EFFECT OF POTASSIUM FERTILIZATION ON THE YIELD, QUALITY AND POTENTIAL ETHANOL YIELD OF FIVE CORN HYBRIDS

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Abstract
We have also determined the potential ethanol yield by fermentation in some samples. The small plot experiments were set up at the same site near Debrecen, on chernozem soil, in 2007 and 2008. We analysed the effect of the same five mineral fertilizer doses on the same five hybrids in four repetitions each year.
Applied fertilizer types and doses were the following: 1.: N and P as ammonium nitrate and superphosphate (100 kg/ha N, 80 kg/ha P_2O_5) without K, 2. and 3.: the same NP with K as KCl (100, and 200 kg/ha K_2O, respectively) 4. and 5.: the same NP with K as KCl+$MgSO_4$ (100, and 200 kg/ha K_2O, respectively).
The effect of Potassium treatments on the yield of corn can be shown in both years, with significant differences between the different hybrids in the yield. The PR36K67 and PR37D25 hybrids resulted in the highest yields in both years.
The treatments did not have a significant effect on the starch, protein and oil contents, however, and the potential ethanol yields measured by fermentation also remained almost the same. The potential ethanol yield pro hectare, calculated from the yield of hybrids and the volume of ethanol through fermentation, was determined by the yield of hybrids.
We found strong significant correlation between the starch content and the ethanol yield estimated by the weight loss, as well as medium strength correlation between the starch content and the ethanol yield determined by HPLC. This effect is influenced by the crop year, and we found different equations and regression coefficients during the evaluation of the average of the two examined year. Our experiences call attention to the uncertainty factors and problems of estimating connection equations of many years, and the importance of their yearly revision and actualisation.

Introduction
The utilization of renewable resources, as the transformation of agricultural products to biofuels, is emphasized again and again by the decrease of mineral oils. European Union plans to cover the 5.75% of total consumption by usage of renewable sources. Bioethanol production from corn is one alternative solution for this.
One of the highest corn production countries of the European Union is Hungary. The decrease of the national livestock was continuously observable on the stored corn stocks in the last decade. It resulted in the first years of the Hungarian introduction of intervention system that the amount offered and stored corn were 4.7 million tonnes by the end of 2006 (Pallagi, 2007), and the overproduction caused strong decrease in market price. One chance is the industrial usage of corn against overproduction and preservation of the balance of market, and the production of glucose-fructose syrup and ethanol are the most suitable ways, as Patzek (2006) found during the analysis of the circumstances in the

United States of America. Because of this, Marko et al. (2009) found that the economy of feed and food raw material production has structural changes by the increase of production volume. Those, who reason against the cereal based biofuel production, think that the area of fields, suitable for food raw material production, will decrease because of the cereal-energy production.
Efficiency of bioethanol production is strongly effected by the properties of breed hybrids and applied agronomy, besides the industrial technology. It is well known, that general terms of good ethanol yield are the high starch and relatively low protein content of grains (Voca et al., 2007). Many references call attention to examine the extractable starch content instead of total starch content with a view to ethanol yield (Singh et al., 2002). As far as we know, there is no generally accepted and standardized method of the determination of ethanol yield, starch content is considered, besides the result of fermentation experiment.
We made a potassium fertilization experiment with different hybrids and applied different potassium forms and doses. We have examined the yield, the chemical composition of grains and we have determined the ethanol yield in a laboratory scale fermentation experiment.

Material and methods
Polifactorial experiment was set up in the region of Debrecen on calcareous chernozem soil on five different corn hybrids:

1. PR38B12 (FAO 310)
2. PR37D25 (FAO 330)
3. KWS 353 (FAO 350)
4. DKC 5211 (FAO 460)
5. PR36K67 (FAO 490)
6.

Five levels of mineral fertilization were applied (Table 1). Nitrogen was applied as ammonium nitrate, Phosphorus as superphosphate. The effect of potassium was examined partly as potassium chloride, partly as Korn-Kali. The Korn-Kali contained 40% of K_2O as KCl and 6% MgO as ($MgSO_4.7H_2O$).

