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FERMENTATION OF SUGARCANE BAGASSE HYDROLYSATES BY *Mucor indicus*

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The aim of the research was to analyze the fermentability of sugarcane bagasse prehydrolysates using *Mucor indicus*. The prehydrolysates were obtained by acid prehydrolysis of sugarcane bagasse and were detoxified before fermentation. A mold strain was also adapted to the inhibitors contained in the prehydrolysates. The production of ethanol and sugar consumption were investigated under aerobic and oxygen limited conditions. For the original strain, the consumption of sugars was incomplete and ethanol was produced at a yield of 0.39 ± (0.02) g g^{-1}. The increased tolerance of *M. indicus* to the inhibitors resulted in a complete fermentation with total glucose consumption. Most of the xylose was consumed in all experiments, with the highest consumption in aerobic fermentations. Ethanol was the main product of fermentation and its yield was 0.41 ± (0.02) g g^{-1} at oxygen limited conditions and 0.37 ± (0.02) g g^{-1} at aerobic conditions. The use of other carbohydrates besides the monosaccharides was also investigated. Another advantage of *M. indicus* detected during the investigation was its ability to ferment pentoses, hexoses and oligosaccharides.

Key words: bioethanol, sugarcane bagasse, acid hydrolysis, *Mucor indicus*, filamentous fungi.

The increase of oil prices, the imminent of oil depletion and the need to mitigate the greenhouse effect lead to the increase of interest for the use of fuel ethanol. The use of biofuels such as ethanol reduces the environmental impact generated by the greenhouse gas emissions [1].

During ethanol production by fermentation, saccharine, amylaceous and lignocellulosic raw materials are used. The saccharine materials have the advantage that their component carbohydrates are monosaccharides and oligosaccharides, whereby the substrate can be fermented directly by the ethanologenic microorganisms. The production of ethanol from sugar and starch-based raw materials is a well-established industrial process that has been used for many years. However, the availability of these raw materials is not enough to cover the demands of fuel ethanol production. In addition, when using materials such as sugar cane and maize, the raw material represents between 60 and 70% of the cost of ethanol production [2].

Lignocellulosic materials such as agricultural, industrial and forest residues are abundant and cheap feedstocks for bioethanol production [3]. Sugarcane bagasse is one of the most promising materials for ethanol production with approximated 163 Lt^{-1} as potential ethanol yield [4]. From economic point of view, this residue is appropriated for industrialization since it is produced in higher amounts and is usually available in central collection sites, such as sugar mills. A large fraction of this lignocellulosic material is often disposed of by burning, which is not restricted to developing countries alone, but is considered as a global phenomenon [5].

Bagasse is the separated residue after the extraction of sugarcane juice and is the main by-product of the sugar industry. Bagasse has high carbohydrate content and is of interest to produce ethanol. The literature reports that bagasse contains 41–52% cellulose, 25–30% pentosans and 18–25% lignin [4]. Xylans are, after glucans, the most important carbohydrates in the bagasse. Xylose is almost one third of the sugar content in bagasse hydrolysates [6]. Arabinose, is another pentose that is found in significant quantities in hydrolysates. For the production of ethanol from bagasse to be economically viable, an efficient conversion of all its sugars, including pentoses, into ethanol is required.

An important requirement for the organisms to be used in the industrial

production of biofuel is the ability to metabolize pentoses, since xylose and arabinose are significant components of the hydrolysates of this type of material, especially in hardwoods and agricultural waste. The yeast *S. cerevisiae*, despite its strengths as an ethanologenic organism, cannot ferment pentoses. On the other hand, it is known that most organisms with natural ability to use xylose have several drawbacks that limit the potential of their industrial use [7].

