

## COMPARATIVE EVALUATION OF QUALITATIVE VALUES IN GERMINATED AND UNGERMINATED WHEAT GRAIN GROWN IN DIFFERENT TECHNOLOGIES

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### Abstract

To estimate an influence of different growing technologies (organical and conventional), separate cultivars of wheat (Širvinta-1 and Zentos) and germinating time upon wheat grain nutritive composition (dry matter, ash, protein, fat and fibre), activity of enzymes ( $\alpha$ - and  $\beta$ -amylase, xylanase, cellulose, catalase and protease). It was established that the amount of dry matters reduced in germinated wheat and the content of mineral substances, protein and activity of ferments  $\alpha$  and  $\beta$ -amylases, cellulase, proteases and maltase was greater than in ungerminated wheat. Furthermore, the amount of cellulase reduced due to growing conditions and the distribution of fat amount was ambiguous regarding wheat species. (Širvinta-1: germinated > ungerminated and Zentos: germinated < ungerminated). The amounts of cellulase and the activity of ferments in ecologically grown wheat, both germinated and ungerminated were greater than in those conventionally grown, with exception of ferments of cellulase and protease. The activity of cellulase in ecologically grown grain was 39 % less and the content of protease was almost the same as in those conventionally grown.

Keywords: wheat grain, grow technologies, germinated, ungerminated wheat, nutritive composition, enzymes.

### 1. INTRODUCTION

The greater part of nutritional wheat grain is milled; during this process bran (with aleuronic layer) and germ are separated from endosperm. Thus, nutritional value of flour is lost, as bran contains many B group vitamins (thiamin B<sub>1</sub>, riboflavin B<sub>2</sub> and niacin B<sub>3</sub>), trace minerals and insoluble dietary fiber [37, 44]. Food, containing fiber, reduces the risk of cancer diseases, especially the cancer of large intestine [31], for the mentioned reasons, nutritional experts strongly recommend using not milled (whole) grain and products of their germination and fermentation [37].

Germinated grain was used in China not only for food but for medicine 5000 years ago [4]. It was observed that grain sprouts increase bio disposal of food products i.e. human body can promptly assimilate substances [20, 22]. Wheat grain sprouts may also scavenge free radicals in a human body, reduce the level of cholesterol and improve immune system [36]. Thus, at present germination is more and more widely used not only for improvement of nutritional grain quality but also as a raw material for healthy food production [20, 21, 33, 44].

The amount of dry matters and starch in germinated grain reduces. However, the

amounts of amino acids compositions, polyunsaturated fatty acids, B group vitamins, sugar increases and the content of anti nutritional substances reduces [5, 10]. Germinated grain contains huger quantum of essential amino acids (lysine, methionine etc.), which take part in protein production in a human body [11, 16, 35, 40]. Besides, dietary fiber in grain bran is not lost [36]. It was established that the longer is the period of germination, the vaster is the content of vitamin C, beta carotene and other antioxidants [6, 12, 13, 44]. Antioxidant biosynthesis at the time of grain germination depends on temperature, lighting, air and humidity [33, 34, 36].

The content of bioactive substances – ferments significantly increases in germinated grain [7, 18, 21]. It was measured that germination ferments stimulate the processes of anti-oxidation biosynthesis [27]. Ferments activity and grain biochemical composition depend on many factors: climatic conditions at the time of wheat growing and harvesting, fractioning while milling etc. [28].

Nowadays healthy food is becoming more and more popular. Firstly, it must not be contaminated with harmful for human body chemical substances. Ecological farming is one

of the solutions to reduce damaging anthropogenic influence to biota and produce free of chemicals raw material. The research results of many scientists proved that ecological products contain significantly fewer amounts of nitrates [1, 42, 43], pesticides [3] and heavy metals [19]. However, the mineral substances in ecologically grown grain increase only slightly [14, 43] and the amount of proteins reduce [8, 38]. Accordingly, one of the methods to improve the quality of ecologically grown wheat could be germination. Further, scientific literature provides very few research data on ecologically grown germinated wheat nutritional qualities. Thus, it is inevitably important to define the alternation of nutritional qualities in ecologically grown wheat during germination.

**The goal of this research is** to study the nutritional qualities of ecologically grown germinated winter wheat of different species and perform comparative analysis of grain grown in different technologies.

