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MAIZE HYBRIDS AS RAW MATERIAL FOR BIOETHANOL PRODUCTION

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Abstract

Last decades, the importance of alternative energy source has become more necessery due to the continous depletion of limited fossil fuel stock and their negative enviromental impact. Bioethanol produced from renewable biomass such as sugars, starch or lignocelluloses could be one of alternatives. The maize is one of the best renewable raw material for ethanol production, due to the high content of starch in grain. Ten maize hybrids of differnt genetic background created in Maize Research Institute Zemun Polje, were investigated for bioethanol production. The highest ethanol yield, 87.84% of theoretical content, as well as the highest bioethanol yield per arable land (5783,4 kg ha-1) had hybrid ZP434. Energy effectivnes of hybrids was from 8,35MJ(ZP341) to 9,47MJ (ZP434). Since that production of one liter of ethanol requires on average 2,5 kg of maize grain, it can be calculated that production of maize grain per hectare of arable land would be more profitable. Identification of maize hybrids with high potential for ethanol yield could significantly rise efficiency of ethanol production from maize.

Key words: *bioethanol, maize grain, starch, production*

Introduction

With the depletion of fossil fuel resources, as well as the increase in environmental pollution, interest in various agricultural crops as renewable and biodegradable feedstock’s for biofuel production are growing (Agarwal, 2007). Bioethanol is a liquid biofuel which can be produced from several different biomass feed stocks and conversion technologies. It can be made from many agricultural products and food wastes if they contain sugar (sugar cane, sugar beet, sweet sorghum and fruits), starch (maize, wheat, triticale, rice, potatoes, cassava, Jerusalem artichoke, sweet potatoes and barley), or cellulose (wood, straw and grasses), which can further be fermented and distilled into ethanol.

Maize, the dominant feedstock in the starch-to-ethanol industry worldwide, is composed of 60 to 70% of starch. There are two major traditional industrial processes for producing ethanol: wet and dry milling. In the dry-milling process, the grain is ground to a powder, which is then hydrolyzed and the sugar contained in the hydrolysate is converted to ethanol while the remaining flow containing fiber, oil, and protein is converted into a co-product known as distillers grains (DG) used as animal feed.

The use of ethanol as a fuel has a long history, starting in 1826 when Samuel Morey used it with the first American prototype of the internal combustion engine. At the beginning of 20th century, Henry Ford built a vehicle that could run either on gasoline or alcohol (Freudenberger, 2009). Nevertheless, ethanol failed to be used as a common fuel at that time because of the abundant and cheap supply of petroleum and natural gas. In Brazil, the commercial use of bioethanol produced from sugar cane, started back in 1925. The first bioethanol plant in the United States was built in the forties of the twentieth century to supply

the U.S. military with fuel. The disruption of oil supply from the Middle East in 1970s, however, re-boosted the production of ethanol (Bothast and Schleicher, 2005).
According to the World Association for Renewable Fuels (GRFA) in cooperation with Licht, world production of bioethanol is 23.4 billion gallons during 2013 (RFA 2013), Table 1. Brazil and the USA are the world leaders, which together accounted for about 60% of the world ethanol production exploiting sugarcane and maize respectively. In Europe, the production of bioethanol constantly increases, with 1371 million gallons during 2013.

Table 1. World fuel ethanol production by countries (mil gallons)

	2007	2008	2009	2010	2011	2012	2013
USA	6521.00	9309.00	10938.00	13298.00	13948.00	13300.00	13000.00
Brasil	5019.20	6472.20	6578.00	6921.54	5573.24	5577.00	6267.00
Europe	570.30	733.60	1040.00	1208.58	1167.64	1179.00	1371.00
China	486.00	501.90	542.00	541.55	554.76	555.00	696.00
Canada	211.30	237.70	291.00	356.63	462.30	449.00	523.00
Rest of world	315.30	359.40	914.00	984.61	698.51	752.00	1272.00
World	13123.10	17643.80	20303.00	23310.00	22404.09	21812.00	23429.00

