

COMMODITY SCIENCE IN RESEARCH AND PRACTICE

FUTURE TRENDS AND CHALLENGES IN THE FOOD SECTOR



EDITED BY MAŁGORZATA MIŚNIAKIEWICZ & STANISŁAW POPEK

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AND CHALLENGES
IN THE FOOD SECTOR**

**Edited by
Małgorzata Miśniakiewicz, Stanisław Popek**

**Polish Society
of Commodity Science**



**International Society
of Commodity Science and Technology**



**Faculty of Commodity Science
Cracow University of Economics**



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Suggested citation:

Author A., 2014, Title of the paper, in: M. Miśniakiewicz, S. Popek (eds.)
Commodity Science in Research and Practice – Future trends and challenges in the
food sector, Polish Society of Commodity Science, Cracow, pp. xx-xx. ISBN 978-
83-940189-0-0

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of Economics, Cracow 2014

ISBN 978-83-940189-0-0 (printed version)
ISBN 978-83-940189-4-8 (html)

Year of publishing: 2014
Number of pages: 188

Publisher
Polish Society of Commodity Science
Sienkiewicza 4, 30-033 Cracow, Poland

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“The best way to predict the future is to create it.”

Peter Drucker

The challenges that face the present food industry are inseparably connected with the trend for innovativeness, as innovations are the core of modern economic growth strategies, company development and shaping the social prosperity in the modern world.

In the complex process of product innovation development a particular role is assigned to consumers, especially in current market conditions characterised by production growth, expanding commercial offer and civilisational and cultural changes. Product innovations in the food market are connected mainly with product development, thus with expanding and deepening commercial consumer-oriented offer to meet his/her expectations and needs related to food and nutrition.

The problem of innovativeness is well established in many scientific researches that deal with the economic importance of innovations based on knowledge, signification of innovations in regional development, Polish consumer behaviour towards branch innovations and identification of commercialisation conditioning and product innovation diffusion. This is reflected also in commodity sciences, including food commodity science.

The monograph entitled “**Future Trends and Challenges in the Food Sector**” is a part of “Commodity Science in Research and Practice” series. It is aimed at the presentation of modern perception of commodity sciences, thus evidencing that the Authors seek for innovative research solutions and applications.

Monograph contains 16 chapters written by the Authors being scientists from the European commodity science centres.

The monograph is published to present both the challenges that face food commodity science and observed innovative trends in this science and therefore it can be a valuable source of knowledge for practitioners and theorists of economic life as well as economics students.

*Małgorzata Miśniakiewicz
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ROMANIA'S EXPERIENCE REGARDING THE PRODUCTION, MERCHANDISING AND CONSUMPTION OF PRODUCTS BASED ON GMOS

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Introduction

Genetically modified food is defined by its method of production, which involves new technologies based on genetic engineering. Since their early years, genetically modified organisms have had a significant impact on both economic and social side. Despite of their substantial contribution for increasing the power of food and agricultural markets, GMOs had caused many cross-cultural differences, regarding their acceptance.

Starting with the highlighting of the biotech risks, there are worldwide mixed opinions regarding their benefits. In this context, the main objective of this paper is to identify the main position of a developing country, regarding its GMO production, merchandising and consumption and the way in which GMO's benefits and risks are perceived and acknowledged. Based on its GMO experience, the research was focused on Romania, a European developing country, with more than 14 years of GMO production.

The secondary objectives of the research are to clarify the legislation in Romania regarding GMO based products. The legislation in this country defines GMOs as "any organism except human, whose genetic material has been altered other than by crossing and / or natural recombination or any biological entity capable of replication or of transferring genetic material." (Law 214/2002 on the regime of production, testing, use and trade of genetically modified organisms by modern biotechnology and products derived from them).

On the other hand, due to ethical issues and related risks, most consumers do not accept the application of modern biotechnology to food production, although in general, the same technologies are accepted in the medical field and the environment. However, interestingly , some applications of genetic

engineering are more easily accepted than others in positions of perceived benefits to consumers.

The present research is structured into three main parts as follows: identifying the key points regarding Romania’s production and merchandising experience in GMO based products in a global context; a case study presenting the analysis of Roundup Ready soy species distribution in Bucharest supermarkets and the results of an exploratory study on a sample of availability concerning GMO’s.

The research methodology consists of the above mentioned Case Study developed in the ELISA Microbiology Laboratory of al Institute of Food Bioresources – Bucharest, the questionnaire based research and the statistical data retrieved from the scientific materials.

GMOs – Key points regarding Romania’s production and merchandising experience in a global context

Worldwide, genetically modified organisms have been produced, for commercial purpose, for about two decades. The first GMOs were created in the 1960’s and caused conflicting opinions that rapidly developed at a global scale. Despite the general context, regions like USA and South America registered an extensive growth regarding the areas sowed with GMOs, since early years.

Based on the potential benefits of genetic engineering, Romania represents one of the first European countries that embraced GMO production. Having as starting point the first USA area sown with GM crops (1996), Romania’s GMO production debuted in 1999, when several European counties, including Spain, France, Portugal and Ukraine, approached modern biotechnology methods (Table no.1)

Table 1. Area and Counties where GM crops are cultivated, by year (mill ha)

Year	Area	Countries
1996	2.8	US, China, Canada, Argentina, Australia and Mexico
1997	12.0	US, China, Canada, Argentina, Australia and Mexico
1998	27.8	US, China, Canada, Argentina, Australia and Mexico
1999	39.9	US, China, Canada, Argentina, Australia, South Africa, Spain, France, Portugal, Romania and Ukraine

Source: Timo Kaphengst, 2011

Amongst the GM crops grown worldwide, only four have a significant impact on the global market: soybean, cotton, maize and canola. According to the annual reports published by the International Service for the Acquisition

of Agri-Biotech Applications Organization (ISAAA) in 2013, the four GMO cops reached a total of 174 million ha worldwide (with soybean being the leader – Figure no 1).