Table 1 – The mineral fertilizer doses applied in the experiment

Treatments	N, kg/ha	P_2O_5, kg/ha	K_2O, kg/ha	MgO, kg/ha
1.	120	80	-	-
2.	120	80	100 (KCl)	-
3.	120	80	100 (Korn-Kali)	15
4.	120	80	200 (KCl)	-
5.	120	80	200 (Korn-Kali)	30

Experiment was set up in random block design with four repetitions, same treatments and hybrids both in 2007 and 2008.
Type and properties of soil: moderately leached chernozem, loamy clay, formed on loess. pH($CaCl_2$): 5.6, Humus 2.9%, AL-P_2O_5 314 mg/kg, AL-K_2O 355 mg/kg, AL-Ca 4142 mg/kg, AL-Mg 422 mg/kg.

Weather conditions: Weather conditions were favourable for corn production in 2008. The precipitation of April, June and July was lower than the 30-years-average in 2007, while the temperature was higher than it, except in September.
The precipitation of the vegetation period exceeded the 30-years-average in 2008. The temperature conditions, what were similar to the 30-years-average, were favourable for fertilization and grain filling.
Yield harvesting was made by Sampy harvester by plots. We have determined the moisture content of grains and we have calculated to 86% dry matter content the yield data.
We have determined the starch, protein and fat content of samples of three selected treatments, 120-80-0, 120-80-200 (KCl), 120-80-200 (Korn-Kali) from two field repeats in two laboratory repetitions by the directives of Hungarian Standards (moisture content by MSZ 6367-3:1983, protein content by MSZ 6830-4:1981, fat content by MSZ 6830-6:1984 and starch content by MSZ 6830-18:1988). Results presented in the percentage of dry matter.

Besides, we have determined the ethanol yield by fermentation experiment. Corn samples were milled by Retsch SR2 laboratory grinder using 0.4 mm sieve. 160 g of corn-water mash with 27% dry matter content was fermented. Liquofication was made by Liquozyme amylase enzyme (Novozymes) on 83°C.
Saccharification was made by Spirizyme glucoamylase enzyme (Novozymes) simultaneously by the fermentation. Fermentation was performed by Ethanol Red Yeast (Fermentis) complemented by AYF1177 yeast nutrient (Ethanol Technology), urea, 50069 alcalase (Novozymes) and LactoStab (BetaTec Hopfenprodukte GmbH) for 72 hours. Final ethanol concentrations of mashes were determined by HPLC method by UV detector and estimated by weight loss, supposed that all weight losses are the result of microbial ethanol fermentation.
We made analysis of variance to evaluate the effect of mineral fertilization on the yield and quality parameters and correlation analysis to reveal the connection between starch content and ethanol yield.

Results and discussion

The same tendencies are observable in the formation of the yield of hybrids. PR36K67 (FAO 490) and PR37D25 (FAO 330) hybrids showed the highest yields in both years and the yield increasing effect of potassium is proved. The higher dose of KCl was effective in the case of all hybrids. The application of Korn-Kali increased the yield only in the case of 100 kg dose, the higher dose caused decrease in yield in the most of parcels, so its effect was smaller than of KCl on this soil, relatively well provided with Magnesium. Yield data of the experiments are shown in Table 2.

Table 2 – Yield of the examined corn samples (t/ha, on 86% dry matter base), 2007-2008

Hybrids	Treatment	2007	2008
PR38B12 (FAO 310)	120-80-0	9.03	10.36
	120-80-100 (KCl)	10.60	13.16
	120-80-200 (KCl)	11.63	14.02
	120-80-100 (Korn-Kali)	9.72	13.06
	120-80-200 (Korn-Kali)	11.22	12.14
Mean		**10.44**	**12.55**
LSD 5%		**0.52**	**0.80**
F value		**39.55*** **	**28.93*** **
PR37D25 (FAO 330)	120-80-0	11.77	10.88
	120-80-100 (KCl)	13.25	14.18
	120-80-200 (KCl)	13.12	15.12
	120-80-100 (Korn-Kali)	13.02	13.80
	120-80-200 (Korn-Kali)	12.29	12.93
Mean		**12.69**	**13.38**
LSD 5%		**1.39**	**0.71**
F value		**1.98**	**49.26*** **
KWS 353 (FAO 350)	120-80-0	9.80	9.07
	120-80-100 (KCl)	10.55	10.47
	120-80-200 (KCl)	11.65	10.74
	120-80-100 (Korn-Kali)	10.29	10.82
	120-80-200 (Korn-Kali)	11.20	9.84
Mean		**10.70**	**10.19**
LSD 5%		**0.90**	**0.53**
F value		**6.21** **	**18.21*** **
DKC 5211 (FAO 460)	120-80-0	8.59	8.60
	120-80-100 (KCl)	9.96	10.93
	120-80-200 (KCl)	10.53	11.37
	120-80-100 (Korn-Kali)	8.94	10.15
	120-80-200 (Korn-Kali)	8.74	8.70
Mean		**9.35**	**9.95**
LSD 5%		**0.83**	**0.70**
F value		**9.81** **	**31.02*** **
PR36K67 (FAO 490)	120-80-0	12.78	13.35
	120-80-100 (KCl)	14.32	15.94
	120-80-200 (KCl)	14.62	16.37
	120-80-100 (Korn-Kali)	13.39	17.15
	120-80-200 (Korn-Kali)	13.30	14.74
Mean		**13.68**	**15.51**
LSD 5%		**1.29**	**0.74**
F value		**3.32**	**38.39*** **

