### **BREEDING WHEAT FOR ORGANIC CROPPING SYSTEM**

### OPLEMENJIVANJE PŠENICE ZA EKOLOŠKU POLJOPRIVREDU

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"The next technological revolution will be led by eco-innovation." Timo Makela, Director DG Environment

### ABSTRACT

The breeding methods for organic and conventional production may overlap in the early generations, when selection focuses on an ideotype for organic production. But in later generations, the environmental variation must be accounted for during selection. Free access to genetically diverse pool is a prerequisite for combining desired parental characteristics.

At Krizevci, ideotype building for organically grown wheat started in the 1990's. In addition to high tillering and stress tolerance, our breeding goals included premium grain quality and resistance to diseases and insects. Of course, grain yield was considered in all cases. Both pedigree and recurrent selection were applied as in conventional wheat breeding. Cv. Divana -- the first Croatian premium grain quality wheat suitable for organic cropping systems -- was registered in 1995. The next step was introducing resistance to prevalent diseases, and the last step was introducing resistance to leaf beetle (*Oulema melanopus*) – an emerging threat to the future of wheat cultivation in the country. It should be noted that some of the listed ideotype characteristics are negatively correlated with others. Therefore, improvement in some pairs of characters, for instance high yield and high quality, proceeds more slowly than it would for either trait alone. Following a historical approach to development of scientific thought and achievements, we review conclusive experimental evidence that organic agriculture can feed the world.

Key words: wheat, breeding, thought development, ideotype, bioethics and organic cropping.

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### SAŽETAK

Vodeći računa o ideotipu biljke za ekološki uzgoj, kod oplemenjivanja pšenice u samom početku oplemenjivačkog procesa metode oplemenjivanja za konvencionalni i ekološki uzgoj mogu se podudarati. No u kasnijim generacijama tijekom selekcije treba primijeniti različite okolišne uvjete. Da bi mogli kombinirati željene roditeljske karakteristike, neophodan je slobodan dostup do genetski divergentnog materijala. U Križevcima je stvaranje ideotipa za ekološku poljoprivredu započeto devedesetih godina. Osim snažnog busanja i toleriranja stresnih uvjeta, ovdje je uključena i vrhunska pekarska kvaliteta u kombinaciji s otpornošću na biljne bolesti i štetnika. Primijenjene metode bile su pedigre i rekurentna selekcija.

Prva sorta vrhunske pekarske kakvoće, prikladna za ekološki uzgoj bila je Divana (1995.). Sljedeći korak bio je unos otpornosti na prevladavajuće bolesti pšenice, a potom otpornosti na lisni balac (*Oulema melanopus*), štetnik koji će u bliskoj budućnosti praviti dosta neprilika. Treba imati na umu da su neke od nabrojenih osobina u međusobno negativnom odnosu, npr. visoka rodnost i dobra pekarska kakvoća, pa će njihovo poboljšanje biti postepeno.

Opisan je razvoj ideje oplemenjivanja tijekom gotovo pola stoljeća: od induciranih mutacija, preko hibridne pšenice, konvencionalnog oplemenjivanja do oplemenjivanja za održivu ekološku poljoprivredu, te je na kraju rezultatima pokusa dat potvrdan odgovor na pitanje: Može li ekološka poljoprivreda hraniti svijet?

Ključne riječi: pšenica, oplemenjivanje, razvoj ideje, ideotip biljke, bioetika i ekološki uzgoj

# SHORT OVERVIEW OF THE THOUGHT DEVELOPMENT ABOUT ORGANIC AGRICULTURE

The following is a historical overview of how scientific thought has developed, parallelling the development of one scientific career through half a century.