## 2. RESEARCH METHODS

The studies focused on analysis of germinated and ungerminated winter wheat of two species *Širvinta-1* and *Zentos*. These species were chosen for their high and constant fertility, good grain and winter crop qualities. Samples for research were selected from the year 2005 harvest grown in ecology centre of Lithuanian University of Agriculture. The sample soil is carbonated shallow sticky leached – (IDg8-k), up to the FAO – Calcari-Epihypogleyic Luvisols (LVg-p-w-cc), its granule metric composition is slight clay loam on middle heaviness and heavy claim loam. The basic soil agro chemical data:  $\text{pH}_{\text{KCl}} - 7.0$ ; humus – 2.10 %;  $\text{P}_2\text{O}_5 - 108.30 \text{ kg}\cdot\text{ha}^{-1}$  ir  $\text{K}_2\text{O} - 107.80 \text{ kg}\cdot\text{ha}^{-1}$ .

Ecological farm wheat of both species was fertilized –  $30 \text{ t}\cdot\text{ha}^{-1}$  with organic fertile mixture (50 % animal and 50 % vegetable mixture), conventional farm wheat was fertilized in spring –  $\text{N}_{60} - 200 \text{ kg}\cdot\text{ha}^{-1}$  once more in summer with nitre  $\text{N}_{60} - 200 \text{ kg}\cdot\text{ha}^{-1}$ , and in June on the leaves –  $\text{N}_{15} - 30 \text{ kg}\cdot\text{ha}^{-1}$ . Later it was twice fertilized with carbamide  $46.2 \% - 30 \text{ kg}\cdot\text{ha}^{-1}$ .

Seeds from conventional farm were pickled with Maksim –  $1 \text{ kg}\cdot\text{ha}^{-1}$ , growing regulator Cicocel –  $1 \text{ l}\cdot\text{ha}^{-1}$  was used. During vegetation they were sprayed with Folkon against diseases –  $0.5 \text{ l}\cdot\text{ha}^{-1}$ . No additives were used on ecological farm.

Meteorological conditions were favourable for wheat wintering and growing in the year 2005. Chemical composition, ferment activity and energetic value were measured in ungerminated and germinated for two days wheat seeds with three repetitions ( $n=3$ ).

*The method of germination:* grain was soaked for 12 hours in water (1 part of grain: 4 parts of water), germinated in dark premises at  $20^\circ\text{C}$  in Swiss *Bionatura* equipment and periodically washed with water in the morning and in the evening. This enabled to improve the access of oxygen and to keep the necessary amount of humidity.

*Sample preparation for analysis.* Germinated wheat was milled in laboratory mixer *Turrax T25*. Dry wheat grain was milled in laboratory mill with freezing system *Knifetec 1095 Sample Mill*.

Wheat grain biochemical composition indexes: humidity, amount of mineral substances, albumen, fat and cellar were evaluated in Olstein Warminsko-Mazurski university (Poland), in accordance with the standards at present valid in Lithuania: dry substances – LST 712:2000 [26]; protein – LST 1497:97 [23]; fat – ISO 7302:1982 [15], fibre – LST ISO 6865:2001 [25], ash – LST ISO 2171:2000 [24].

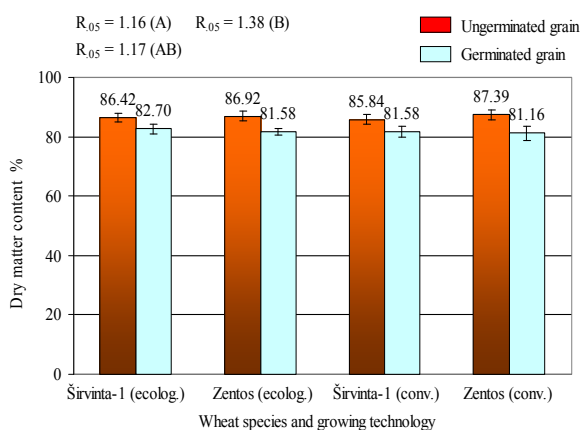
Grain ferments:  $\alpha$  and  $\beta$  amylases, cellulase, maltase and proteases activity research was performed in Olstein Warminsko-Mazurski university (Poland) ferment laboratory. The activity of cellulase, maltase,  $\alpha$  and  $\beta$  amylases in ferment preparations was established applying the method of DNS (3,5-dinitro salicylic acid) [2]. Ferment proteases activity was settled with Tomarelli method [41].