Many researchers have devoted attention to the study of some molds of the *Zygomycetes* class, such as *Mucor indicus* and *Rhyzopus oryzae*, which are of interest to produce cellulosic ethanol. The conversion of pentoses and hexoses into ethanol by *M. indicus* and *R. oryzae* both in mineral media and in lignocellulosic hydrolysates has shown [8–13]. It is known that the morphology of molds of the *Mucor* genus depends on aeration and nutrition conditions. In aerobic conditions, filamentous growth occurs, whereas in anaerobiosis the growth is yeast-like. On the other hand, yeast growth occurs only using hexoses as carbon sources, while in mycelial growth molds can use different types of sugars. The yeast growth is usually accompanied by the production of ethanol, which is minimal when the growth is mycelial. However, it has been reported that some *Mucor* molds are Crabtree-positive organisms, so they also produce ethanol under aerobic conditions [14]. There are reported studies with *M. indicus* in hydrolysates from forest residues [10, 11, 15], rice straw [9], orange peels [16, 17], peanut shells [13] and spent sulfite liquor [18]. High yield ethanol production is among the most important features of this strain [19].

In the present work, the fermentability of bagasse prehydrolysates with *M. indicus* was investigated. A mold strain was adapted to the inhibitors contained in the prehydrolysates . The production of ethanol and the consumption of sugars in cultures of the adapted strain were investigated under aerobic and oxygen limited conditions.

Materials and Methods

Acid prehydrolysis

Bagasse prehydrolysis was carried out with H_2SO_4 in a ratio of 2 g per 100 g of dry biomass, a solid concentration of 10%, at 121 °C and for 1 h. When the prehydrolysis finished, the prehydrolyzate was separated by filtration. $Ca(OH)_2$ was added to adjust the pH at 5.0; the calcium sulphate formed was deposited and the supernatant was centrifuged to remove the solids that could not be separated by sedimentation. The clarified prehydrolyzate was preserved at 4 °C until its later use.

Detoxification

Bagasse acid hydrolysates were detoxified by treatment with $Ca(OH)_2$ according to the process known as overliming [20]. A solution of 20% $Ca(OH)_2$ (m/v) was added to the hydrolyzate until reaching a pH 10. After one hour of treatment, the hydrolysate was filtered under vacuum, adjusted to pH 5.5 with H_2SO_4 and filtered again. The hydrolysates used were supplemented with glucose, $(NH_4)_2SO_4$, KH_2PO_4, $MgSO_4 \cdot 7H_2O$, $CaCl_2 \cdot 2H_2O$, vitamin and mineral solution as referred [9].

Preparation of the inoculum

The strain *M. indicus* CCUG 22424 donated by the University of Borås, Sweden was used. It was conserved at 4 °C in the medium PDA, consisting of glucose (20 g L^{-1}), potato extract (4 g L^{-1}) and agar (15 g L^{-1}).

The inoculum from *Mucor indicus* was prepared by adding 10 ml of sterile distilled water on the mycelium developed in solid PDA medium and, with the help of a sowing loop, the mycelium was vigorously separated. Another 10 mL of sterile distilled water was added, the spore suspension was transferred to a sterile bottle, and stored at 4 °C. The preculture, inoculated with 1–2 mL of the spore suspension, was prepared with 10 mL of synthetic medium incubated at 32 °C under agitation (75 rpm) for 16 h. One milliliter of the spore suspension contained $5 \cdot 10^7$ spores mL^{-1}.

Fermentation regimes

Aerobic fermentation: One liter of hydrolyzate prepared as was indicated above and 100 ml of the inoculum were added into a 2 L capacity bioreactor. Antifoam was added in concentration 0.1 mLL^{-1}, the pH adjusted to 5.5 and incubated at 32 °C with 300 rpm stirring and 300 mL min^{-1} air supply. Samples were taken every two hours until 12 hours of fermentation and after 24 hours.

Fermentation under oxygen limited conditions: Bottles of 30 mL containing 25 mL of medium were used, equipped with rubber caps and needles for the removal of CO_2, and subjected to moderate agitation (50 rpm). Samples were taken as under aerobic conditions but the time of fermentation was extended until 72 hours.

The fermentability criteria of the hydrolysates were: final concentration of ethanol (g L^{-1}), yield of ethanol (g g^{-1}), maximum volumetric productivity of ethanol

(g L^{-1} h^{-1}), speed of glucose consumption (g $L^{-1}h^{-1}$).

Adaptation of M. indicus to prehydrolysates

As a first stage of the adaptation process, mold tolerance was evaluated at different concentrations (0, 25, 50, 75, 100%) of prehydrolysate. The liquid medium for adaptation was prepared by diluting the bagasse prehydrolysate with water and adding the nutrients (Table 1).