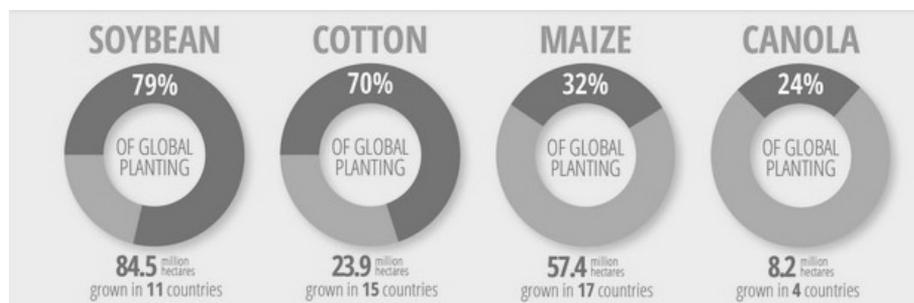


Figure 1. The 4 Major GM Crops and their share in the global market

Source: ISAAA Brief 45 -Global Status of Commercialized GM Crops: 2013

The first GMO adopted by Romanian agriculture was Roundup Ready soybean, a crop that, by the year of 2006, covered more than 100.000 ha. The main characteristic of biotech soybeans grown in Romania was the herbicide tolerance.

Cultivation of GM soybean in Romania has registered a fast growth since 1999. By the year of 2001, the area sown with GM soybean reached 14.250 ha, which represented 15% of the national soybean crop area of approximately 100,000 ha. In just 6 years, GM soybean areas extended more than 8 times, the crops being spread across an area of more than 115.000 ha. These numbers highlight a biotech rate of conversion of 79%, reported to the total area cultivated with soybean of 145.000 ha. Seven years of experience with GM soybean crops transformed Romania in an important European producer, more exactly the third largest producer in Europe, next to France and after Italy and Serbia Montenegro.

The year 2007 was marked by the adherence of Romania to the European Union. According to the country's new status, Romania had to transpose and apply the European GMO legislation, which was very rigid regarding key aspects like labelling, traceability and, most important, authorizations for commercial use. In this sense, a list of European regulation had to be adopted and implemented within the national law system (Table no 2).

Because of the fact that soybean was and still isn't approved for commercialized planting in the European Union, Romania had to cease the use of this GM technology. This Governmental decision had a huge economic impact on

Table 2. Basic Legislation regarding GMOs in EU and Romania

EU Regulations	Romanian Legislation
EC Regulation no. 1829/2003 on genetically modified food and feed	Resolution no. 173 from 09/02/2006 on traceability and labeling of GMOs and traceability of food and feed derived from genetically modified organisms.
EC Regulation no. 1830/2003 on GMOs traceability, labelling & derived food & feed	
EC Regulation no. 1946/2003 on cross-border movements of GMOs	Decision no 497 from 23/05/2007 on establishing European measures on transboundary movements of GMOs
Directive 2002/53 - common catalogue of agricultural plant varieties	Order no. 730 from 01/11/2006 on expelling GM soybean varieties from the official Romanian crops catalog.
EC Directive 2001/18 on the deliberate release into the environment of genetically modified organisms	OUG no. 43 from 23/05/2007 on the deliberate release into the environment and placing of GMOs the market.
Decision 2004/204 - Rules for registers on GMOs	Order no. 237 from 07/04/2006 on authorizing GM crop growers.

Source: original, based on data from the European Commission and Romanian Government

Romania's soybean production: in just 3 years, areas cultivated with soybean had reduced with 70% and national production has decreased with 80%. In other words, the total area cultivated with soybeans has dropped shapely from 177.000 ha in 2006 (including conventional and GM soybeans) to only 46.000 ha in 2008.

Romania's merchandising experience registered a significant growth right after the this main event from 2007. The decision taken by Romania's Ministry of Agriculture (MADR) had an important impact on the national market, triggering a major change in the country's net commercial balance and global status. Starting 2007, transgenic soybean was imported from non-EU countries, where soybean crops were still authorized for commercial purposes.

In other words, this impact was materialized in exchange of roles that occurred at an economical and commercial level: Romania took the role of net GM soybean importer after 2007. Before this year, Romania was considered a role model regarding the GM soybean production and was one of the leading global market exporting country.

Other effects of increasing soybean net imports have been immediately experienced by Romanian farmers that activated in other agricultural and industrial sectors: a rapid growth in the cost of animal feed and, thus, in the final cost of meet (especially poultry and pork).

The potential loss of farmers, which activated in this sector of GM soybean cultivation, has been estimated at 11.1 mil EUR in 2007 and 19.85 mil EUR in 2008. According to the latest publication regarding the Romanian

academic environment position on GMOs, the economic effects of ceasing the cultivation of GM soybeans are:

- mil EUR in 2007 and 1.725 mil EUR in 2008, invested in chemical fertilizers, that became necessary in the absence of nitrogen, that was provided to future crops, by the symbiotic bacteria from the GM soybean roots,
- 2.3 mil EUR in 2007 and 4.14 mil EUR in 2008, invested in additional mechanical works.

According to MADR, Romania represents a major soybean consumer with more than 500.000 tons annually (mostly consumed as animal feed in the agricultural sector). But, on behalf of Romania’s propitious pedo-climatic conditions, its annually production capacity can rise up to 2 million tones. These statistics highlight a difference of 1.5 million tons, which can be exported.

In 2007 the only GM plant approved in the EU for commercial use was the transgenic maize MON 810 (developed by Monsanto – France). After 7 years of GMO experience (acquired by the GM soybean cultivation period), Romania soon embraced GM maize as well, but on a lower scale than soybean, in 6 years the largest area cultivated with GM maize did not exceed 7.200 ha (Table no 3).

The slow evolution of maize crops had bureaucratic and financial causes, imposed by the new European standards and, also, organizational causes determined by the limited supply of GM maize seeds. Another general cause of the slow evolution of new GM crops was the severe economic recession that Romania faced starting 2008.

Table 3. Area cultivated with GM maize in Romania (in ha)

Year	2007	2008	2009	2010	2011	2012	2013
Area	321	7.146	3.243,5	756	584	216.5	834.62

Source: original, based on data from the Romanian Ministry of Agriculture

Comparing Romania’s situation with other Center and Eastern European countries’ GMO experience, we encounter similarities or major differences.

Poland’s GM Bt maize experience, for example started in 2006, with a total area of 100 ha. After a slow evolution, areas sowed with GM maize reached 3000 ha and the trend was stable until 2012, when Bt maize was banned in the country. No crops were officially registered; in spite of the planting acknowledged (Table no 4).

Table 4. Area cultivated with GM maize in Poland (in ha)

Year	2006	2007	2008	2009	2010	2011	2012
Area	100	327	3000	3000	3000	3000	N/A

Source: ISAAA Brief 44 -Global Status of Commercialized GM Crops: 2012

Other European countries like Austria, took a total different approach regarding the GMO subject, right from the early years, as EU authorized, in certain situations, the banning of GMOs at national levels, in its member countries. Like Poland in 2012, other countries have invoked the so-called 'safeguard clause earlier (Art. 23 Dir. 2001/18/EC). This involves temporary restrictions or total prohibition regarding the use and/or sale of the GM product on their territory. Next to Austria; Greece, Hungary, Germany and Luxembourg adopted this position.