**Induced mutation** - In 1960, as a student of agronomy a senior author of this report was lucky enough to start his career as a technical coworker of

academician Aloiz Tavcar. At that time Tavcar was the most famous plant geneticist in the former Yugoslavia. The scientific project was entitled "Induction of plant mutations by gamma rays". For the student, who had no experience in scientific research, it was a new, exciting world. At that time he did not recognize the fact that treating plant cells with gamma rays is like treating a Rembrandt painting with a shotgun. Nothing better could come from that type of experimentation, and in fact - it did not. The only useful thing was the experience gained by the young scientist.

**Hybrid wheat breeding** – While finishing his studies he was engaged in another, at that time very promising, scientific project: "Creation of hybrid wheat". At the beginning the project was led by professor Josip Milohnic. During 15 years of work on this project, he strongly believed in the idea of hybrid wheat and spent all his energy on this research.<sup>[1,2,3,5,7,8]</sup> The research team had some good, internationally recognized results in development of restorer-line parents that transferred pollen fertility to the hybrid. From 1979 to 1982, he organized and conducted four International Wheat Restorer Germplasm Screening Nurseries.<sup>[10]</sup> The collection of papers and results of hybrid wheat research were published in a special edition of Agriculturae Conspectus Scientificus, No.38(48) 1976. In 1977, at the 14<sup>th</sup> Hard Red Winter Wheat Workers Conference in Lincoln, Nebraska, he was a guest speaker and had a very provocative presentation "Challenging the myths of hybrid wheat". Professor Virgil Johnson, the organizer of that conference, left the auditorium during the presentation, because he did not support the idea of hybrid wheat, but at the same time did not want to contradict a presentation by a foreign guest.<sup>[6]</sup> Today, the speaker is grateful to Professor Johnson for that act, because now he understands that Professor was right and he was wrong. Fifteen years of hybrid wheat research was a long period in which to gain extensive experience and develop germplasm, but it also presented an opportunity for changing a way of thinking. As a bonus, some of the germplasm developed during hybrid wheat research was later used in conventional wheat cultivar development programs.

**Conventional wheat breeding** – Starting in 1980, the senior author's main scientific occupation was cultivar development based on the knowledge and germplasm he collected during hybrid wheat research project.<sup>[8,9]</sup> For a wheat breeder starting a cultivar-development program in Croatia at that time, there

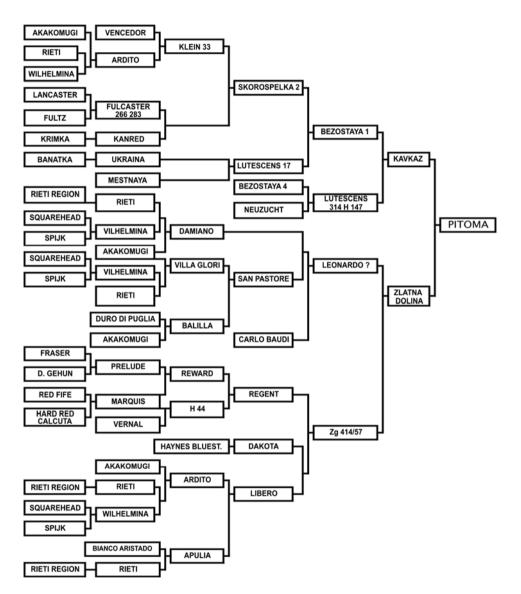


Fig. 1. Pedigree of wheat cultivar Pitoma (1983): 1BL.1RS chromosome translocation, Ppd1, Rht8, Saitama 27, Pm2, Pm8, Lr10, Lr26 and Vrn gene