In the media in which the prehydrolysate was concentrated at 25 and 50%, good growth was achieved and the consumption of sugars was total, while in the prehydrolysates concentrated at 75 and 100%, little metabolic activity was observed. Therefore, a portion of the culture obtained in the 50% concentrated prehydrolysate was isolated, seeded in a solid medium enriched with prehydrolysate and incubated at 30 °C.

After 72 h of incubation at 30 °C, a spore suspension was prepared, from which 2 mL was added to a 250 mL Erlenmeyer flask containing 100 mL of the M1 medium (Table 1). Two replicates were incubated at 30 °C with shaking (130 rpm). After observing an abundant growth, after 24 h, a portion of the culture was transferred to the M2 medium and incubated under similar conditions. Similarly, when the growth was abundant, a portion of the crop was transferred to the M3 medium. Successive transfers to media with increasing concentrations of prehydrolysate continued. Finally, a culture portion of the more concentrated medium was taken and transferred to a plate with prehydrolysate and incubated at 30 °C for 72 h.

Analytical methods

The metabolites in the culture media, were quantified by HPLC (Waters, Milipore, USA). Glucose, xylose and arabinose, ethanol, glycerol and acetic acid were separated on an Aminex HPX-87H column at 60 °C with 0.6 mLmin^{-1} of 5 mM H_2SO_4 as eluent.

For the analysis of oligosaccharides an acid post hydrolysis was carried out followed by the chromatographic analysis of the hydrolysates.

The sugars were analyzed as total reducing sugars (TRS) using the 3.5-dinitrosalicylic acid method.

The cellular biomass content was analyzed gravimetrically. Culture samples of 5 mL were centrifuged (4 000 rpm) for two minutes in pre-weighed tubes. After washing the cells with distilled water, the centrifugation was repeated under the same conditions. Once the supernatant was expelled, the tubes were dried to constant weight at 45 °C. The biomass content during the fermentations was monitored by measurements of optical density at 620 nm, using a spectrophotometer.

Calculations

The calculations were made based on the mean values of the experimental measurements. The yield of ethanol, biomass and glycerol from the total sugars (g g^{-1}) were calculated as the amount of ethanol, biomass and glycerol formed (g) divided by the initial amount of total sugars (g). The maximum volumetric productivity (g L^{-1} h^{-1}) was based on the amount of ethanol produced per liter of medium per hour during the first 6–8 hours of fermentation. The speed of consumption of sugars (g L^{-1} h^{-1}) was calculated by dividing the amount consumed between the fermentation time.

All the experiments were triplicated. Calculations and diagrams construction were done using the Microsoft Excel 2016 software. The standard deviation is shown in the tables in parentheses. The data obtained was processed by statistical analysis methods using the Student's *t*-test with confidence $P < 0.05$.

Results and Discussion

Fermentability of hemicellulosic hydrolysates of bagasse with M. indicus

Initially the ability of the *M. indicus* CCUG 22424 strain to ferment a prehydrolysate

Table 1. **Composition of the media used in the adaptation of a strain of *M. indicus* to bagasse prehydrolysates**

Medium	Prehydrolysate,%	Water,%	Nutrient solution,%
M1	50.0	45.0	5.0
M2	65.0	30.0	5.0
M3	80.0	15.0	5.0
M4	90.0	5.0	5.0
M5	92.5	2.5	5.0
M6	95.0	0.0	5.0

of sugarcane bagasse was investigated. The prehydrolysate was obtained by acid prehydrolysis of bagasse and was detoxified before fermentation. The fermentation was performed under aerobic conditions. The fermentation time was 24 h, taking into account previous results with this strain in hydrolysates of other materials.

During the fermentation a relatively fast consumption of the total reducing sugars (TRS) and acetic acid was observed, which was exhausted in 12 h (Fig. 1). The consumption of sugars was incomplete, since certain amounts of residual sugars were detected in the medium at the end of the fermentation. The incomplete consumption may be due to the sensitivity of the mold to some inhibitors contained in the prehydrolyzate, of which acetic acid is excluded because it was completely consumed at the end of fermentation. This result agrees with criteria of other researchers in this subject [18]. The short duration of fermentation may also have influenced the low consumption of sugars.