Starch, protein and fat content of corn samples are shown in Table 3.

Table 3 – Starch, protein and fat content of examined corn samples (%,, on dry matter base), 2007-2008

Hybrids and treatments	Starch content		Protein content		Fat content	
	2007	2008	2007	2008	2007	2008
PR38B12 (FAO 310)						
120-80-0	72.19	68.51	11.65	9.52	4.93	4.94
120-80-200 (KCl)	72.70	71.19	11.84	9.47	5.04	5.02
120-80-200 (Korn-Kali)	72.52	65.30	11.77	9.93	5.15	4.97
Mean	**72.47**	**68.33**	**11.75**	**9.64**	**5.04**	**4.97**
LSD 5%	**1.24**	**0.74**	**0.96**	**0.66**	**0.02**	**0.51**
F value	**0.21**	**76.42****	**0.05**	**0.68**	**121****	**0.03**
PR37D25 (FAO 330)						
120-80-0	73.15	68.95	10.72	9.21	4.99	4.51
120-80-200 (KCl)	73.31	69.73	10.91	9.38	4.80	4.37
120-80-200 (Korn-Kali)	73.82	70.57	10.64	9.29	4.83	4.38
Mean	**73.43**	**69.75**	**10.76**	**9.29**	**4.87**	**4.42**
LSD 5%	**0.62**	**0.98**	**0.46**	**0.40**	**0.04**	**0.40**
F value	**1.52**	**3.27**	**0.44**	**0.24**	**24.14****	**0.18**
KWS 353 (FAO 350)						
120-80-0	72.32	68.11	11.70	10.42	5.22	4.73
120-80-200 (KCl)	71.73	69.00	12.35	10.53	5.25	4.86
120-80-200 (Korn-Kali)	71.31	68.45	12.33	10.51	5.10	4.37
Mean	**71.79**	**68.52**	**12.13**	**10.48**	**5.19**	**4.65**
LSD 5%	**0.77**	**1.36**	**0.70**	**0.47**	**0.17**	**0.70**
F value	**2.09**	**0.51**	**1.32**	**0.08**	**0.98**	**0.62**
DKC 5211 (FAO 460)						
120-80-0	72.74	68.98	11.36	9.91	5.12	4.58
120-80-200 (KCl)	72.30	69.26	11.31	9.90	5.02	4.68
120-80-200 (Korn-Kali)	72.84	69.23	11.20	9.56	4.94	4.84
Mean	**72.63**	**69.16**	**11.29**	**9.79**	**5.03**	**4.70**
LSD 5%	**0.18**	**0.40**	**0.35**	**0.39**	**0.09**	**0.14**
F value	**12.18***	**0.69**	**0.25**	**1.25**	**4.28**	**3.72**
PR36K67 (FAO 490)						
120-80-0	74.04	68.69	9.85	9.05	4.86	5.35
120-80-200 (KCl)	73.48	72.04	10.64	8.93	4.89	4.90
120-80-200 (Korn-Kali)	73.19	68.75	10.33	9.11	4.89	4.88
Mean	**73.57**	**69.83**	**10.27**	**9.03**	**4.88**	**5.04**
LSD 5%	**0.68**	**3.16**	**0.08****	**0.44**	**0.11**	**0.22**
F value	**1.92**	**1.75**	**111.37**	**0.44**	**0.09**	**6.93**