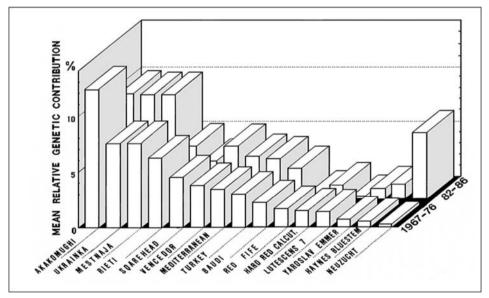
was available a good germplasm base, built around the famous Italian shortstrawed cultivars Leone (1955) and San Pastore (1958); the Russian highquality cvs. Bezostaya 1 (1962) and Aurora (1971); and the first -- also famous -- Croatian semi dwarf, high yielding cv. Zlatna Dolina (1971)<sup>[4]</sup>. His first released cultivar was Pitoma (1983), which carried the 1BL.1RS chromosome translocation, was higher in yield than Zlatna Dolina, and approached the quality of Besostaya 1. The main breeding goal at that time was strong straw, to confer resistance to lodging under heavy N application. Cv. Pitoma was able to resist lodging at application of 300 kg pure N per hectare. At that time, he took great pride in this achievement. But today he understands that once again, he was wrong.

Genetic erosion - Among 250 thousand species of flowering plants, human beings have cultivated only about 150 on a commercial scale. Indeed, the bulk of the human food supply comes from approximately 20 crop species, which are grown in the greater part of the planet's cultivated acreage. To make matters worse, genetic diversity is shrinking within those few species. Genetic erosion, within species comes from growing a small number of the best yielding, highly efficient cultivars and using a narrow range of parents in breeding programs. Although a certain amount of genetic diversity still exists within individual cultivars, especially of open pollinated species, the introduction and dissemination of high-vielding hybrids with similar genetic backgrounds has severely eroded that diversity. Continuous genetic erosion leaves vulnerable not only our crops but our own human species as well. Changes in environmental conditions or outbreaks of disease (often triggered by the appearance of new virulent races) could have devastating effects. Just two examples: a) the destruction of maize in the U.S. in 1970s by southern leaf blight fungus on hybrids having 'Texas' type of cytoplasmic male sterility,<sup>[19]</sup> or b) the stripe rust race 24 of barley that appeared in Colombia in 1975. It is almost certain that a plant breeder attending a barley conference in the UK transported race 24 urediospores on his clothing. Race 24 became rampant in South America, causing crop losses valued at millions of dollars.<sup>[20]</sup>

**Germplasm study** - In hybrid wheat breeding, and later in conventional breeding, one of the most important parts of the research was to examine patterns of diversity in the available germplasm base. In the days before plentiful molecular genetic markers, pedigrees of wheat cultivars were the most

useful source of information for plant breeders. That is why, in cooperation with Dr. Thomas S. Cox of the U.S. Department of Agriculture, the author analyzed pedigrees of 142 Yugoslav winter wheats. As a result, more than 20 thousand coefficients of parentage (rw) for all pairwise combinations of the cultivars were computed and cluster analyses were done.<sup>[11,12,13]</sup> (Up to that time, such winter wheat germplasm analyses were available only for U.S. wheat germplasm.)

Relative genetic contribution of predominant ancestors to the Yugoslav gene pool could be computed from the rw values.<sup>[14]</sup> Although over the past 20 years winter wheat cultivars have been released at a rapid rate by domestic breeding programs, results showed that many cultivars arose from similar parentage. Genetic uniformity expressed as mean coefficient of parentage (rw=0.13) was lower among Yugoslav than among U.S. soft (rw=0.19) or hard (rw=0.26) red winter wheat, respectively. That is, in that time the Yugoslav gene pool was more diverse. The most important ancestor, as evaluated by mean coefficient of parentage, was the Japanese cultivar 'Akakomugi' - source of the dwarfing gene *Rht*8 and the photo-period insensitive gene *Ppd1*.<sup>[13,14]</sup> (Fig.1.)