The main products detected were mycelial biomass, ethanol and small amounts of glycerol. The formation of ethanol under aerobic conditions indicates that *M. indicus*, like other filamentous fungi is a Crabtree-positive organism [14]. The yield of ethanol (0.39 g g^{-1}) is comparable to the yields of 0.40–0.45 g g^{-1} obtained in the fermentation of acid hydrolysates of forest residues with the same organism [10, 15, 17], but the volumetric productivity of ethanol was lower in the present work (Table 2). These results were superior to those achieved by [9], who demonstrated that fermentations of rice straw hydrolysates with *M. indicus* can achieve volumetric productivity comparable to those achieved with the yeast *P. stipitis*, which is known for its wide range of sugar utilization. The results with orange peel acid hydrolysates were also surpassed [16]. These results confirm the potential of *M. indicus* as an ethanologenic organism and support its use in bagasse hydrolysates.

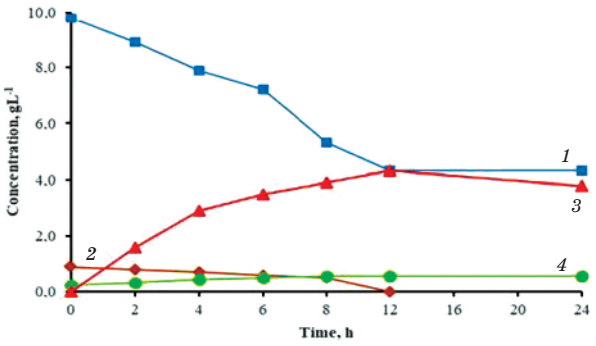

Fig. 1. **Consumption of TRS (*1*) and acetic acid (*2*); formation of ethanol (*3*) and glycerol (*4*) during the fermentation of a sugarcane bagasse acid prehydrolysate with *M. indicus***

Evaluation of fermentation with a strain of M. indicus adapted to bagasse prehydrolysates

As the fermentation with the *M. indicus* CCUG 22424 strain was inhibited by some of the toxic components of the bagasse prehydrolysate, it was decided to adopt a strategy to increase mold tolerance. This alternative of adaptation to different cultivation conditions, substrate and inhibitors was also suggested by [19] as one of the ways for contribution to the improvement of the results in this area.

A process of adaptation of the study strain to bagasse prehydrolysates was developed using the methodology described above.

Once the adaptation process was completed, the adapted strain was isolated and its behavior was evaluated in fermentation of bagasse prehydrolysates. The fermentations were carried out both in conditions of oxygen limitation and in aerobic conditions and extended for 72 h. In Fig. 2 is shown the data obtained at aerobic conditions.

The behavior of the adapted strain in the fermentation of the prehydrolysates indicates that the adaptation process was effective. The increased tolerance of *M. indicus* to the inhibitors resulted in a complete fermentation with total glucose consumption (Fig. 2, *a*). Some aspects were observed in which the behavior of the adapted strain differs from the behavior of strain CCUG 22424, which is used in most of the works on *M. indicus*. For example, with the adapted strain the glucose consumption started just from the beginning of the fermentation, unlike the experiments reported by [15], in which the mold needed some time to adapt to the hydrolysates, and the consumption of glucose only started after 24 h. Good results with the same strain of *M. indicus* also were achieved in fermentation with spent sulfite liquor. It was the best and fastest ethanol producer from the tested fungi *Aspergillus oryzae*, *M. indicus* and *Rhizopus oryzae* but it was necessary to dilute hydrolyzates [18].