There are other European countries, like Italy or France, where Bt maize planting has never been allowed, in spite of earlier GM Soybean experience (Thomas Venus, 2011). Also, there is no cultivation of GM crops in Slovenia, no field trials being identified in this country as well. (The Role of GMOs in the New Member States – a critical appraisal, European Parliament)

In 2013, Romania is still one of the 27 countries worldwide that planted GMO crops (according to Global Status of Commercialized Biotech/GM Crops: 2013). Furthermore, Romania is one of the 5 European countries that still cultivate GM crops, including Spain, Portugal, Czech Republic and Slovakia.

As a 2007 accession country to the EU, Romania's positive experience over the last eight years with biotech soybeans has important policy implications concerning the cultivation of biotech crops in all other EU accession countries like Bulgaria, and other neighboring countries in the Black Sea region (ISAAA Brief 44 -Global Status of Commercialized GM Crops: 2012). Romania represents today a role model regarding the growth of biotech crops, having an important implication in the European and global GMO market.

Case Study: Analysis of Roundup Ready soy species distribution in Bucharest supermarkets

In Romania, genetically modified Roundup Ready soybean was cultivated legally since 2000, although it was not authorized and the European Union. In 2005, from the 130,000 ha planted with soybeans in Romania, about 88,000 ha were represented by GM crops. Romanian Government decided to ban the cultivation of genetically modified soybean plants as of January 1, 2007 according to EU regulations.

At EU level soy is approved for use as food or feed. This was rather a political decision, taking into account the perspective of Romania's EU accession in 2007. In the same year, Roundup Ready soybean was authorized only for testing by the State Institute for Variety Testing and Registration

(ISTIS). Testing was done in five centers of the ISTIS network: Mircea Voda, Tecuci, Targoviste, Peciu Nou and Satu Mare and the amount of soy used for testing was 475 kg. The purpose of testing was the registration of varieties in the Official Catalogue of Varieties of Romania.

For the strict monitoring and evaluation of the presence of the GM crop on the market, the Ministry of Agriculture and Rural Development (M.A.D.R) has developed a detailed plan of inspection and control. M.A.D.R. has done conventional soybean seed testing in order to determine contamination by genetically modified seed in the ELISA Microbiology Laboratory from the National Institute of Research and Development for Food and Bioresources, Bucharest IBA .

a. The objective of the study

The objective of this study was the analysis and evaluation of soybean samples, fresh or processed, randomly chosen from the supermarkets of Bucharest. Vegetal food (soy) and also food products of animal origin which contain vegetal protein (soy) have been analyzed. The method used in the Microbiology Laboratory – ELISA of IBA Bucharest for the extraction of the nucleic acids was recommended by the Roche producer – High Pure GMO Sample Preparation Kit.

b. The samples

The analyzed samples have included a wide range of soy products, from the less processed ones such as flour, to the highly processed ones such as tofu, soy milk, soy drinks and sweets. Out of the total samples, 24 samples have been identified with genetically modified soy content, from 0,2 to 100% and 56 samples which do not contain genetically modified soy. The possibility of quantifying genetically modified insert and establish the limit of detection to allow rapid identification of products containing genetically modified soy and the need to label products containing more than 0.1 % GMOs were also a priority in conducting this study .

The samples analyzed included a wide range of soy products , from the very little processed as flour to the high processing such as tofu , soy milk , soy drinks and sweets. The results of 80 tests are given in Table 6 soybean on the basis of two indicators:

- lectin gene - specific plant ;
- p35S promoter (amount of target DNA , GM) - GM specific .

Table 5. Summary results

Nr. crt.	Sample name	Nr of analyzed samples	Number of positive samples for lectin gene	Number of negative samples for p35S promoter	Number of positive samples for p35S promoter
1.	Drinks	5	5	5	0
2.	Soy milk	10	10	10	0
3.	Soy Flour (producer 1)	4	4	4	0
4.	Vegetal Pate	7	7	7	0
5.	Cereals that contain soy	1	1	1	0
6.	Textured protein	8	8	8	0
7.	Sweets	5	5	5	0
8.	Tofu	5	5	5	0
9.	Soy Fiber	1	1	0	1
10.	Soy granules	1	1	0	1
11.	Soy Flour (producer 2)	2	2	0	2
12.	Soy Schnitzel	1	1	0	1
13.	Soy protein concentrate	1	1	1	1
14.	Textured Soy Schnitzel	1	1	1	0
15.	Vegetal meatballs	1	1	1	0
16.	Vegetal Pate	1	1	1	0
17.	Soy Beans	3	3	0	3
18.	Textured soy protein	7	7	0	5
19.	Vegetal Mushroom Pate	1	1	1	0
20.	Soy Cubes	1	1	0	1
21.	Soy Flour	2	2	0	2
22.	Vegetal Salami	1	1	0	1
23.	Soy Schnitzel	1	1	0	1
24.	Soy granules	1	1	0	1
25.	Vegetal Hamburger	1	1	1	0
26.	Bird baloney	1	1	1	0
27.	Meat roulade	1	1	1	0
28.	Indigen Soy Beans	1	1	0	1
29.	Granulate textured soy	1	1	0	1
30.	Kaizer	1	1	1	0
31.	Baloney	2	2	1	1
32.	Soy Lecitine	1	1	0	1
	Total	80	80	56	24

Source: own research. ELISA Microbiology Laboratory of al Institute of Food Bioresources - Bucharest.

The samples used in this study consisted of seeds and processed products purchased from different manufacturers or importers of the market. Seed samples of 500 grams and 300 g were milled in a laboratory mill until a fine powder was obtained. During milling sterile equipment and materials were used to avoid cross-contamination.

The samples were packaged and labeled as grinding and homogenization and stored at 4 ° C until analysis. Samples were tested by molecular biology techniques (PCR and Real Time PCR conventional) and the results are presented for each sample. (see Table 5)

c. Reference Materials

These materials are represented by Roundup Ready soybean homogeneous powder in different concentrations: 0.0%, 0.1%, 0.5%, 1%, 2% and 5% (w / w) supplied in bottles of about 1 g, which is maintained at 4 ° C in the dark.

From the total of the analyzed samples, 24 samples were found containing genetically modified soybeans between 0.2 and 100 % , and 56 samples that do not contain genetically modified soybean . The data presented shows that although some food containings GMOs , the labeling of these products does not contain this information.