*Fig. 2. Changes in main relative genetic contribution in wheat gene pool of former Yugoslavia over the period of 15 years* 

Those analyses raised a question: Can progress in developing successively higher-yielding winter wheat cultivars be sustained if it relies exclusively on the current elite, narrow gene-pool?<sup>[19]</sup>

**Development of a broad-based wheat population and recurrent selection** – Based on pedigree analysis, thirty-five divergent ( $r_w=0.06$ ) pure-line parents (15 of domestic and 20 of foreign origin) were randomly intermated in 105 double cross (F1 x F1) combinations, and the resulting intercross (I1) plants were intermated once more in 97 combinations to form the I2 generation of the base population for the study.<sup>[15]</sup>

The best families were selected in the first cycle of recurrent selection, and their selfed progeny (I2S0:2 families) were tested in two environments: optimal (Croatia) and climatically stressed (Kansas, USA). Tremendous genotypic diversity of created intercross populations (I2S0:2 families) could be illustrated by the range of yield recorded in Croatia -- 3.6 to 9.8 t/ha – and the range of winterhardiness recorded in Kansas -- 0 to 100% survival. Considerable withinfamily segregation supported the expectation that selection of pure lines could produce even better yielding genotypes - selection of the top 20% families for yield was expected to increase mean yield by 1.09 t/ha.

Recurrent selection is, by its very nature, a long-term task. Even with no interruptions in the schedule, only one cycle was completed within the 5-year project period. Because allelic diversity is the lifeblood of each breeding program, and the results achieved are significant, it would be necessary to continue with maintaining and future improvement of the intercross population, but a lack of financial support led to termination of the project. Intercrossed populations are very useful foundations for selection of superior genotypes in the range of environments. Therefore, after finishing 5 years of the project (JFP648), the release of germplasm on the model of free germplasm exchange is proposed, and the germplasm has been sent to several universities and research centers in the USA. (*Note: The USDA has awarded the research team for the results achieved in the project.*)

**Studies on bread baking quality** – The increase in grain yield after introduction of Italian wheat germplasm (Leone, San Pastore, Produttore, Libellula) was accompanied by softer endosperm and poorer bread-making characteristics. The positive effect of the 1B/1R chromosome translocation

(introduced with Russian cvs. Aurora and Kavkaz) on grain yield was negatively correlated with bread-making quality. In Croatia, breeding for milling and breadmaking quality has not attracted sufficient attention for a long time.

Genetic variability of high molecular weight glutenins (HMW Glu) in the germplasm pool was also studied.<sup>[16,17,18]</sup> Cv. Divana, with the highly valued HMW Glu subunits 2\*, 7+9, and 5+10, was the first Croatian premium grain quality wheat suitable for organic cropping systems. It was registered in 1995. High quality characteristics, including 16 to 18% kernel protein, were inherited from the Nebraska wheat line NE7060 79Y335. Results of wheat breeding for bread making quality in Croatia have been comparable with those in some Western European countries.<sup>[21]</sup> Today, cv. Divana is still the best bread quality cultivar on the Croatian market.

**Powdery mildew resistance** - The next step was introducing resistance to the most prevalent disease, powdery mildew, from Romania, New Zealand and Kansas germplasm.<sup>[23,24]</sup> A set of completely resistant lines with *Pm* genes from KS92WGRC21 and KS92WGRC22<sup>[22]</sup> were developed and are currently being tested. The last step, introduction resistance to leaf beetle (*Oulema melanopus*) from GoldCorn wheat, is still in progress.

The route from induced mutation to breeding for organic practice was a long process with many wrong turns. But all of the experiences of those four decades, even the negative ones, provided a fertile medium for growth of the thought processes on organic agriculture and the ideotype of wheat suitable for organic production.

### WHY SUSTAINABLE ORGANIC AGRICULTURE?

As a result of long term engagement in research and breeding of wheat the senior author has become aware of genetic erosion in agriculture,<sup>[19]</sup> and has developed a deep interest in the bioethics of science and technology,<sup>[25]</sup> especially when a threat of new biotechnology and recombinant organisms (GMOs) to environment and human health is in question.<sup>[26,27]</sup>

However, today even contemporary western science across the disciplines rediscovers how nature is organic, dynamic and interconnected.