Ethanol production process only uses energy from renewable energy sources. Hence no net carbon dioxide is added to the atmosphere, making ethanol an environmentally beneficial energy source. What bioethanol placed in front of other renewable energy sources at the moment is the fact that the entire existing oil infrastructure can be used for the distribution of biofuels without problems (or with minor modifications) that burns in petrol or diesel engines.
Fuel ethanol is used in several manners in internal combustion engines: as 5% to 25% anhydrous ethanol blends with gasoline (5% maximum in Europe and India, 10% in the United States and China, 20 to 25% mandatory blends in Brazil), as pure fuel (100% of hydrated ethanol) in dedicated vehicles, or up to 85% in FFVs (Gnansounou, 2009). When anhydrous bioethanol is blended with gasoline in small proportion (up to 15%), the influence of the lower heating value has no significant effect. For higher blend levels, the fuel economy is reduced compared to that with conventional gasoline.
Current production of bioethanol in Serbia is based on molasses (50%) and cereals (50%) (Mojovic *et al.*, 2012). The average annual maize yield is approximately 40% higher than the calculated domestic needs in last few years This means that there is enough maize for other purposes besides the food, therefore significant amounts can be used for the bioethanol production.
The feedstock for bioethanol production is currently based mostly on agricultural crops, which can be devoted to both food and ethanol markets or dedicated solely to ethanol. In case of a high world production of bioethanol, the correlation between food and ethanol markets may generate a high volatility of agricultural crops with regard to fluctuations in energy prices.
The aim of our study was to analyze suitability of selected ZP maize hybrids for the bioethanol production.

Material and methods

Ten maize hybrids of different genetic background and maturity produced in Maize Research Institute Zemun Polje: ZP341 (FAO300), ZP 434 (FAO400), ZP 505, ZP548 (FAO500), ZP 600, ZP606, ZP666 (FAO 600), ZP704wx, ZP747 (FAO 700), ZP808, ZP877 (FAO 800) were investigated for bioethanol production.
Before ethanol fermentation two-step enzymatic hydrolysis of maize meal by commercially

available α-amylase (from *Bacillus licheniformis*, Termamyl® SC, Novozymes, Denmark) and glucoamylase (SAN Extra® L, Novozymes, Denmark, from *Aspergillus niger*) was done (Nikolić *et al*., 2009). Starch hydrolysates were subjected to fermentation by *Saccharomyces cerevisiae* (collection of BIB-TMF, Belgrade) yeast. The ethanol concentration was determined based on the density of alcohol distillate at 20ºC and expressed in weight % (w/w), (Official Methods, 2000). The starch content was determined by Ewers polarimetric method (ISO, 1997), protein content by Kjeldahl metod and oil content by Soxhlet method (Oficial method of analysis, 2000).
The average bioethanol yield per arable area for each hybrid was calculated on the basis of percentage of theoretical bioethanol yield obtained in fermentation and average yield potential of the hybrid per arable area. The calculation of the cost of production of maize per hectare of arable land based on the prices of certain operations, the cost of fuel and the price of maize seeds, fertilizers and herbicides used. In the calculation of production costs of bioethanol used data on costs of production plants in the United States (Mousdale, 2008). The budget is approximated to the ideal case that Serbia has a production facility which is currently operational.

Results and discussion

Characteristics regarding chemical composition of the hybrids are shown in Table 2. Starch, protein and oil content differ between analyzed hybrids. Hybrid ZP 877 had the highest starch content of 74.68% and ZP341 the lowest, only 70.40%. Hybrid ZP877 has the highest soluble carbohydrate content and hybrid ZP434 the lowest one. All hybrids containing proportion of starch in the grain, more than 70%, which is one of the main prerequisites for the achievement of high yields of bioethanol in the process of dual-enzymatic hydrolysis and fermentation. According to Gulati *et al.* (1996) from one kg maize with the 15.5% moisture, containing an average of 0.61kg starch and 0.022kg of carbohydrates, theoretically 0.50l of ethanol could produce. In this case, 0.44l of ethanol originating from starch, 0.14l from soluble carbohydrates, 0.034l from cellulose and 0.015l from hemicelluloses. This means that soluble carbohydrates found in maize affect the yield of bioethanol (approximately 2.8% of the total yield). Protein content ranged from 8.86% (ZP808) to 10.30% (ZP704wx). Oil content ranged from 5.06 (ZP600) to 7.18% (ZP747).

Table 2. Chemical composition of ten maize hybrids

Genotype	Dry matter content %	Moisture content %	Starch content %	Soluble carbohydrates content %	Protein content %	Oil content, %
ZP 341	88.04	11.96	70.40	0.20	9.75	6.28
ZP 434	88.44	11.56	72.04	0.10	10.15	6.02
ZP 505	88.86	11.14	73.38	0.40	9.88	6.38
ZP 548	90.19	9.81	72.04	0.40	9.19	6.08
ZP 600	86.78	13.22	74.42	0.35	10.19	5.06
ZP 606	87.26	12.74	73.16	0.25	10.22	5.45
ZP 666	87.90	12.10	74.26	0.16	9.42	5.55
ZP 704wx	89.13	10.87	74.13	0.37	10.30	5.71
ZP 747	86.92	13.08	74.08	0.21	9.31	7.18
ZP 808	88.12	11.88	74.55	0.16	8.86	5.24
ZP 877	88.52	11.48	74.68	0.41	9.77	5.26

Values of the parameters important for bioethanol production determined after 48h of fermentation of maize flour hydrolysates of investigated hybrids are presented in Table 3.