Exploratory study on a sample of availability concerning genetically modified organisms

A questionnaire based research was developed during 1.03.2011 – 1.04.2011 among 50 persons of Romanian nationality aged above 18 years old which were familiarized with the genetically modified organism notion. The central objective of the research has succeeded to identify the perception that consumers have about the "new technologies" applied to food, especially the contradictions and challenges that genetically modified organisms can bring.

The secondary objectives of the research consisted in the consumer's attitude concerning genetically modified organisms, their knowledge concerning these products based on GMO's, the level of satisfaction obtained by consuming a GM organism based product, the consumption of certain food identified with genetically modified soy content put on sale in the supermarkets from Bucharest, the safety problems concerning GM food, the advantages and also the characteristics of GMO products which mostly influence consumers. The figure below shows where all these issues stand in relation to Romania's main actors on the market of GMO based products.

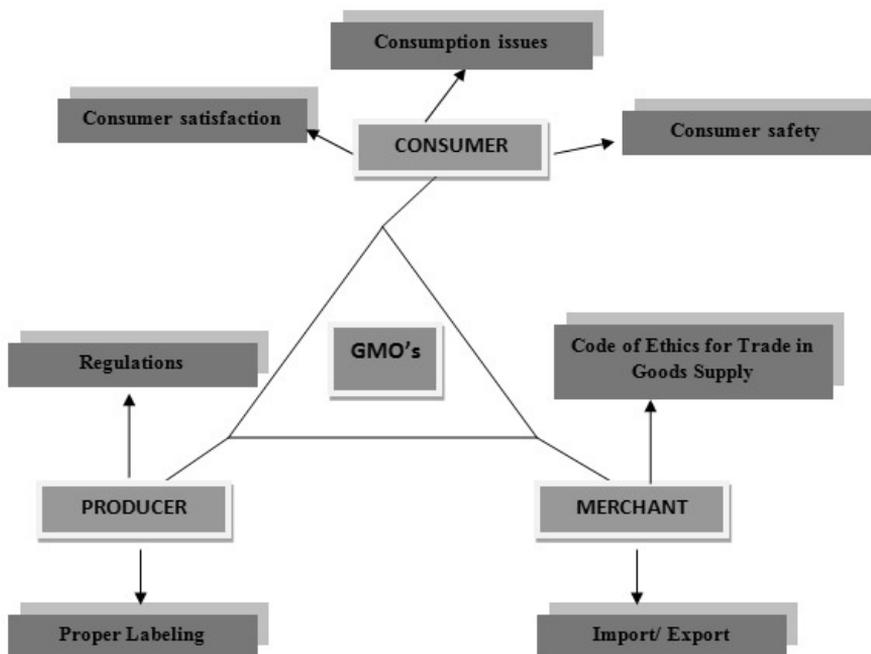


Figure 2. Romania's main actors on the market of GMO based products

Source: own research.

The exploratory study has shown the following results :

- increased interest of the consumers for the products made through modern biotechnology.
- bioremediation, medicine and genetic testing are encouraged.
- interestingly, some applications of biotechnology are more easily accepted than others, depending on the benefits perceived by consumers according to the available information.
- customers were not very satisfied from consumption of products containing genetically modified soy, which leads to the following conclusion: the assortment of such products should be improved in the future (Cristescu, Pamfilie 2011).

Conclusions

Several important conclusions have resulted from the present research. This paper aimed and managed to identify Romania's positioning on the EU market as a producer, merchandiser and consumer of GMO based products. Romania was one of the biggest Roundup Ready soybean producers in

Europe, before 2007 when the country adhered to the European Union and GM soybean production was prohibited by European regulations. Romania's merchandising experience registered a significant growth right after the this main event from 2007.

The consumer's distrust in the application of modern biotechnology to food production has led to the proposal of new directives and laws to ensure better food labeling and traceability of genetically modified products or of products that contain genetically modified ingredients.

Along with the GM soy termination, the country experienced an economic impact which affected many farmers and local producers, that had a high degree of acceptance regarding GMOs. In 2013, Romania is still one of the 27 countries worldwide that planted GMO crops. Furthermore, Romania is one of the 5 European countries that still cultivate GM crops, including Spain, Portugal, Czech Republic and Slovakia.

The data presented in the Case Study regarding the analysis of Roundup Ready soybean shows that although some food contains GMOs, the labeling of those products does not contain this information.

The distrust of most consumers that do not accept the application of modern biotechnology to food production has led to the proposal of new directives and laws to ensure better food labeling and traceability of genetically modified products or of products that contain genetically modified ingredients. If proper labeling is made, consumers are better informed and can choose between foods that are genetically modified and non- GM.

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THE PROBLEM OF ENRICHING YOGHURTS WITH ASCORBIC ACID

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Introduction

Yoghurts and their uses

Contemporary consumers use yoghurts as drinks, snacks or self-sufficient meals, though a variety of cooking applications have been found, like adding them to soups, sauces, cold borscht, pates, cakes, cocktails and desserts. Dishes based on or including additions of yoghurt have gained popularity in some countries. Chicken soup with yoghurt is a popular Bulgarian speciality, while meat in yoghurt sauce is a Kazakh invention. The latter is based on meat and bone extract seasoned with pepper, salt, onion, chives, sorrel, turnip, and horseradish, then fermented with yoghurt and thickened with egg yolk. Yoghurts (*Ayran*) mixed with salted water are popular in Turkey as they are perfect at quenching thirst and supplement sweated salt on hot days. Mango, saffron and rosewater are added to yoghurt in India, on the other hand. Germans like their yoghurt with muesli, the English with jams, and Scandinavians with almonds (Mojka 2013).

A growing interest in consumption of yoghurts can be observed in Poland as well. The National Office for Statistics figures for 2006 show that the monthly consumption of yoghurt and fermented dairy drinks averaged 4.94 kg a head (an average European Union citizen consumed about 15 kg at that time) (Krasowska, Salejda 2008). K. Świetlik (Świetlik 2013), who used macroeconomic data published by the National Office for Statistics, surveys of household budgets and her own analysis of market statistics, implies that, in effect of a considerable slowdown of Polish economy, consumption collapsed, real-terms spending on food, retail and consumption of food products by households declined. Household consumption of a number of food articles was considerably lower in 2012 compared to 2008. Several product groups, including yoghurts, resisted the crunch and their consumption even rose.

