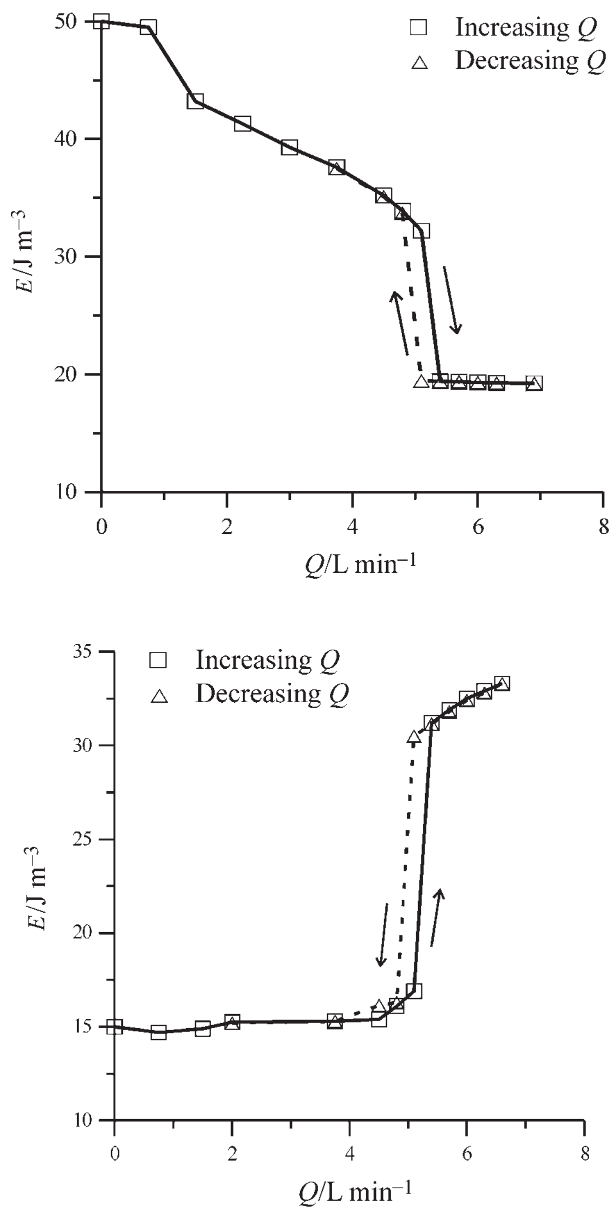

Fig. 5. Indication of the loading-flooding effect: (a) close to the Rushton turbine; **(b)** above the impeller

In the first position, still in the loading regime at increasing *Q*, there were practically no changes in energy *E*. However, upon setting up the flooding regime, the *E* value increased instantly about 2.5 times, then, at further increase in *Q*, the *E* value grew approximately proportionally. At the same time, in the second position (the one close to the impeller), the behavior was opposite, *i.e.* at the beginning, the *E* value slightly decreased with an increase in *Q* and dropped about 1.7 times upon achieving the flooding point. Further increase in *Q* did not change the *E* value in this point.

In both the positions, a hysteresis effect could be also observed, *i.e.* by decreasing *Q* from the flooding zone, loading occurred again at a lower *Q* value.

The ratio of the energy E values above the impeller and the impeller plane was chosen as the flooding effect criterion. As can be seen, in the moment when the loading state is present, this ratio is within 2.2–2.3. As the flooding regime sets up, this ratio falls to 0.6. On the contrary, as transition loading/flooding occurrs, this rate changes more than 3.5 times. Probably, this ratio can vary with varying conditions. However, in any case, this will be essential enough to serve as an indicator of flooding set up. As such information is received in the production process, the operator performs operations to prevent the flooding regime, for example, decreases the flow rate *Q*.

Control of mixing in the candy (caramel) production process

In many processes used in food industry, mixing regime is determined by the state of the rheological properties of the medium. A typical example is the case when the medium solubility impairs as a result of an increase in viscosity. In this case, it is important to increase the rotational speed of the impeller to ensure necessary turbulence for better solubility in the greater part of the reactor, wherever possible.

