Bioconversion of rye straw by micromycetes

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Micromycetes *Myrothecium verrucaria* and *Galactomyces geotrichum* were investigated for their ability to utilize remnants of *Secale* straw and to enrich it with proteinous substances. The investigation showed that *Galactomyces geotrichum* was a more successful micromycete in rye straw bioconversion. The fungus after a 60-day cultivation on straw increased the protein amount 2.06 times and the total amount of amino acids 1.25 times as compared to the control. The regularity in changes of proteinous substances depending on the cultivation time in *Galactomyces geotrichum* and *Myrothecium verrucaria* was different. After a long-time cultivation on rye straw in the case of *Galactomyces geotrichum*, the content of proteinous substances increased and in the case of *Myrothecium verrucaria* decreased.

Key words: micromycetes, total nitrogen, proteins, amino acids

INTRODUCTION

Microorganisms play an important role both in traditional and new technological processes. Recently in many countries the growing interest has been given to biosynthesis of enzymes (phenoloxidases, amylases, cellulases and hemicellulases) under solid-phase fermentation conditions. The traditional enrichment of fodder with proteins is also urgent under these conditions.

The advantage of solid-state fermentation against submerged one is its cheapness, more effective growth of particular fungi-producers, a possibility to do some operations under non-sterile conditions, an increased product yield because of a high concentration of a substrate, easy separation of the product, etc. The disadvantage is an insufficient standardization and control of the processes and a more difficult thermoregulation of a culture [1].

An urgent problem is the use of biotechnologies in industrial processing of the plant raw-material [2, 3]. All kinds of plant remnants possess high amounts of lignin, cellulose and hemicellulose, although the percentage of these components in various kinds of raw differs.

Thus, various kinds of straw differ greatly in the content of lignin and cellulose and in their digestibility.

Micromycetes play an important role in the biodegradation of various substrates rich in lignin and cellulose, and they could be used for biotechnological purposes. Attention of researchers is paid to microbiological degradation of lignin as a renewing raw-material [4–7].

Search of microbiological means for fodder enrichment with proteins is an urgent problem of cattle-breeding in Lithuania. Therefore, the aim of the work was to ascertain the ability of some micromycetes to utilize remnants of *Secale* and enrich them with proteins.

MATERIALS AND METHODS

The object of the study was rye (*Secale*) straw. Micromycetes – producers of phenoloxidases isolated from polymeric materials were used in experiments. The experiments were conducted with microorganisms – most successful degraders of lignin–cellulose complex in plant remnants – *Galactomyces geotrichum* (Butl. et Petersen) Redhead et Malloch and *Myrothecium verrucaria* (Alb. et Schweinitz) Ditmar ex Fries.

The plant remnants were moistened with a mineral medium (0.3 g NH_4NO_3 and 0.1 g KH_2PO_4 were added to 10 g of dry material). Micromycetes on the rye straw were cultivated 60 days at a temperature of 28 °C. Then the composition and content of total nitrogen, proteins and amino acids were analyzed.

The total nitrogen content in a product after cultivation of micromycetes for 20, 40 and 60 days was evaluated with an automatic Kontoflo analyzator. Mineralization of the plant raw material was performed by the Kjeldal method [8]. The ratio of a sample and mineralizing acid was 1:20. The amount of proteins was found by multiplication of the total nitrogen by 6.25.

The error of the analysis did not exceed 5%. The composition and content of amino acids in the biomass was analyzed with an automatic T339 (Czech Republic) analyzer of amino acids

RESULTS AND DISCUSSION

The obtained results showed that after 20 days of cultivation the straw was most significantly enriched with proteins by *Myrothecium verrucaria* (Fig. 1). Their content was 1.78 times higher than in control. During the further cultivation, after 30 days protein content decreased 1.1 and after 60 days 1.05 times in comparison with control.

The opposite results were obtained when *Galactomyces geotrichum* was cultivated (Fig. 2). After 20 days of cultivation the micromycetes enriched plant remnants with proteins by 7.08%. Their content was 1.1 times higher than in control. During the subsequent cultivation after 30 days, the amount of protein increased 1.49 times and the maximum content (up to 2.06 times higher in comparison with the control) was reached after 60 days of cultivation.

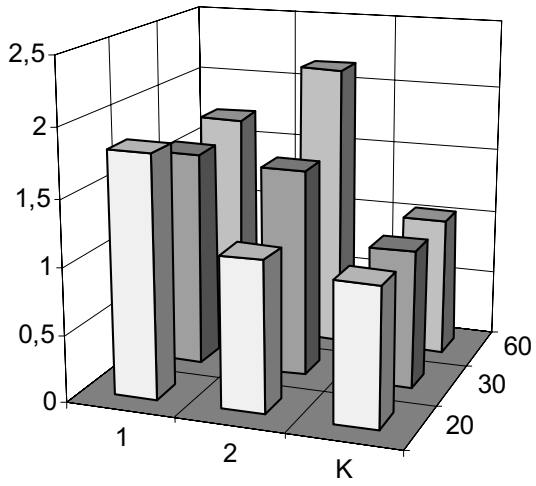

Fig. 1. Change of the total N in rye straw after 20, 30 and 60 days of cultivation of *Myrothecium verrucaria* (1) and *Galactomyces geotrichum* (2)