Some facts:

- 90 to 95% of all the species that ever lived are extinct;
- Thanks to "technology revolution" 90 thousand species are dying out annually;
- The human race is destroying soil 13 times faster than it can be created;
- Conventional agriculture has turned organic soil, which is a carbon sink, into a carbon source, and generates other greenhouse gases;
- Freshwater consumption is almost twice that of its annual replenishment;
- Conventional agriculture uses up to 7 times more energy per tonne of food than does organic agriculture.

Plant breeders have developed high yielding and hybrid forms of cereals, vegetables and fruits primarily for high yield and profit, not for nutritive value. The results of their efforts and of new modern technology – including heavy applications of mineral fertilizers and pesticides -- at first brought tremendous vield increases. But that did not last long. In the 1980s, the vield increase curve started to flatten. Conventional farming technology and powerful fertilizers and pesticides had practically sterilized the soil - reducing microbial life and leaving it with little or no available micronutrient fertility. If the soil does not have available nutrients, there is no way for cereals, vegetables and fruits to absorb them. It is not the nitrogen, phosphorus, and potassium macronutrients that are in question, but rather a number of very important micronutrients. Consider magnesium: if soil is poor in magnesium, plants grown in it will be also deficient in this microelement, and its nutritive value for humans will be diminished. Magnesium regulates over 300 functions in human body. The element is so critical that low levels in human blood can lead to serious health problems, including diabetes, heart disease, high blood pressure and heart attack.

As early as 1936, a group of scientists issued a dire warning that the mineral content of American soils was eroding, when they wrote Document No. 264. It was introduced on the floor of the United States Senate, but without effect.<sup>[28]</sup> A 2001 cover story in *Life Extension* magazine warned that vitamin and mineral

content of vegetables had drastically dropped, and little was done to reverse the trend.  $^{\left[ 29\right] }$ 

In the last fifty years, new breeds of wheat and new practices in cereal production have increased grain yields. But at the same time, the increased use of N fertilizers has caused pollution of underground water and atmosphere, thus creating ecological problems.

The pollution risk could be reduced by:

- genetic changes in the wheat plant that make it suitable for sustainable agriculture and
- introduction of legume species into crop rotation and significant changes in agricultural practice, including compost and stone meal application.

Breeding low imput, high quality wheat cultivars acceptable for sustainable agriculture will become even more important as years pass. Today, the objective should no longer be maximum grain yield, but rather rational and environmentally safe production. The goal should be a good compromise between production cost, yield and quality, while keeping the impact on the environment within tight limits. Rejecting the current doctrine of broad adaptation, organic farmers and consumers are demanding more varieties of locally adapted wheat.<sup>[30]</sup>

The plant ideotype suitable for organic wheat-cropping systems should include

- adaptation to local environmental conditions
- fast early growth
- high tillering capacity at a lower seeding rate
- lodging resistance in a 100-cm-tall semidwarf
- Resistance to prevalent wheat diseases and pests
- Good bread making quality, with grain protein content of at least 15 %

The best cultivar for that purpose to date is Divana. It is the best-quality bread wheat cultivar ever grown in Croatian fields (Fig.2., Tab.1. and Fig.3.), which compensates for its reduced yield potential. With conventional practices, it needs lower seeding rates, less fertilization, and less protection from pests and diseases, at the same time satisfying the needs of sustainable agriculture.