The preparation of the caramel (a special type of candies) corresponds to the above-mentioned situation. The automatic preparation of this mixture proceeds in accordance with Fig. 3 and this article's section **Materials and Methods**, *Description of a mass dosage system for high-viscous liquids*.

In accordance with the scheme shown in Fig. 6, a piezotransducer is placed in lower vessel of the dosage system (Fig. 3) in a characteristic point of the vessel. In this case, the characteristic point defines the site where the variations in energy *E* owing to the change in viscosity and rheological properties are reflected best. In its turn, SIMD-s1, which processes the signal received from the piezotransducer, forms an analogue output signal of 4–20 mA, which is proportional to the energy *E* value. To ensure variations in the rotation rate of the mixer engine M2 depending on energy *E* values, SIMD-s1 is connected at the analogue output to the analogue input of the inverter. The latter is designed for PID control.

This appliaction is important, as the caramel mass aparent viscosity (60–250 p/s) can vary rather dramatically in the production process depending on the product form (*e.g.* the selected recipe) and raw materials (mainly syrup-type) applied. However, in its turn, as is shown in the present study, energy *E* values tend to decrease with increasing viscosity at a constant rotational speed of the impeller. At higher viscosity values, a higher rotational speed of the impeller is required to ensure a better mixing of the components and prevent the sedimentation of sugar. As an SIMD transducer is placed in the vicinity of the M2 impeller, energy *E* can be measured in the given point. At a constant rotational speed rates of the impeller, energy *E* tends to decrease with increasing viscosity. Hence, to ensure a constant intensity of mixing (*E* = constant), the M2 impeller rotational speed should be increased, which is performed using a SIEMENS inverter.

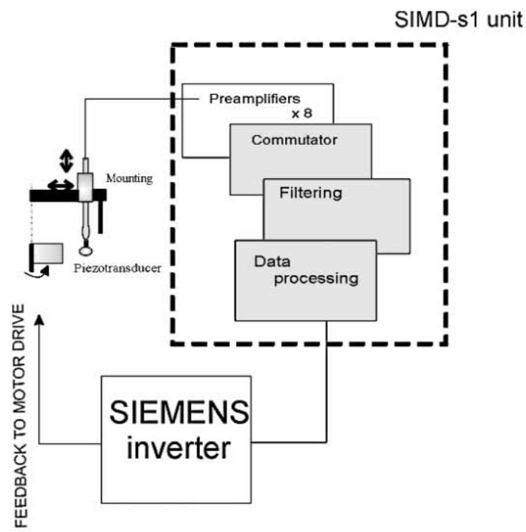

Fig. 6. Local mixing control during the caramel mass preparation process using SIMD-s1; number of mixers – 2. Diameter of mixer d = 210 mm. Distance between the transducer head and the lower mixer 24 mm

Control of mixing in biotechnology

Hundreds of annual publications, including our studies (*2,3,7–9*), deal with this phenomenon. Within the context of the present work, the main point is that SIMD and FAS-5.2, having minimum vibrations for correct *E* measurements, are valuable tools for solving process optimization problems (*10–16*). Two examples could be mentioned here illustrating the use of SIMD. In all cases, the function of the energy *E* distribution across the bioreactor volume *E(r,h)* was used as primary information on mixing in the bioreactor. To reduce the number of the measured points, energy *E* was measured in the zones where the intensity of *E* was the highest. These zones reflect the change of *E(r,h)* distribution, influenced by different factors (*n*, *Q*, biomass, *etc.*) most sensitively. When studying the effect of mixing on cultivation of *A. niger*, various mixing systems were used in a 3 L-bioreactor:

- the standard turbine mixer STMS,
- the counter-flow mixer with one drive CFMS 1,
- the counter-flow mixer with two drives CFMS 2.