Euromonitor International data indicate the average yoghurt consumption in Poland should reach 9.2 kg by 2015 (<http://rme.cbr.net.pl/archiwum/maj-czerwiec-nr-43/51-rolnictwo-w-unii-europejskiej/125-ronie-spoycie-yoghurtow-w-polsce.html>) [accessed 21May 2014]).

Yoghurts in the opinions of Polish consumers

Considerable interest in yoghurts is confirmed by surveys among Polish consumers. Research by J. Stankiewicz and M. Lange implies the fermented dairy drinks most popular with pre-school children are yoghurts (n = 200), with merely 7.4% of respondents claiming to prefer other drinks, such as buttermilk or kefir (n = 16). 61% of those surveyed claimed they consumed yoghurts on a daily basis (n = 132), 36% pointed to 2–3 times a week (n = 78). J. Stankiewicz and M. Lange cite Adamczyk and Szymandera-Buszki to say that yoghurt is the most popular fermented dairy drink with youth (Stankiewicz, Lange 2012). G. Krasowska and A. Salejda report, referring to results published by Adamczyk, that yoghurt consumption is declared by 70% of children and young people, attracted chiefly by its nutritional value (Krasowska, Salejda 2008). Participants in the survey, students of Wrocław universities aged 18 to 26 in the academic year 2006/2007, stated the fermented dairy products they most often consumed were yoghurt and kefir (consumption of yoghurts was declared by 40% of female and 36% male students). A significant number of female students (47%) and only 21% male students claimed they consumed yoghurts daily, with 32% respondents drinking it several times a week (Krasowska, Salejda 2008). Nearly all respondents (98%), solely shoppers for whole families familiar with fermented dairy products, declared they bought and consumed fermented dairy products, most commonly yoghurts (97%), in a survey conducted by M. Nowak, T. Trziszka and M. Szołtysik - 39% of those surveyed consumed yoghurts daily, 43% 1-3 times a week (Nowak, Trziszka, Szołtysik 2007).

Yoghurts – nutritional value

Basic ingredients of natural yoghurts: 3.4-4.8% of protein, 0.8-3.0% of fats, and 3.8-11.8% of carbohydrates; of fruit yoghurts: 2.8% to 4.5% of protein, 1.5% to 3.3% of fats and 8.8% to 15.7% of carbohydrates (Kunachowicz, Kłys 2002).

Yoghurts are a good source of minerals, especially of easily available calcium, of considerable importance for adequate bone structure and function. Research by A. Kunachowicz and W. Kłys indicates that natural yoghurts contain 106 to 170 mg calcium per 100g of the product and 92-122 mg of phosphorus/100g of the product, whereas fruit yoghurts: 104-134 mg calcium per 100g of the product and 87-96 phosphorus/100g of the product,

respectively (Kunachowicz, Kłys 2002). Yoghurts are a source of vitamins soluble both in water and in fats (Kunachowicz et al. 2005). Table 1 presents the energy value and contents of the basic nutrients in yoghurts, table 2 presents contents of selected minerals and table 3 the content of vitamins in yoghurts.

Table 1. Energy value and contents of the basic nutrients in yoghurts

Product name	Nutrients (per 100g of the product)				
	Energy value (kcal)	Protein (g)	Lactose (g)	Sacharose (g)	Cholesterol (mg)
Natural yoghurt 2% fat	60	4.3	4.6	1.0	8
Blueberry yoghurt 1.5% fat	62	3.7	5.0	2.2	7
Cherry yoghurt 1.5% fat	61	3.7	5.1	1.7	6
Apricot yoghurt 1.5% fat	63	3.7	5.0	2.9	7
Strawberry yoghurt 1.5% fat	60	3.7	5.0	2.4	6
Strawberry cream yoghurt 8% fat	139	2.3	3.2	10.0	30
Strawberry yoghurt reduced fat	46	4.2	5.8	0.0	2

Source: (Kunachowicz et al. 2005, Mojka K. 2013)

Table 2. Contents of selected minerals in yoghurts

Product name	Minerals (per 100g of the product)				
	Calcium (mg)	Phosphorus (mg)	Potassium (mg)	Magnesium (mg)	Zinc (mg)
Natural yoghurt 2% fat	170	122	200	17	0.46
Blueberry yoghurt 1.5% fat	134	96	170	14	0.42
Cherry yoghurt 1.5% fat	134	98	182	15	0.38
Apricot yoghurt 1.5% fat	133	96	200	14	0.38
Strawberry yoghurt 1.5% fat	134	97	179	15	0.37
Strawberry cream yoghurt 8% fat	88	64	109	9	0.24
Strawberry yoghurt reduced fat	150	116	188	15	0.48

Source: (Kunachowicz et al. 2005, Mojka K. 2013)

Table 3. Content of vitamins in yoghurts

Product name	Vitamins (per 100g of the product)				
	A (w µg)	D (w µg)	B ₂ (w mg)	B ₁₂ (w µg)	C(w mg)
Natural yoghurt 2% fat	16	0.03	0.216	0.50	1.00
Blueberry yoghurt 1.5% fat	20	0.02	0.183	0.36	2.8
Cherry yoghurt 1.5% fat	24	0.02	0.189	0.36	2.5
Apricot yoghurt 1.5% fat	57	0.01	0.185	0.35	1.90
Strawberry yoghurt 1.5% fat	19	0.01	0.187	0.35	9.90
Strawberry cream yoghurt 8% fat	70	0.08	0.113	0.24	1.70
Strawberry yoghurt reduced fat	13	0	0.205	0.40	1.50

Source: (Kunachowicz et al. 2005, Mojka K. 2013)

Yoghurt – yoghurt bacteria

Presence of viable bacteria cultures is an essential advantage of yoghurts from the viewpoint of health. As defined by the Polish standard PN-A-86061 'Milk and dairy products. Fermented milk', yoghurt is fermented milk containing specific, symbiotic cultures *Streptococcus thermophilus* and *Lactobacillus delbrueckii subspecies bulgaricus*. This microflora must remain active, viable and numerous in a finished products by the end of its shelf life. In accordance with recommendations of FAO/WHO Codex Alimentarius and FIL/IDF, specific microflora of yoghurt should total a minimum of 10^7 cfu/g on the last day of storage. Such a definition of yoghurt stresses the role of micro-flora in this product. Yoghurt fermentation bacteria are symbiotic. Streptococci initially grow faster, producing lactic acid as well as acetic acid, acetic aldehyde, diacetyl and formic acid. The presence of formic acid and the reduced redox potential of the medium foster growth of yoghurt rods. The yoghurt rods in turn, displaying higher proteolytic activity, release small-molecule peptides and amine acids from milk protein, which enables further growth of normally low-protelytic strains of *Streptococcus thermophilus*. The living processes as in yoghurt fermentation bacteria result in multi-direction changes in milk's ingredients. The changes comprise primarily:

- the production of acids (mostly lactic acid)
- partial disintegration of milk sugar
- disintegration of proteins resulting in increased amount of free amino acids
- the increase of some vitamins' content, mostly folic acid and vitamin B 12 (Tamime, Robinson 1985).