According to [11] using CCUG 22424 strain, the concentration of xylose in aerobic cultures remained constant, both in

Table 2. **Yield and volumetric productivity of ethanol in fermentation of hemicellulosic hydrolysates of different materials, obtained by acid hydrolysis**

Material	Organism	Aeration regime	$Y_{E/TS}$, g g^{-1}	Q, g L^{-1} h^{-1}	References
Sugarcane bagasse	*M. indicus*	Aerobic	0.39	0.48	This work
Forest residues	*M. indicus*	Aerobic	0.45	0.83	[10]
Forest residues	*M. indicus*	Aerobic	0.40	–	[15]
Rice straw	*P. stipitis*	Aerobic	0.38	0.05	[9]
Rice straw Orange peels	*M. indicus* *M. indicus*	Aerobic Aerobic	0.24 0.36	0.03 –	[9] [16]
Forest residues	*M. indicus*	Anaerobic	0.46	–	[15]
Forest residues	*M. indicus*	Anaerobic	0.44	–	[11]
Rice straw Orange peels	*M. indicus* *M. indicus*	Anaerobic Anaerobic	0.38 0.35	0.04 –	[9] [16]

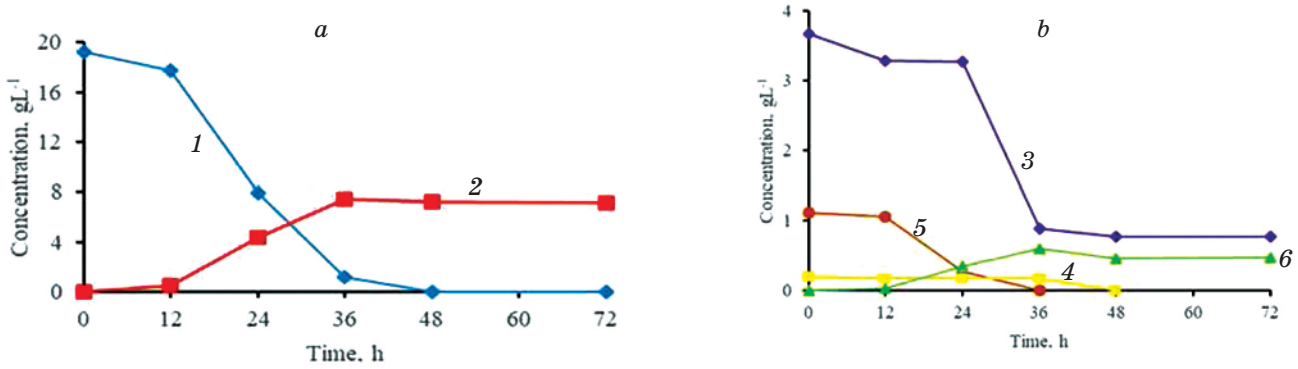

Fig. 2. **Kinetics of the fermentation of bagasse hydrolysates with the strain adapted from *M. indicus*:** *a* — glucose consumption (*1*) and ethanol formation (*2*); *b* — consumption of xylose (*3*), arabinose (*4*) and acetic acid (*5*) and formation of glycerol (*6*)

synthetic media and in hydrolysates while there were traces of hexoses in the media. It is associated with the diauxic behavior that *M. indicus* exhibits. In the present investigation the consumption of xylose, although slow, started from the beginning of the fermentation (Fig. 2, *b*). As the concentration of glucose in the medium decreased, especially below 8 gL^{-1}, the consumption of xylose was accelerated. At the end of the fermentation, only 0.7 gL^{-1} of residual xylose remained, which indicates that practically 80% of the xylose contained in the prehydrolysate was consumed.

Most of the xylose was consumed in all experiments, with the highest consumption in aerobic fermentations (Table 3). The consumed xylose was not converted to xylitol, because this metabolite was not detected in the medium. This is a relevant result that, in agreement with previous results [15], shows an important difference between *M. indicus* and yeasts capable of fermenting xylose, such as *P. stipitis* and *P. tannophilus* [7]. The other pentose present in the bagasse prehydrolysate, arabinose, was completely consumed in 48 h of aerobic culture (Fig. 2, *b*), while under conditions of oxygen limitation it was not consumed. This result differs from previous reports with the original strain [15], in which arabinose consumption was poor even in aerobic conditions.