Mineral fertilizer treatments did not cause significant differences in starch, protein and oil contents.
Weather conditions caused stronger differences. The higher yield was accompanied by higher starch and lower protein content in the second year. Effects of genotype also can be seen, there were differences in the chemical compositions of different hybrids. The highest starch contents were shown by PR36K67 (FAO 490) and PR37D25 (FAO 330) hybrids in both years. The higher starch contents were accompanied by lower protein contents. The effect of the examined parameters on the fat content is negligible.
We have estimated by weight loss and measured with HPLC equipment the potential ethanol yield in g/100 g dry matter base in fermentation experiment. The analysis by HPLC requires expensive equipment and materials and costly, but more accurate. On the other hand, the estimation method is less reliable, but its advantages are rapidity and cheapness, so it is simply to use in industry and production practice, and in the valuation of hybrids, as in our experiment. Estimation usually reports higher yields, since the theoretical base of estimation is that assumption that the weight losses, measured during fermentation, come completely by the vaporizing CO_2 from the alcoholic fermentation of carbohydrates. As it is well known, other fermentation processes are running beside this.
Besides potential ethanol yield, we have calculated the ethanol yield pro hectare as well (Table 4).
Similarly to the parameters of chemical composition, we did not find nor statistically proved nor tendency-like changes in the potential ethanol yield, due to effect of mineral fertilization. Year effect is visible in the results; values in 2008 are slightly lower, probably in connection with lower starch content. Performances of hybrids did not differ significantly.
However, significant differences in ethanol yield pro hectare calculated by yield and potential ethanol yield is visible, and it is determined by different yields. Best results were shown by PR36K67 (FAO 490) and PR37D25 (FAO 330) hybrids, as in the case of yield.
We made correlation analysis between starch content of grains and potential ethanol yield, determined by both ways (Figure 1 and 2). The strength of connections formed similarly in the different crop years, but when we made the analysis on the results of both years we experiences that the strength of connection decreases in the case of ethanol yield estimated by weight loss, but increases in the case of the ethanol yield measured by HPLC, due to effect of crop year. The correlation analyses confirm that the connection can be modified in different years by other factors.

Conclusions

We found that the effect of Potassium treatments on the yield of corn can be shown in both years, with significant differences between the different hybrids in the yield. The PR36K67 and PR37D25 hybrids resulted in the highest yields in both years.
The treatments did not have a significant effect on the starch, protein and oil contents, and those hybrids resulted the highest starch content what has the lowest protein content. The potential ethanol yields measured by fermentation also remained almost the same, so Potassium fertilization had no proved effect on this technological parameter. The potential ethanol yield pro hectare, calculated from the yield of hybrids and the volume of ethanol through fermentation, was determined by the yield of hybrids.
We found strong significant correlation between the starch content and the ethanol yield estimated by the weight loss, as well as medium strength correlation between the starch content and the ethanol yield determined by HPLC. This effect is influenced by the crop year, and we found different equations and regression coefficients during the evaluation of the average of the two examined year. Our experiences call attention to the uncertainty factors and problems of estimating connection equations of many years, and the importance of their yearly revision and actualisation.

Table 3 – Potential ethanol yield and ethanol yield pro hectare of examined corn samples (%,, on dry matter base), 2007-2008