Student and Fisher tests were used for the comparison of the data average values (to 0.95 the probability level). Least significant difference ( $R_{.05}$ ) were calculated for factor A (ungerminated grain composition and ferment activity), for factor B (germinated grain composition and ferment activity) and their

reciprocity AB, applying SAS statistic method [30].

### 3. RESEARCH RESULTS AND DISCUSSION

One of the essential factors influencing grain relevance to nutritional raw material is grain's chemical composition, which depends on grain species and growing conditions. Basic grain chemical composition indexes are: dry matter, albumen, mineral substances and carbohydrates.

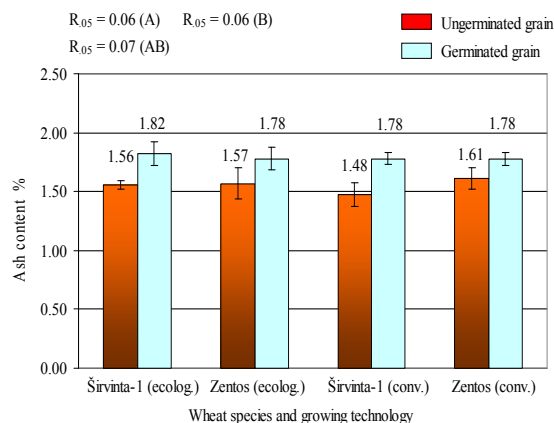


**Figure 1. Dry matter amount in ecologically and conventionally grown germinated and ungerminated wheat**

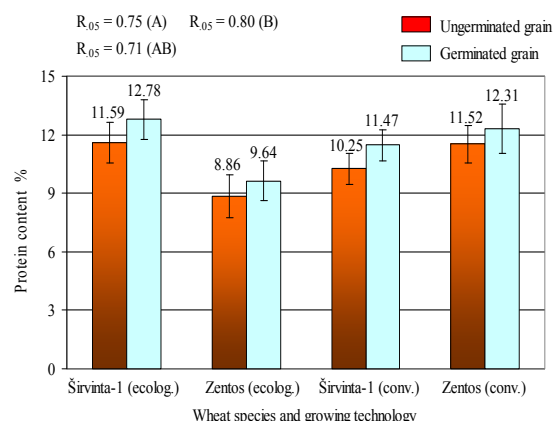
Dry matter analysis proved that its amount in both germinated wheat grown in ecological and conventional technologies is less (Figure 1). Estimating the results on both species, statistically significant data was obtained as a difference between averages, exceeding least significant difference limit ( $R_{0.05}(AB)=1.17\%$ ). In germinated wheat the amounts of dry matters were on average 5.64 % lower than in ungerminated. The obtained data validates the research conclusions of other researches who have established that while germination the amount of dry matters reduces [12, 21]. Statistically significant link comparing dry matters in different species or growing technologies was not observed.

While burning the product, organic substances burn and become ash. Food products contain from 0.03 % to 2.00 % [9] of mineral matters. Essential differences of mineral substances in analyzed wheat grain were noticed only between germinated and ungerminated winter

wheat. Though the differences are not significant but they exceed index  $R_{0.05}$  value 0.07 % (Figure 2). Growing conditions and wheat species did not have any influence on this index.



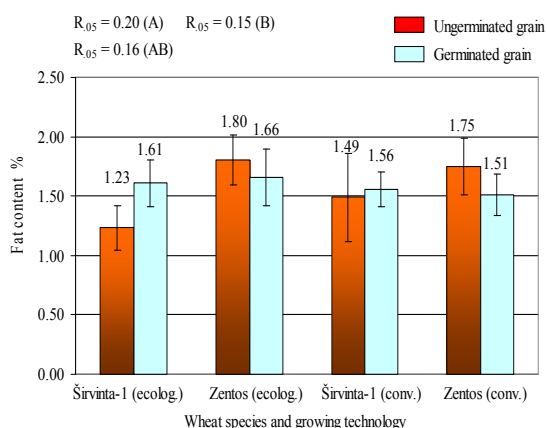
**Figure 2. Amount of ash in ecologically and conventionally grown germinated and ungerminated wheat**