Bacteria in yoghurts have a variety of effects on functioning of the human body (Adolfsson et al. 2004). M. Piaia et al. compared yoghurts containing viable bacteria cultures and thermised yoghurts (Piaia et al. 2003). Their results show that, seen from a nutritional/functional point of view, heat treatment dramatically alters some intrinsic beneficial properties of yoghurt. There is emerging evidence that fermentation of milk has positive effects on the release of bioactive peptides, and that consumption of yoghurt has beneficial effects on the gut equilibrium, on the immune system, on prevention of infections, on mutagenesis and carcinogenesis, on oral health and (in animals) on prevention of collagen-induced arthritis. These beneficial effects have not been shown for heat-treated fermented milk (Piaia et al. 2003).

Yoghurt bacteria are not typical probiotics. M. Elli et al. report that yogurt bacteria, especially *L. delbrueckii* subsp. *bulgaricus*, can be retrieved from feces of healthy individuals after a few days of ingestion of commercial yogurt. Moreover, their results indicate that very careful setup of the analytic procedures can dramatically improve the reliability of studies of the survival of yogurt starters (Elli et al. 2006). Kudeřka W. says *Lactobacillus bulgaricus* strains have stronger probiotic properties than *Str. thermophilus* as they show greater capacity for settlement in the gut, overcoming the high acidity barrier of gastric juices, exhibiting resistance to physiological concentrations of bile acids and production of bacteriocins (Kudeřka 2008).

Yoghurt – enrichment

Popularity and properties of yoghurts give rise to attempts at enriching them with a variety of ingredients: vegetable substances of bioactive activity (Stankiewicz 2009), calcium and magnesium salts (Żbikowska, Żbikowski 2011). Interesting results of supplementation with milk powder and amine acids are presented by M. H. Naji, Z. Hashemi, M. Hoseini (Naji, Hashemi, Hoseini 2014). The British scientists suggest the products are fermented appropriately to enrichment with L-ascorbic acid and iron as they contain plenty of organic acids (Teucher, Olivares, Cori 2004).

L-ascorbic acid plays a major role in the human body. It also serves to prevent a range of diseases, including cancer and cardio-vascular conditions (Borek-Wojciechowska). Reports by scientists indicate that vitamin C can be used to treat *Helicobacter pylori* (Borek-Wojciechowska 2009)]. Oh Y. et al. show yoghurt microorganisms may adversely affect *Helicobacter pylori* (Oh et al. 2002). L-ascorbic acid has strongly oxidising properties. When weakly oxidised, it converts into L-dehydroascorbic acid via a radical intermediate compound known as L-monodehydroascorbic acid (Gawęcki, Hryniewiecki 1998). The latter has the ability of reducing the oxidation-reduction potential (Zaleski 1985).

The redox potential is a physico-chemical parameter defining oxidative or reductive properties in the environment and dependent on food composition (content of thiol amine acids, peptides, proteins and reducing sugars), pH, temperature and, to a great extent, on free oxygen concentration. The parameter plays an important role in microorganism cell physiology as it affects growth potential (Alwazeer 2003).

The research described below was designed to establish the moment for yoghurt enrichment with L-ascorbic acid.

Material and methods

Yoghurts were produced in laboratory conditions. 1L of UHT milk was inoculated with *Lbc. bulgaricus* and *Str. thermophilus* strains using quantities computed for purposes of acidifying 1L as instructed by the vaccine manufacturer. The liquid was then filled into three plastic containers (PS). Contents of one were enriched with L-ascorbic acid in the form of an aqueous solution. The containers were sealed and the cultures were incubated at 42±1°C for 5 hours. After that time, L-ascorbic acid in the form of an aqueous solution was added to the curd in one of the non-enriched containers, the contents were mixed and sealed, and all the yoghurts were stored at 4±1°C for 28 days.

A well-mixed representative sample of yoghurt, out of which 10±1g was weighed to produce a 10⁻¹ and further dilutions. A deep inoculation of the appropriate sample dilutions was produced on two parallel Petri dishes, covered then with a medium appropriate to the given culture. M-17 (Biomerieux) medium was applied to *Str. thermophilus* and MRS (Biomerieux) was applied to *Lbc. bulgaricus*. Colonies were counted for each separate micro-organism grade at the end of the incubation.

Results

Test results for the yoghurts (mean values of three runs) enriched with L-ascorbic acid prior to and post incubation, as well as for the non-enriched yoghurts after the first (X = 0 - after 5 hrs of incubation) and successive days of storage are illustrated in figures 1 and 2.

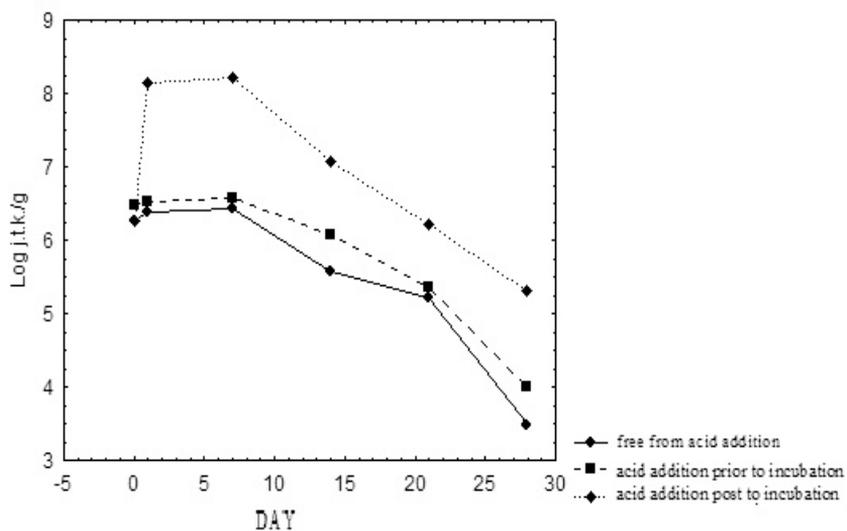


Figure 1. *Str. thermophilus* changes at the time of yoghurt storage as dependent on the moment of L-ascorbic acid enrichment