Acetic acid was also rapidly consumed in the presence of oxygen and was depleted in 36 h (Fig. 2, *b*), whereas in anaerobic fermentation consumption was very poor. This observation partially agrees with results reported in the literature, since in the work

[15] acetic acid is also consumed in aerobic conditions, although consumption is much lower than in this work, while [11] also report a small consumption in anaerobic conditions. It is also noteworthy that the formation of pyruvic, succinic, lactic and formic acids was not observed, some of which have been detected in previous research [9,11].

Ethanol was the main product of fermentation and there was no significant difference between the aeration schemes used (Table 3). The yield achieved under conditions of oxygen limitation is comparable to that achieved with *S. cerevisiae* from glucose, although the volumetric productivity is low. However, other authors report ethanol productivities with *M. indicus* comparable to those achieved with *S. cerevisiae* [9, 15].

Under aerobic conditions, the ethanol yield was 0.37 g per g of total sugars (Table 3). This yield is lower than the 0.42–0.44 g g^{-1} achieved in aerobic mold cultivation in hemicellulose prehydrolysates of forest residues [10,15]. This is due to the fact that the prehydrolysates used in the previous reports were from softwoods, so they had high contents of mannose and glucose. They are hexoses easily metabolized by *M. indicus* in aerobic conditions [15], whereas in the prehydrolysates used in the present work the main sugar was the xylose, whose conversion in ethanol by the mold is inhibited by the presence of oxygen in the medium [11]. On the other hand, a part of the sugars was used in the formation of mycelial biomass. There was no significant biomass formation (Table 3), but in anaerobic fermentation of wood hydrolysates, it was zero [11, 15] and ethanol yields were high (Table 2). It should be noted that with *M. indicus* the yield of ethanol is at the expense of all sugars, including pentoses, while *S. cerevisiae* only ferments the hexoses.

A peculiarity of aerobic cultures is that the ethanol produced was subsequently consumed partially. It was shown that the consumption of ethanol by *M. indicus* increases proportionally to the increase in the level of aeration [11]. It was also confirmed by [18] when the concentration of ethanol decreased slowly at the end of cultivation of spent sulphite liquor with *M. indicus*. The consumption of ethanol by the ethanologenic organism itself represents a challenge for the design of industrial processes based on the use of *M. indicus* strains.

In addition to ethanol and biomass, another product formed was glycerol. Glycerol formation was low in all fermentations (Fig. 2, *b*), but there is no significant difference between the aeration schemes used (Table 3).

In the present work, the use of other carbohydrates besides the monosaccharides was also investigated, considering that in the revised literature this investigation was not reported. Oligosaccharide determinations were made in the prehydrolysates before and after fermentation. The experiment revealed that the contents of xylo-, gluco- and arabino-oligosaccharides decreased with fermentation (Table 4). This result indicates that *M. indicus* has an enzyme system that allows it to hydrolyze the oligosaccharides that have been solubilized during the pretreatment. This is another strength of this organism for the production of ethanol, especially for the simultaneous scheme of hydrolysis and fermentation (SSF), in which the use of *M. indicus* could be dispensed with the addition of β-glucosidase. It has a significant economic importance considering the high cost of enzymes.

It was confirmed that sugarcane bagasse prehydrolysates can be fermented to ethanol by *M. indicus* under aerobic conditions. The investigated strain is similar to *S. cerevisiae* in terms of the fermentation products and the ethanol yields achieved

Table 3. **Parameters of the fermentation of prehydrolysates with the adapted strain of *M. indicus*. $YE_{/TS}$, $Y_{X/TS}$, $Y_{G/TS}$, yield of ethanol, biomass and glycerol per gram of total sugars; Q, volumetric productivity of ethanol; GCS, glucose consumption speed; Cxyl, consumption of xylose**

Aeration regime	$Y_{E/TS}$, g g^{-1}	$Y_{X/TS}$, g g^{-1}	$Y_{G/TS}$, g g^{-1}	Q, g L^{-1} h^{-1}	GCS, g L^{-1} h^{-1}	Cxyl,%
Aerobic	0.37 (0.03)	0.21 (0.02)	0.03 (0.00)ы	0.22 (0.02)	0.54* (0.03)	79.10* (0.70)
Oxygen limited conditions	0.41 (0.03)	0.12 (0.01)	0.02 (0.00)	0.21 (0.02)	0.48 (0.03)	66,80 (0.70)

Note: * — $P < 0.05$ compared to Oxygen limited conditions.