In counter-flow mixers, the upper and lower mixing flows interact in opposite directions. Besides, in CFMS 1, axial flows interact oppositely, and in CFMS 2, axial and tangential flows interact in opposite directions. Thereby, an even distribution of the introduced energy is ensured. As a criterion of the evenness of energy distribution, k_D has been used:

$$k_D = \frac{1}{V} \iint_{(V)} \sqrt{(dE/dr)^2 + (dE/dh)^2}\; r\, dr\, dh \qquad /2/$$

It may be assumed that the greater k_D, the greater the maximum shear stresses in the bioreactor.

In all the three mixing systems, *A. niger* cultivation was carried out at the same mechanically introduced power. An inversely proportional dependence between citric acid productivity and k_D was established. In its turn, the dependence between k_D and the productivity according to the sugar source indicates that the maximum unevenness of k_D is ≈1100 J/m^3 (Fig. 7). This is confirmed also by the cultivation practice, which shows that each *A. niger* strain has a pronounced mixing intensity optimum.

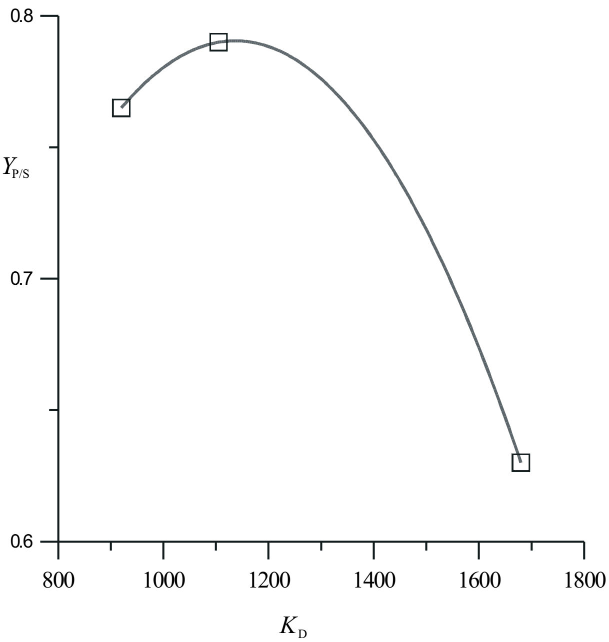

Fig. 7. Dependence of citric acid productivity $Y_{P/S}$ on the unevenness K_D of energy *E* in *A. niger* cultivation

It has been established that, for bacterial microorganisms, the optimum mixing in STMS is achieved at higher mixing rotational speed values rather than for mycelial fungi. The lysine producer, *Brevibacterium flavum*, was investigated regarding the correlation between the conventional maximum energy *E* (which was measured at some distance from the impeller blades in its central plane) and the activity of the Krebs cycle enzyme aconitase, since it reflects the dynamics of the growth and biosynthesis process more sensitively. It has been established that there is an obvious proportionality between the aconitase activity and energy *E* (Fig. 8).

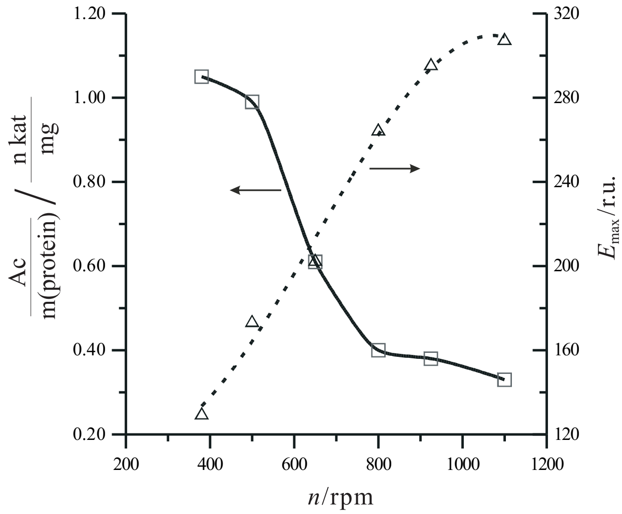

Fig. 8. Dependence of aconitase activity *Ac* and energy *E* in *Brevibacterium flavum* fermentations

As can be seen from the above examples, the SIMD application way depends on the fact, which mixing aspect limits the process yield.