Table 3. Bioethanol content, percentage of the theoretical yield, volumetric productivity and energy value in SSF

Genotype	Bioethanol content % (w/w)	% of theoretical yield %	Volumetric productivity (gl^{-1} h^{-1})	Energy value MJ after 24h	Energy value MJ, after 48h
ZP 341	7.97	79,94	1.66	6.27	8,35
ZP 434	8.96	87.84	1.87	6.39	9.43
ZP 505	8.01	77.02	1.67	5.66	8.46
ZP 548	8.83	86.57	1.84	6.24	9.47
ZP 600	8.33	79.03	1.74	5.59	8.60
ZP 606	8.46	81.66	1.76	5.50	8.78
ZP 666	8.61	81.84	1.79	6.07	9.00
ZP 704wx	8.07	76.86	1.68	5.50	8.55
ZP 747	8.75	83.41	1.82	5.45	9.05
ZP 808	8.20	80.68	1.78	5.60	8.93
ZP 877	8.41	79.49	1.75	5.84	8.85

The highest bioethanol content after 48h of fermentation had hybrids ZP434, ZP 548, ZP 747, ZP 666, ZP 808, ZP 606 (8.96%, 8.83%, 8.75%, 8.61% 8.52% and 8.46, respectively). The similar ethanol yield (approximately 79%) was detected in the hybrids ZP 877 and ZP 341. These two hybrids significantly differ in starch content. The relationship between grain starch and ethanol yield is not complete understood. The highest starch producing hybrid was not the highest ethanol producer. Volumetric productivity varied from 1.66 gl^{-1} h^{-1} (ZP341) to 1.87 gl^{-1} h^{-1} (ZP434). The hybrid ZP434 had the highest bioethanol content, volumetric productivity, a percentage of theoretical ethanol yield and bioethanol yield. If compared ethanol yield of ZP 434 with ethanol yield of another hybrids (Drinić *et al.*, 2011; Semečenko 2013), it can be concluded that ZP434 is very suitable for bioethanol production. The lowest yield detected in ZP704wx can be attributed to the high percentage of hard endosperm. The hybrid ZP434 has the highest amount of soft endosperm. Semečenko *et al.* (2013) found a very good association between bioethanol yield and amount of soft endosperm. Srcichuwong *et al.* (2010) concluded that the hybrid with the highest starch content and the lowest content of lipids and proteins achieve the best yield of ethanol.

During the production of bioethanol, starch is removed from the grain and converted to alcohol and carbon dioxide. As a result of starch removal, the concentration of the remaining nutrients in the grain increases approximately three-fold. Valuable by-product dried distillers grains with soluble (DDGS) contains high percentages of protein, fiber and lipid can be used as a substitute for traditional feedstuff. Energetic value of bioethanol, which can be obtained from one kg of maize, expressed in MJ is calculated. After 24 hours of fermentation the most effective hybrid was ZP 434 (6.39 MJ) and the lowest one ZP737 (5.45 MJ). After 48 hours of fermentation the most effective hybrid was ZP 548 (9.47 MJ), following by ZP 434 (9.43 MJ) and the lowest one ZP737 (5.45 MJ).

Beside the fermentation efficiency, an important issue in the maize evaluation for bioethanol production is land requirement, e.g. bioethanol yield per land area. Hybrid ZP434 gives the highest yield per area, 5783.4tha^{-1}, followed by ZP704wx (4646.5tha^{-1}). The average bioethanol yield per arable area for each hybrid was calculated on the basis of percentage of theoretical bioethanol yield obtained in fermentation and average yield potential of the hybrid per arable area. It is also important to note that the best bioethanol producer, hybrid ZP 434, can be successfully cultivated on a poor land quality.

According to Mousdale (2008) 1GJ production of ethanol from starch or sugar feedstock in the European Union and the United States costs between € 16.2 to 23 or from € 0.29 to 0.41l^{-}

[1]. This means that 17.82 to 17.90 MJ is consumed to produce one liter of ethanol or about 5.5 MJ less energy than the energy value of one liter of ethanol. One liter of ethanol achieves higher profit than selling one kilogram of maize grain, but the production of one liter of ethanol requires on the average 2.5 kg of maize grain so the production of maize grain per hectare of arable land would be more profitable.

Conclusion

Identification of maize hybrids with higher bioethanol yield potential could significantly increase production efficiency of this biofuel from maize. The hybrid ZP434 is the most promising ethanol producer. High yield potential per hectare makes it the best candidate for the commercial bioethanol production. This hybrid is also extremely tolerant to drought and stalk lodging.

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