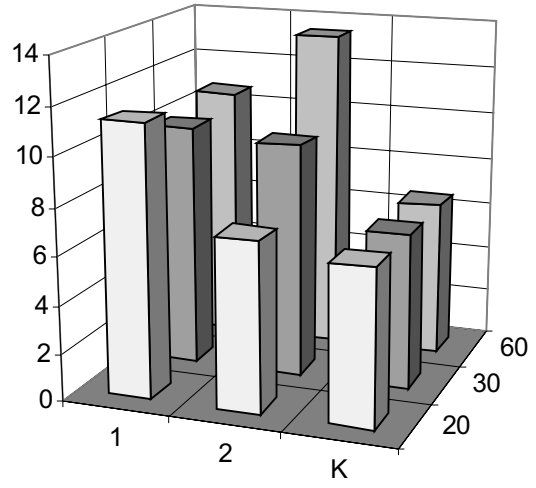

Fig. 2. Change of protein content in rye straw after 20, 30 and 60 days of cultivation of *Myrothecium verrucaria* (1) and *Galactomyces geotrichum* (2)

There were 6040 mg of amino acids/100g air-dried material detected in the control *Secale* straw (Table 1). After cultivation of *Myrothecium verrucaria* for 60 days, the total amount of amino acids increased 1.02 times when compared to the control.

The control straw contained 2250 mg of irreplaceable amino acids/100 g air-dried material. When micromycete *Myrothecium verrucaria* was grown for 60 days, the total amount of amino acids increased 1.05 times in comparison with the control. A more significant increase was noticed for valine, isoleucine and proline, and their amount was 1.12, 1.11, 1.02 times respectively higher when compared to the control.

When *Galactomyces geotrichum* was cultivated on rye straw for 60 days, the total amount of amino acids increased 1.25 times in comparison with the control and 1.22 times when compared with cultivation of *Myrothecium verrucaria* (Table 2).

The content of irreplaceable amino acids reached 3060 mg/100 g air-dried material. This increase 1.36 times exceeded the control. The main amino acids were glutaminic, asparagine, tyrosine and isoleucine (their increase was 1.4, 1.36, 1.67 and 1.76 times respectively higher in comparison with the control).

Table 1. **Composition and content of amino acids in rye straw after 60-day cultivation of *Myrothecium verrucaria* (mg/100 g air-dried material)**

Amino acids	Control	Cultivation with mineral additives
Lysine	300	310
Threonine	290	280
Cystine	30	40
Methionine	70	70
Valine	310	350
Isoleucine	340	380
Leucine	400	450
Tyrosine	360	370
Phenylalanine	470	480
Tryptophan	70	60
Histidine	250	260
Arginine	470	450
Asparaginic acid	550	550
Serine	320	300
Glutamic acid	700	710
Proline	470	480
Glycine	330	340
Alanine	310	320
Total	6040	6200

Table 2. **Composition and content of amino acids in rye straw after 60-day cultivation of *Galactomyces geotrichum* (mg/100 g air-dried material)**

Amino acids	Control	Cultivation with mineral additives
Lysine	300	410
Threonine	290	440
Cystine	30	50
Methionine	70	100
Valine	310	370
Isoleucine	340	600
Leucine	400	670
Tyrosine	360	170
Phenylalanine	470	370
Tryptophan	70	100
Histidine	250	390
Arginine	470	500
Asparaginic acid	550	750
Serine	320	410
Glutamic acid	700	980
Proline	470	490
Glycine	330	430
Alanine	310	360
Total	6040	7590

Thus, the obtained product contains all irreplaceable amino acids and has a high biological value.

Galactomyces geotrichum was found to be most successful in rye straw bioconversion.

References

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RUGIŲ ŠIAUDŲ BIOKONVERSIJA MIKROMICETAIS

S a n t r a u k a

Ištirta mikromicetų *Myrothecium verrucaria* ir *Galactomyces geotrichum* gebėjimas utilizuoti *Secale* šiaudų atliekas ir praturtinti jas baltyminėmis medžiagomis. Atlikti tyrimai parodė, kad perspektyvesnis mikromicetas rugių šiaudų biokonversijoje buvo *Galactomyces geotrichum*, kurį kultivuojant 60 parų ant šiaudų proteinų kiekis padidėjo 2,06, suminis aminorūgščių kiekis 1,25 karto, palyginti su kontrole. Baltyminių medžiagų kitimo dėsningumas priklausomai nuo *Galactomyces geotrichum* ir *Myrothecium verrucaria* kultivavimo trukmės buvo skirtingi. Kultivuojant ilgesnį laiką ant rugių šiaudų *Galactomyces geotrichum*, baltyminių medžiagų kiekis didėjo, o *Myrothecium verrucaria* – mažėjo.