Control of oxygen partial pressure pO2

Taking into account flexibility, which is ensured by the bioprocess controller BIO-2, it is possible to perform also other process control tasks connected with local mixing. For example, it ensures the necessary minimum and maximum mixing levels during the pO_2 control process.

In fermentation processes, a definite level of dissolved oxygen partial pressure pO_2 is normally provided by changing the automatic rotational speed rate *n* of the impeller and the supplied air *Q*. To secure the limit of the minimum and maximum mixing level, the impeller minimum and maximum rotational speed rates, respectively, are set. Taking into account the fact that, in the fermentation process, rheological properties can vary and that different amounts of the supplied air change the distribution of the introduced energy, rotational speed rate does not reflect the mixing level.

To ensure the given control task, it is necessary to place 2 piezotransducers in the fermentation vessel, *i.e.* one close to the impeller, and the other approximately in the middle of the plane between the impeller blades and the reactor wall. The maximum mixing level E_{max} is controlled with the first transducer, and the minimum mimimg level E_{min} with the second transducer. It means that revolutions will be increased according to needs for pO_2 control until $E < E_{max}$, and decreased until $E > E_{min}$ are secured.

Thus, for example, in the case of *Trichoderma viride* (the fermentation was carried out in a FU-8 bioreactor with a working volume of 3 L), it has been established how, controlling pO_2 according to the energy *E* limit, the rotational speed continues to vary (in this case, to increase) (Fig. 9). This is connected with the fact how the

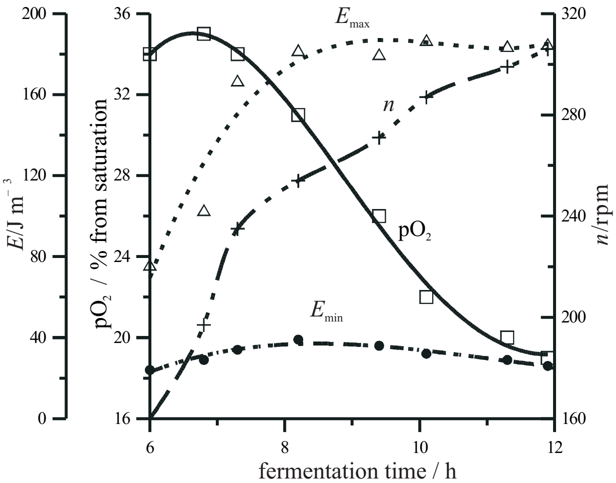

Fig. 9. Dynamics of variations in energy *E* and stirrer speed *n* during *Trichoderma viride* fermentations

rheological properties of the medium vary as the fungal biomass increases.

Conclusions

The results of the present study show that the application of a special device such as SIMD is essential in processes where mixing aspects can affect the product quality and production economy. Till now, information on the application of analogous equipment in the food industry has not been available. Therefore, apart from the above mentioned examples, there can be also other cases that would exemplify the attraction of these applications.

The potentialities of applying local flow fluctuation kinetic energy measurements are increased by the development of a frequency inverter. By connecting SIMD with the frequency inverter, conditions arise for automatic control of local mixing, that could ensure both product quality improvement and economic application of electric energy.

Acknowledgements

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Mjerenje intenziteta lokalnog miješanja u prehrambenoj industriji i biotehnologiji

Sažetak

Prikazana je primjena lokalnog miješanja u prehrambenoj industriji i biotehnologiji. Primjenom uređaja za mjerenje intenziteta miješanja (SIMD-f3 i SIMD-s1), specijalno konstruiranih za tu svrhu, kontrolirano je optimalno miješanje u bioreaktoru, izmjena pri opterećenju i nadiranju u procesu aeracije te razina lokalnog miješanja. Primjena uređaja tipa SIMD omogućuje njegovo povezivanje s motorom tjeranim inverterom frekvencije.