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Fig. 3. Heads and kernels of cv. Divana

Tab. 1.	Kernel characteristics of wheat cultivar Divana – Average and the best values
	achieved until now, location and year

Characteristics	Value		Location and year	
Characteristics	Average*	The best	Location and year	
Grain yield (t/ha)	5.867	8.212	Osijek, Croatia - 1994	
TKW (g)	40,1	52.2	Zagreb, Croatia - 1993	
Hectoliter wt . (kg/hl)	79,8	83.4	Osijek, Croatia - 2000	
Ash (%)	0.538	0.614	Osijek, Croatia - 1997	
Grain protein (%)	15,6	18.3	Probsdorf-Reichersberg, AUSTRIA – 2004	
Wet gluten (%)	35,3	45.7	Belje, CROATIA – 2006	
Zeleny sediment (ml)	62	72	Osijek, Croatia - 1999	
Milling (%)	71.97	79.95	Zagreb, CROATIA – 1999	
Bread yield (g/100 flour)	153,4	161,4	Zagreb, CROATIA – 2000	
Bread volume (ccm)	463	743	Probsdorf-Reichersberg, AUSTRIA – 2005	

\* Average based on four years and two locations of the State Commission Trials for Cultivar Release

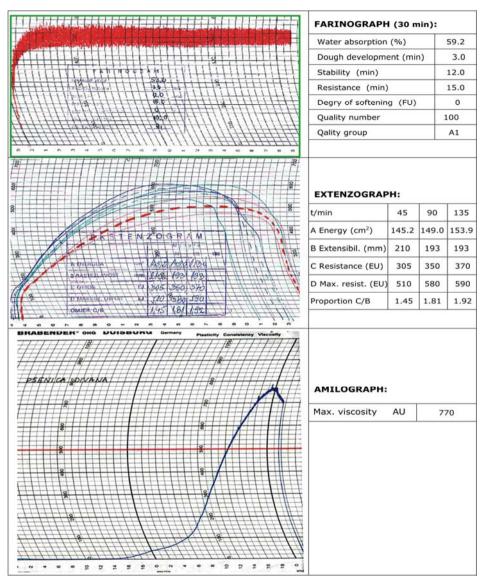


Fig. 4. Quality characteristics of cultivar DIVANA – Farinograph: even after 30 minutes there is no dough softening recorded

## CAN ORGANIC PRODUCTION BE COMPETITIVE WITH CONVENTIONAL PRACTICES?

This question is often answered in the negative by opponents of organic agriculture. But evidence is different. In fact there are many examples that organic yields beat the conventional.<sup>[32, 33, 34, 36, 37, 38]</sup> Substantial increases in per hectare organic food production are reported: 50-100% for rain-fed crops, and 5-10% for irrigated crops.<sup>[31]</sup> A summary of the impact of organic and near-organic on agricultural productivity in Africa, comprising the 286 projects in 57 countries, reported the global average increase of 79%.<sup>[34]</sup> All the case studies that focused on organic food production showed increases in per hectare productivity of food crops, which challenges the popular myth that organic agriculture conference in Addis Ababa (26-28 November 2008) is: "Ecological agriculture holds significant promise for increasing the productivity of Africa's smallholder farmers, with consequent positive impacts on food security, food self-reliance, environmental benefits and mitigation of climate change.<sup>[39]</sup>

To obtain experimental evidence bearing on that question in the Croatian context, we performed replicated tests in Križevci during the 2007/08 growing season. Eleven wheat genotypes -- three standard cultivars (Divana, Koleda, and Renan) and eight new breeding lines -- grown under two treatments in separate fields -- conventional and organic -- with 5 replications in  $5m^2$  plots.

The conventional field had received heavy application of chemical fertilizers and pesticides for at least four decades, and the preceding crop was corn. The soil (stagnic luvisol) was characterized by high acidity (pH = 4.8), good supply of phosphorus (18 mg  $P_2O_5$  per 100g of soil) and potassium (20 mg  $K_2O$  per 100g of soil), but low organic matter content (1.5 %). Microbial activity - the size of soil biomass, measured as soil respiration index (SRI) was 0.09 mg CO<sub>2</sub>/g of soil per day. Before seeding 400 kg/ha fertilizer NPK 7:20:30 and 200 kg/ha urea were added. Herbicide (Cougar 1.8 l/ha), insecticide (Chromorel D 0.5 l/ha), and fungicides (Bavistin 0.3 l/ha + Tilt CB 2 l/ha) were applied. Additional N was provided at tillering by applying 200 kg/ha KAN 27% N.