**Figure 3. Amount of protein in ecologically and conventionally grown germinated and ungerminated wheat grain**

Vegetable protein, due to their complex structure, is digested and assimilated more complicated than animal fat. A human body needs from 70 g to 100g protein a day, depending on work specification; animal protein should compose 60 % of this amount and vegetable protein accordingly – 40 % [39]. The research results show that the essential factors influencing the amount of protein are genetic peculiarities of wheat species. In our case the species influence is related to growing technology (Figure 3). Species Širvinta-1

protein analysis results of both germinated and ungerminated wheat were 10.91 % higher in ecological farm in comparison with Zentos, while conventionally grown Zentos protein analysis results were on the contrary even 22.4 % greater than in Širvinta-1. The summary of the research results on both species determined that germinated grain contains on average 8.62 % more protein than ungerminated. Thus, germination could be successfully applied aiming to increase protein in conventionally grown wheat.

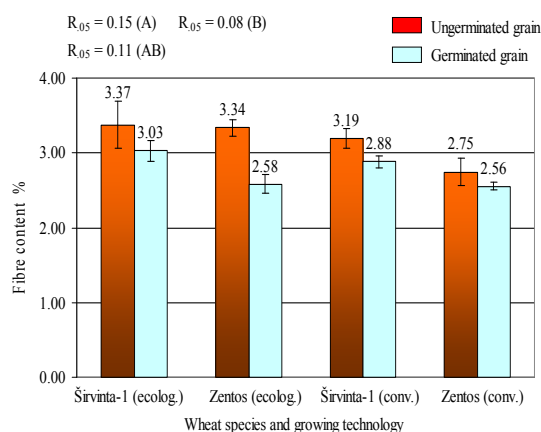


**Figure 4. Amount of fat in ecologically and conventionally germinated and ungerminated wheat grain**

Fat are the compositional part of cells and the source of energy in human body. Fat has twice as much calorie as protein and carbohydrates. Vegetable lipids contain vital for human's body unsubstantial fatty acid (oleinic, linoleic, linolioc), which serve as vitamins [32]. Lipids safeguard from oxidation and destruction of vitamins soluble in fat, regulate their resorption in human body, they protect internal organs from temperature changes [39]. In contrast to the described indexes, genotype ambiguity influenced germinated grain. Zentos ungerminated wheat grain has on average 10.75 % greater amount of fat than germinated, however, germination of Širvinta-1 increased this index for ecologically grown wheat even for 23.6 % (Figure 4).

Fibre is the basic matter of vegetable cells. Human body does not assimilate it, however, it stimulates peristalsis, influences cholesterol circulation, absorb compounds which are hardly soluble and eliminate them from the

organism [17]. In ungerminated wheat the difference  $R_{05}=0.15\%$  is more expressed than in germinated (Figure 5). Studies on fibre define that this index also depends on species and growing technologies. Ecological wheat grain had on average 8 % more fibre content than conventionally grown.



**Figure 5. Amount of fibre content in ecologically and conventionally grown wheat grain**

The grain itself contains various androgenic ferments, though in small amounts, the activity of these ferments influences the quality of grain raw material. The most relevant are ferments, which split main grain components: starch, protein and cell wall polysaccharides. Breathing and biosynthesis of compositional matters in ripe grain is fermentation process. Fermentation reactions are activated when grain is germinated. Table 1, 2, 3, 4 presents activity of ferments:  $\alpha$  and  $\beta$  amylase, protease, maltase and cellulase.