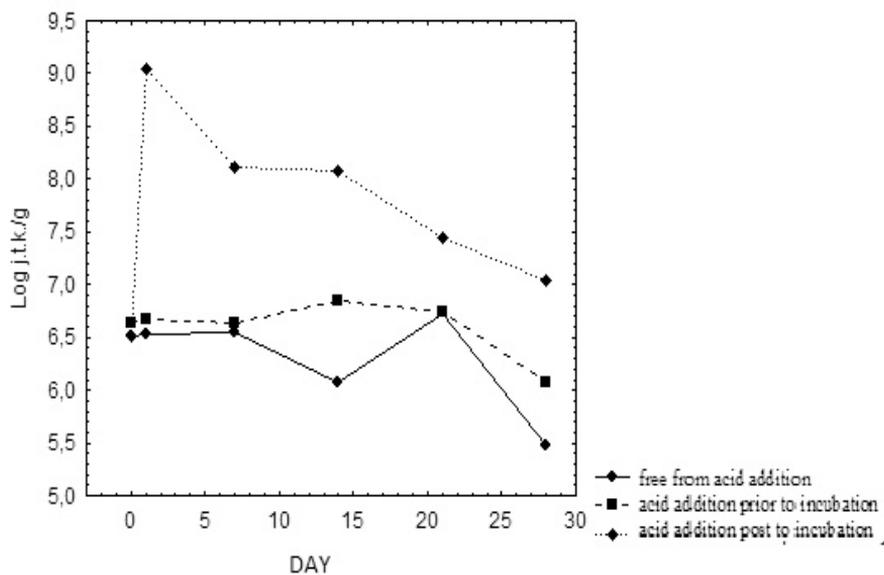


Figure 2. *Lbc. bulgaricus* changes at the time of yoghurt storage as dependent on the moment of L-ascorbic acid enrichment

Discussion of results

The results prove that addition of L-ascorbic acid influenced growth of yoghurt fermentation bacteria. Introduction of L-ascorbic acid after the process of yoghurt incubation produced markedly better effects. Yoghurt fermentation bacteria displayed intense growth during the first day of storage in samples enriched with L-ascorbic acid after the incubation. Luck H. (1952) recommends vitaminising yoghurt on fermentation as vitamin C losses of as much as 50% could be noted, caused by development of yoghurt micro-flora. Bacteria counts stabilised over the following seven days in the case of *Str. thermophilus*, to decline gradually afterwards. *Lbc. Bulgaricus* exhibited a different behaviour – its quantity fell dramatically in the first seven days of storage, to stabilise during the next 7 days and begin a gradual decline only after 14 days. This effect points to a time shift in the bacteria growth; *Str. thermophilus* continues in the state of equilibrium when the *Lbc. bulgaricus* population is decaying. In the case of yoghurts enriched with L-ascorbic acid post incubation, the quantity of *Lbc. bulgaricus* diminishes as early as the first day of storage, whereas the same effect comes later in the remaining sample types (enriched and not). Rajiv I. Dave and Nagendra P. Shah noted that *Str. thermophilus* and *Lbc. bulgaricus* in yoghurts produced by means of four commercially available cultures showed different responses to L-ascorbic acid content – the count of viable *Str. thermophilus* fell, whereas the quantity of *Lbc. bulgaricus* rose as L-ascorbic acid concentration increased (Dave, Shah 1997).

Addition of L-ascorbic acids to milk pre-incubation scarcely raised the count of yoghurt fermentation bacteria in the test yoghurts. Noh et al. also noted somewhat higher quantities of yoghurt fermentation bacteria in their yoghurts enriched with L-ascorbic acid and vitamin A than in control samples (Noh et al. 1994). B. Czarnocka-Roczniakowa and M. Wojewódzka maintain 10-20mg% content of L-ascorbic acid in milk has virtually no impact on growth of yoghurt micro-flora and progress of fermentation (Czarnocka-Roczniakowa, Wojewódzka 1969). Earlier research by the author implies incubation adversely affects L-ascorbic acid levels at the time of yoghurt production. Levels of L-ascorbic acid after the incubation stage in incubated goat milk enriched with lemon juice and analytical grade L-ascorbic acid and in unenriched milk declined by: 14%, 15% and 13%, respectively (Borek-Wojciechowska 1998).

Str. thermophilus and *Lbc. bulgaricus* in non-fortified yoghurts fell dramatically after the 21st day of storage. The decline of viable yoghurt bacteria after 21 days of storage was milder in non-enriched yoghurts.

Conclusion

Yoghurts are indicated as the most popular fermented dairy drink by Polish consumers. It must be emphasised that this opinion is shared across all consumer groups, from children, young people and students to adults. It can therefore be posited that these products have considerable effect on the health of Polish consumers. In effect, maintenance of their requisite quality, especially preservation of appropriately numerous and viable micro-flora, is of particular importance.

The results of the tests suggest that ascorbic acid can influence the amount of living bacteria both *Lbc.bulgaricus* and *Str.thermophilus*. Fortification after the incubation process has turned out to be more beneficial.

In light of results arrived at as part of the U.S. Human Microbiome Project, beside the Human Epigenomic Project the leading research project in the field of human genetics and molecular biology, the problem of yoghurt enrichment to provide adequate counts of yoghurt micro-flora may acquire a new significance in the near future. J. Fiedurek (Fiedurek 2014) relies on results of that project to say: "The human metabolism should therefore be seen as a result of evolutionary coexistence between the world of bacteria and a host organism. In this framework, expression of approximately 20,000 human genes and of an estimated 100 times greater numbers of bacterial genes make up an enormous, highly dynamic human metabolism determining function of the human body understood as a single, extraordinarily complicated "superorganism".

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THE ANCIENT CROP OF QUINOA FOR WORLD FOOD SECURITY

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Introduction

The growing interest in old food crop such as quinoa (*Chenopodium quinoa* Willd), is based on their nutritional value, high adaptability to different pedoclimatic conditions and remarkable yields. Quinoa belongs to the family of Chenopodiaceae and it is considered a pseudocereal. It is a native food plant of the Andean region dating back to 5000 years AD.