Hybrids and treatments	Potential ethanol yield by weight loss, g/100g		Potential ethanol yield by HPLC, g/100g		Ethanol yield pro hectare, by weight loss, l/ha		Ethanol yield pro hectare, by HPLC, l/ha	
	2007	2008	2007	2008	2007	2008	2007	2008
PR38B12 (FAO 310)								
120-80-0	34.92	33.29	30.47	28.64	3991.49	4365.95	3482.84	3755.83
120-80-200 (KCl)	35.12	35.61	30.80	30.70	5170.20	6318.76	4534.23	5448.28
120-80-200 (Korn-Kali)	35.03	32.70	30.47	28.55	4975.15	5025.04	4327.51	4387.30
Mean	**35.02**	**33.87**	**30.58**	**29.29**	**4627.96**	**5236.58**	**4041.21**	**4530.11**
LSD 5%	**0.59**	**0.70**	**1.04**	**0.32**				
F value	**0.13**	**22.93****	**0.16**	**67.00****				
PR37D25 (FAO 330)								
120-80-0	35.39	34.34	30.42	28.70	5272.66	4728.67	4532.19	3952.61
120-80-200 (KCl)	35.25	34.89	30.57	28.99	5854.18	6677.20	5076.94	5548.47
120-80-200 (Korn-Kali)	35.44	35.55	31.19	29.78	5513.39	5818.09	4852.22	4874.12
Mean	**35.36**	**34.92**	**30.73**	**29.16**	**5679.98**	**5741.32**	**4936.25**	**4792.06**
LSD 5%	**0.43**	**0.82**	**0.43**	**0.75**				
F value	**0.26**	**2.60**	**4.23**	**2.58**				
KWS 353 (FAO 350)								
120-80-0	34.99	33.96	30.47	28.83	4340.53	3898.66	3779.82	3309.98
120-80-200 (KCl)	34.57	34.23	30.03	29.88	5097.98	4653.55	4428.47	4062.17
120-80-200 (Korn-Kali)	34.40	34.31	30.12	28.98	4876.96	4273.55	4270.18	3609.66
Mean	**34.66**	**34.17**	**30.20**	**29.23**	**4694.46**	**4275.25**	**4090.38**	**3660.50**
LSD 5%	**0.28**	**0.84**	**0.20**	**1.02**				
F value	**5.47**	**0.23**	**6.56**	**1.46**				
DKC 5211 (FAO 460)								
120-80-0	35.07	34.46	30.73	29.32	3813.31	3751.34	3341.40	3191.80
120-80-200 (KCl)	34.81	34.41	29.67	29.39	4639.86	4952.43	3954.75	4229.93
120-80-200 (Korn-Kali)	35.01	34.33	30.19	29.84	3873.26	3780.37	3340.01	3286.18
Mean	**34.97**	**34.40**	**30.20**	**29.51**	**4138.85**	**4161.38**	**3574.30**	**3569.05**
LSD 5%	**0.28**	**0.48**	**0.31**	**0.26**				
F value	**1.09**	**0.09**	**13.52**	**5.62**				
PR36K67 (FAO 490)								
120-80-0	35.61	34.36	31.09	28.76	5760.71	5806.83	5029.50	4860.08
120-80-200 (KCl)	35.24	35.89	31.03	31.15	6521.63	7435.92	5742.51	6454.75
120-80-200 (Korn-Kali)	35.15	34.26	31.09	29.56	5917.66	6391.84	5234.14	5515.37
Mean	**35.34**	**34.84**	**31.07**	**29.82**	**6119.64**	**6544.86**	**5380.22**	**5610.13**
LSD 5%	**0.27**	**1.55**	**0.98**	**1.71**				
F value	**3.69**	**1.65**	**0.01**	**2.11**				

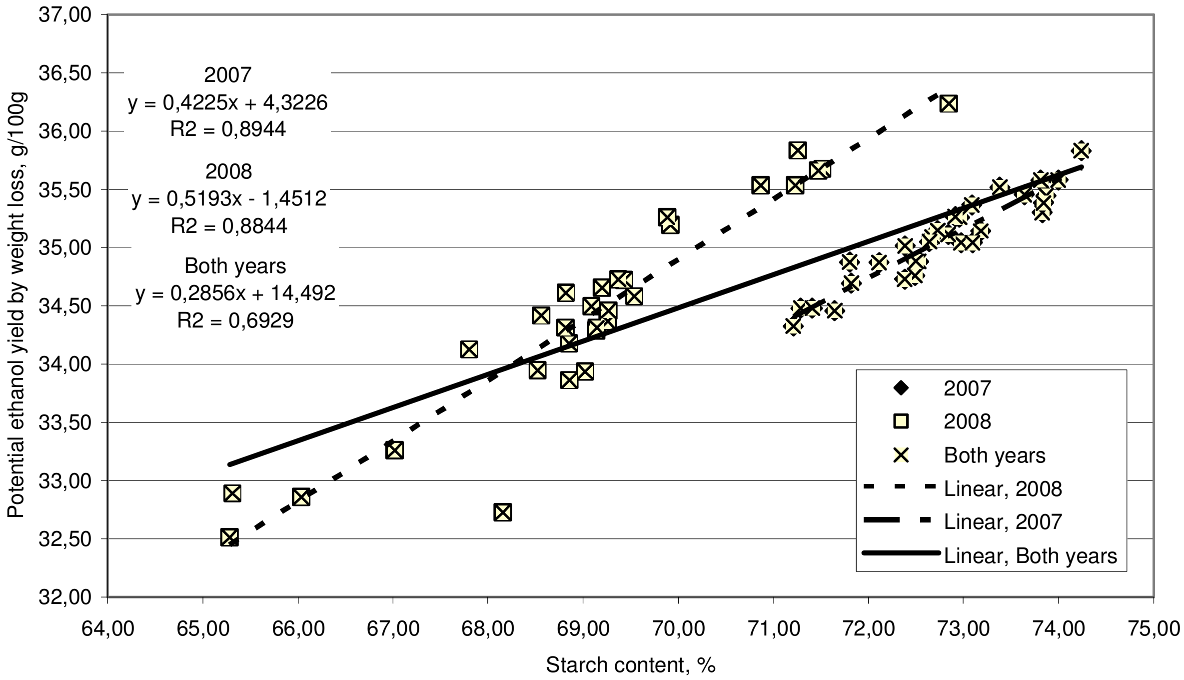

Figure 1 – Connection between starch content and potential ethanol yield, estimated by weight loss