Table 4. **Concentration of oligosaccharides in the hydrolyzate before and after fermentation with *M. indicus***

Prehydrolysates	Xylo-oligosaccharides, g L^{-1}	Gluco-oligosaccharides, g L^{-1}	Arabino-oligosaccharides, g L^{-1}
Not fermented	5.50* (0.46)	5.50* (0.26)	2.30* (0.20)
Fermented	4.00 (0.20)	2.70 (0.20)	1.60 (0.10)

Note: *— $P < 0.05$ compared to fermented hydrolysates.

in anaerobic conditions, but there is an advantage over the spectrum of utilization of sugars, which was demonstrated with its capacity to ferment pentoses, hexoses and oligosaccharides.

A feasible procedure was developed for the adaptation of *Mucor indicus* to lignocellulosic hydrolysates. The adapted strain was suitable for the production of ethanol from sugarcane bagasse hydrolysates.

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ФЕРМЕНТАЦІЯ ГІДРОЛІЗАТІВ ЖОМУ ЦУКРОВОЇ ТРОСТИНИ З ВИКОРИСТАННЯМ *Mucor indicus*

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Метою дослідження було проаналізувати ферментабельність жому прегідролізату цукрової тростини за допомогою *Mucor indicus*. Прегідролізати було отримано шляхом кислотного гідролізу жому цукрової тростини й детоксифіковано перед ферментацією. Штам форми адаптовано до інгібіторів, які містяться в прегідролізатах. Виробництво етанолу й споживання цукру досліджено за аеробних умов та обмеження доступу кисню. Для оригінального штаму споживання цукру було неповним, а етанол вироблявся з виходом 0,39 ± (0.02) г г$^{-1}$. Збільшення толерантності *M. indicus* до інгібіторів сприяло 100% -й ферментації з повним споживанням глюкози. Найбільший рівень споживання ксилози зафіксовано в експериментах з аеробною ферментацією. Етанол був основним продуктом ферментації, і його вихід становив 0,41 ± (0.02) г г$^{-1}$ за умов обмеження доступу кисню і 0,37 ± (0.02) г г$^{-1}$ за аеробних умов. Також було досліджено використання не лише моносахаридів, але й інших вуглеводів. Перевагою *M. indicus*, виявленою під час досліджень, була здатність ферментувати пентози, гексози й олігосахариди.

Ключові слова: біоеталон, жом цукрової тростини, кислотний гідроліз, *Mucor indicus*, ниткоподібні гриби.

ФЕРМЕНТАЦИЯ ГИДРОЛИЗАТОВ ЖОМА САХАРНОГО ТРОСТНИКА С ИСПОЛЬЗОВАНИЕМ *Mucor indicus*

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Целью исследования было проанализировать ферментабельность прегидролизатов жома сахарного тростника с использованием *Mucor indicus*. Прегидролизаты были получены путем кислотного предварительного гидролиза сахарного тростника и детоксифицированы перед ферментацией. Штамм формы также был адаптирован к ингибиторам, содержащимся в прегидролизатах. Производство этанола и потребление сахара было исследовано в аэробных условиях при ограничении доступа кислорода. Для исходного штамма потребление сахаров было неполным, а этанол получен с выходом 0,39 ± (0.02) г г$^{-1}$. Увеличение толерантности *M. indicus* к ингибиторам привело к 100% -й ферментации с полным потреблением глюкозы. Большая часть ксилозы использована во всех экспериментах с наибольшим потреблением в аэробных условиях. Этанол был основным продуктом ферментации, и его выход — 0,41 ± (0.02) г г$^{-1}$ в условиях ограничения доступа кислорода и 0,37 ± (0.02) г г$^{-1}$ — в аэробных условиях. Было также исследовано использование и других углеводов, кроме моносахаридов. Преимуществом *M. indicus*, обнаруженным во время исследования, была его способность ферментировать пентозы, гексозы и олигосахариды.

Ключевые слова: биоэтанол, жом сахарного тростника, кислотный гидролиз, *Mucor indicus*, нитевидные грибы.