**Table 1. Ferment activity in ecologically grown ungerminated wheat grain**

Ferments activity	Ungerminated wheat grain	
	Ecological growing	
	Širvinta-1	Zentos
$\alpha$ -amylase, U·ml <sup>-1</sup>	359.67±12.80	346.54±2.72
$\beta$ -amylase, U·ml <sup>-1</sup>	374.51±4.60	363.19±3.65
Cellulase, U·ml <sup>-1</sup>	2.45±0.17	2.78±0.10
Maltase, U·ml <sup>-1</sup>	7.44±0.13	7.36±0.09
Protease, mAU·min <sup>-1</sup>	12.11±0.30	14.28±0.50

Averages and Standard deviations from 3 repetitions (n=3)



**Table 2. Ferment activity in chemically grown ungerminated wheat grain**

Ferments activity	Ungerminated wheat grain	
	Conventional growing	
	Širvinta-1	Zentos
$\alpha$ -amylase, U·ml-1	288.68±7.66	247.64±4.76
$\beta$ -amylase, U·ml-1	314.60±7.35	262.61±5.18
Cellulase, U·ml-1	2.88±0.12	2.89±0.10
Maltase, U·ml-1	6.55±0.05	6.52±0.04
Protease, mAU·min-1	11.03±0.31	13.96±0.34

**Table 3. Ferment activity in ecologically grown germinated wheat grain**

Ferments activity	Germinated wheat grain	
	Ecological growing	
	Širvinta-1	Zentos
$\alpha$ -amylase, U·ml-1	398.15±6.37	367.94±3.71
$\beta$ -amylase, U·ml-1	400.93±2.25	390.64±5.81
Cellulase, U·ml-1	4.24±0.15	4.49±0.15
Maltase, U·ml-1	8.58±0.07	8.75±0.11
Protease, mAU·min-1	15.96±0.49	16.76±0.52

**Table 4. Ferment activity in chemically grown germinated wheat grain**

Ferments activity	Germinated wheat grain	
	Conventional growing	
	Širvinta-1	Zentos
$\alpha$ -amylase, U·ml-1	307.51±4.96	292.17±6.29
$\beta$ -amylase, U·ml-1	348.94±2.69	331.30±4.14
Cellulase, U·ml-1	4.27±0.05	5.06±0.06
Maltase, U·ml-1	7.83±0.10	6.90±0.08
Protease, mAU·min-1	16.53±0.45	16.11±0.33

Two specific ferments prevail in wheat; they determine the changes in starch structure i.e.  $\alpha$  and  $\beta$  amylases. The basic active ferment in wheat is ferment  $\beta$ -amylases ( $\alpha$ -1.4-gliukozidmaltahydrolysis). Nevertheless, evaluating the quality of wheat in our climatic conditions, a considerable attention should also be paid to the activity of  $\alpha$ -amylases ( $\alpha$ -1.4-gliukozidhidrolases) [29]. The following two factors mainly influence the amount of both amylases in unharmed and ripen wheat grain: species and growing conditions. The performed research proves that all the researched factors

had essential influence to  $\alpha$ - and  $\beta$ -amylases activity (germinated wheat grain > ungerminated wheat grain; ecological growing system > conventional growing system; winter wheat species Širvinta-1 > winter wheat species Zentos). The identical results were obtained researching the activity of maltase, due to which maltose is hydrolyzed into two glucose cells.

Germination positively influenced all the other cellulase and protease ferments. Comparing inter species factor, statistically reliable links were established only in ungerminated wheat grain, where Zentos species is on average 28.8 % more efficient than Širvinta-1 one. Growing technology does not influence the activity of protease ferment. Germination mostly affected the activity of cellulase ferment; it increased even in 39 %. In contract to the other ferments, the activity of cellulase ferment is greater in conventionally grown wheat, both for germinated and ungerminated grains. Statistically significant differences were not fixed in different species.

#### 4. CONCLUSIONS

1. It was established that the amount of dry matters reduced in germinated wheat and the content of mineral substances, protein and activity of ferments  $\alpha$  and  $\beta$ -amylases, cellulase, proteases and maltase was greater than in ungerminated wheat. Furthermore, the amount of cellulase reduced due to growing conditions and the distribution of fat amount was ambiguous regarding wheat species. (Širvinta-1: germinated > ungerminated and Zentos: germinated < ungerminated).

2. The amounts of cellulase and the activity of ferments in ecologically grown wheat, both germinated and ungerminated were greater than in those conventionally grown, with exception of ferments of cellulase and protease. The activity of cellulase in ecologically grown grain was 39 % less and the content of protease was almost the same as in those conventionally grown.

3. Wheat species, irrespective of growing technology, influenced only the activity of  $\alpha$  and  $\beta$ -amylases and maltase. Statistically significant greater ferment activity was

established in wheat grain in both growing technologies.

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