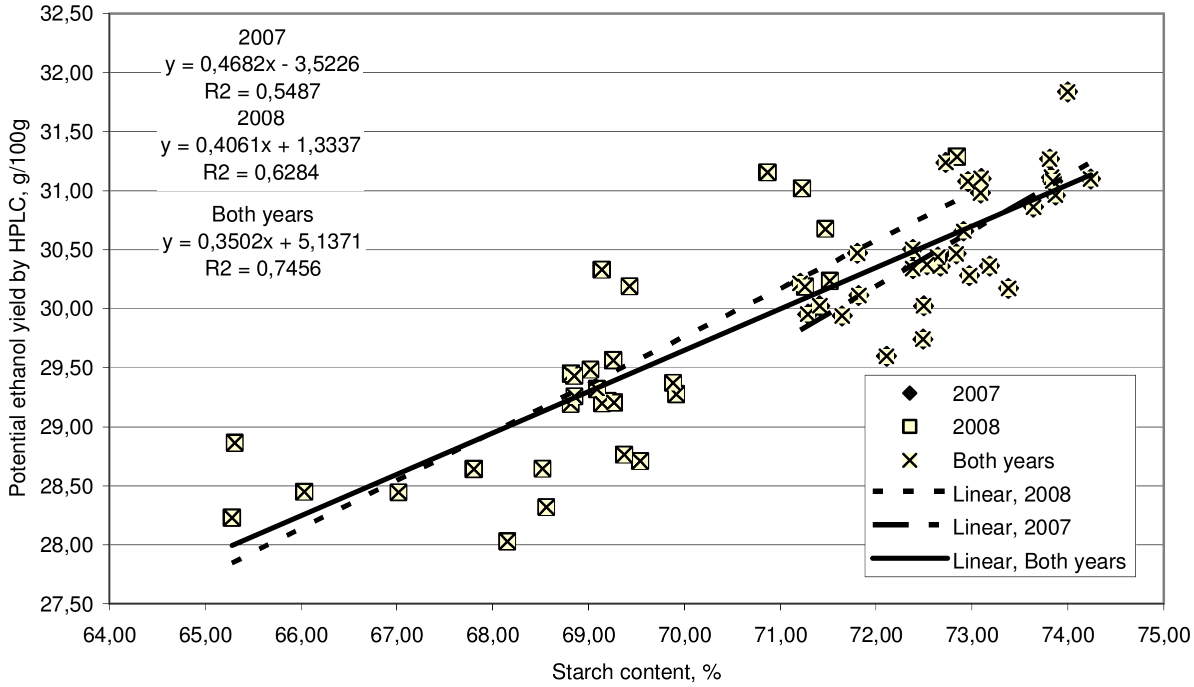

Figure 2 – Connection between starch content and potential ethanol yield, measured by weight HPLC

References

Marko, Popp, Somogyi, 2009. Sustainable land use for food, feed and biofuels, Cereal Research Communications, 37. Suppl, 643-646

Pallagi, 2007. AZ agrártámogatási rendszer aktuális kérdései, gabonaintervenció, A „Tárolás, elsődleges feldolgozás a növényi termék-előállításban" címmel 2007. február 8-án rendezett Interaktív Szaktanácsadási Konferencia szerkesztett anyaga. Debrecen. Szerk.: Győri Z. 7-12. ISSN: 1588-9665.

Patzek, 2006. Thermodynamics of the Corn-Ethanol Biofuel Cycle, Critical Reviews in Plant Sciences, 23(6):519-567

Singh, Paulsen,Tian, Yao, 2002. Site-specific study of corn protein, oil, and extractable starch variability using NIT spectroscopy. ASAE Paper No. 02-1111

Voća, Krička, Janušić, Matin, 2007. Bioethanol production from corn kernel grown with different cropping intensities. Cereal Research Communications, Vol. 35, No. 2 pp 1309-1312