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The 'organic' field was under organic management for 7 years, and plots were sown after bitter lupine for green manure and stone meal application (400 kg/ha). The soil (stagnic luvisol) was characterized by moderate acidity (pH = 5.7), moderate phosphorus fertility (11 mg  $P_2O_5$  per 100g of soil), a good supply of potassium (18 mg  $K_2O$  per 100g of soil), and moderate organic matter content (2.3 %). Microbial activity (SRI) was 0.12 mg CO<sub>2</sub>/g of soil per day. No herbicides, insecticides or fungicides were applied.

On average for the eleven entries, test in organic production gave 1.185 t/ha (15%) higher grain yield than did conventional treatments (Table 2). Such a big difference in favor of the organic method was surprising, but it could be explained: in the 2007/08 growing season, the winter and spring were rather dry and mineral fertilizers, due to the lack of soil moisture, were not available to

Traits	Conventional	Organic	Diference		
Plant height (cm)	92	101	9 *		
Heads per sq m	742	1 235	493 **		
Grain yield (t/ha)	7.891	9 076	1.185 **		
Hectoliter wt. (kg/hl)	80.92	80.44	-0.48 <sup>NS</sup>		
TKW (g)	42.35	43.20	0.85 <sup>NS</sup>		
Bread baking quality parameters					
Grain protein (%)	14.64	12.61	2.03 **		
Wet gluten (%)	32.46	24.54	7.92 **		
Farinograph: Quality number	80.2	66.7	13.5 **		
Quality group	A1 - A2	A2 - B2	-		
Extensograph: Energy (cm <sup>2</sup> )	88.8	58.9	29.9 **		
Amilograph: Max. viscosity (AU)	1 391	1 347	44 <sup>NS</sup>		

Tab. 2. Average values for eleven entries grown in conventional and organic practice Križevci, Croatia – growing season 2007/08

\* significant at P=0.05; \*\* significant at P=0.01; NS - nonsignificant.

wheat plants in the conventional plots. Tillering was weak and plant height and head fertility lower. After heading in June there was a surplus of rain, much of it in the form of storms. This resulted in a surplus of accessible nitrogen that came rather late and could not compensate for poor mean yield components. On the contrary, due to heavy storms and weak plants, heavy lodging occurred. However, surplus nitrogen accessible in grain filling period resulted in the higher bread making quality.

In organic plots, wheat had nitrogen available from bacterial (*Rhisobium lupini*) fixation and green manure and expressed at least 30% stronger tillering (more heads per sq m), with taller plants. However, at the grain filling stage N availability was not so great, and that was reflected on a slightly lower bread making quality. Generally, the 2007/08 growing season was favourable for wheat, and above average yields were common in this region.

Cultivar Divana showed the greatest difference (2.02 t/ha or 35.1%) in grain yield between conventional (5.75 t/ha) and organic (7.77 t/ha) practices. That could be explained as an indication of its suitability for organic cropping system. On the other hand, cultivar Renan had low difference (0.54 t/ha or 6.4%) in grain yield between conventional (8.50 t/ha) and organic (9.08 t/ha) practice, and it might be due to the fact that it is more well-adapted to conventional agricultural practice.

### CONCLUSION

In organic soil, with organic practices, grain yields at the same or even higher level than with conventional practices can be achieved. Due to lower inputs, and no input of mineral fertilizers or pesticides, the production cost per unit of product will be lower, and nutritive quality of the food produced will be higher. The fear that organic practices could not provide enough food to feed the world has no support.

Wheat cultivars suitable for organic production are already available, and new ones are on the way. However, production practices should be adopted that will supply additional N in the grain filling period to enable good bread making quality.

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