Quinoa originated in the Andean region of Bolivia, Colombia, Ecuador and Peru and was domesticated 3000 to 4000 years ago. This crop was so important for Andean peoples as food, animal feed and it was also considered sacred as the “mother grain” during their ceremonials. When Francisco Pizarro, a Spanish conquistador, and his army conquered the Andes (1532), they imposed to Inca communities Old World religion and habits like Catholicism, potatoes, wheat and barley. Despite the risk to have their hands cut off or to be killed for growing quinoa, the Incas continued its cultivation in hidden locations. It has been claimed that quinoa has been continuously cultivated in a given region longer than any other crop (Small, 2013). In the second half of last century, the interest in quinoa plant increased due to the “rediscover” of its nutritional values, its agronomic qualities and its great adaptability to different pedoclimatic and water conditions. The story of Quinoa is very similar to dozens of other crops, like *Amaranthus*, developed by Incas but overlooked by Spanish colonizers. Considering them cheap plants, ignoring their nutritional value and afraid of their ritual importance, conquistadors preferred to destroy to forbid them. Since a long time these crops have been revaluated and grouped in a new and large family called “Lost crops of Incas”. Even if the Spanish must be remember for their role in diffusion of “new crops” like maize, tomato, peanut, potato, etc.

Quinoa is an annual strong herbaceous plant, among over 200 varieties the most utilized is the *Real quinoa*, mainly in the brackish areas of the Salar,

in the Oruro and Potosi regions. The Incas and other earlier cultures appreciated Quinoa high nutritional value, including its leaves and seeds in their diet in order to balancing the lack of animal proteins. Quinoa seeds are gluten free, therefore valuable for celiac diet, rich in protein (14-15%) primarily lysine (50-54 mg/100 g), in minerals (calcium, magnesium, iron, copper, and zinc) and in vitamins. Quinoa seed oil (2-8%) has a high content of oleic (23-29%) and linoleic acid (48-52%) and also a remarkable % of squalene. Furthermore Quinoa is also an excellent source of starch useful in several food and no-food applications (cosmetics and pharmaceutical) because its seeds are small starch granules with low amylose content. Quinoa is also a good source of trimethylglycine, a substance with DNA protection, anti-aging and anti-cancer action (Abugoch James, 2009). Today quinoa is currently grown in Bolivia, Peru, Argentina, Chile, Colombia, and Ecuador. Its cultivation has been tasted in different regions of United State of America, Canada, India and Europe (England, Denmark, Finland, Greece and Italy) where despite the local pedoclimatic conditions, are quite evident positive adaptation results (Small, 2013; Jacobsen, 1997). The first two countries are the main producers and their 2012 total production was approximately equal to 82,500 t. Due to nutritional, economic, environmental and cultural value of Quinoa the United Nations (ONU) has declared 2013 as the “International Year of Quinoa” in order to remark its role as world food security crop.

The objective of this paper is to present a review of quinoa seed characteristics and to analyze its potentialities in rural and urban food market.

Botanical and agronomic characteristics of quinoa

Quinoa was first botanically described by Willdenow in 1778 as a native South America species, whose center of origin, according to Buskasov, is Bolivia and Peru. This plant belongs to Chenopodiaceae family, genus *Chenopodium* and its botanical name is *Chenopodium quinoa Willd.* As just mentioned before, more than 200 varieties are known and the *Real quinoa* is mostly used for its low saponins content. The other species commercialized are: *Bear*, *Cherry Vanilla*, *Cochabamba*, *Dave 407*, *Gossi*, *Isluga*, *Kaslala*, *Kcoito*, *Linares*, *Rainbow*, *Red head*, *Temuco*.

Quinoa may be classified according to the following parameters: a) morphological characteristics, according to the plant and fruit color or inflorescence types; b) great adaptability to grow at different altitudes, five ecotypes are recorded: sea-level, valley, subtropical, salar, and antiplanic; c) free saponins concentration, “sweet” quinoa less than and 0.11 %, “bitter” more than 0.11%. This crop is also known as “Inca rice”, “corn of the Andeans” or occasionally “vegetable caviar” (Jancurova et al. 2009; Vegas-Galvez et al. 2010; Small, 2013).

Quinoa is not a true cereal but being a dicotyledonous plant rich in starch it is included in the pseudocereal family along with amaranthus and buckwheat. Quinoa plant is 0.7-3 m tall, with an erect branched or unbranched cylindrical stem (1-8 cm in diameter). Plant color may vary from white to yellow or from light brown to red. If sown deep in the soil, quinoa roots could be 30 cm deep and besides its high growing flexibility at different altitudes (from sea-level up to 4000 m) quinoa is a frost and drought resistant plant. Low temperature (-1°C) are dangerous for quinoa flowers so it must bloom after frosting season in order to avoid plant and yield damages. High temperature instead (up to 35°C) is well tolerated by the whole plant. This crop has a low water requirement, so annual rainfall ranging from 100-400 mm is sufficient to make quinoa grow even if it has not problem growing in regions like southern Chile where annual precipitations are equal to 3000 mm. Quinoa is also a plant that is not affected by acid/alkaline soil (pH from 6.0 to 8.5) or by nitrogenous poor soil although different pedoclimatic conditions do have an impact over seed yield and consequently on their nutritional value (FAO, 2011; Jacobsen et al., 2005; Coulter, 1993).

Quinoa leaves up to 15 cm long and 12 cm wide, are alternate and dentate, with a “goose foot” shape, have different colors and a surface containing small grains that make the plant look like covered with sand. These grains are rich in calcium oxalate and are able to retain a water film, increasing the relative atmospheric humidity and reducing transpiration. Flowers are very small (30 mm) with no petals and positioned in groups forming glomerules. Blooming period is 10-15 days long. Inflorescence is a typical panicle with a central axis. It could be loose (or amaranthiform) or compact (or glomerulates), 30-80 cm long and its diameter ranges from 5 to 30 cm. From 100 to 3000 seeds are contained in the quinoa inflorescence. The fruit is an achene, cylindrical shaped with a single differently colored seed. The seed is obtained from quinoa fruit without perigonium. It may be conical, cylindrical or ellipsoidal shaped with a diameter ranging from 1.6 to 2.6 mm; 1 g of quinoa seeds contains approximately 350 colored (white, beige, light yellow, light brown, red or black) seeds (Vilche et al., 2003; Jancurova et al., 2009). They present a complex structure with three different parts: episperm (including pericarp, a protective covering and the endosperm), embryo and perisperm. The main starch reserve is in the perisperm and endosperm and embryo are rich of protein and fat. Compared to other seeds the embryo of quinoa has a higher amount of protein equal to 35-40% of the total seed proteins, while the perisperm only 6.3 to 8.3% (Abugoch James, 2009).

In general, depending upon the variety and pedoclimatic conditions, planting season begins in August-September and extends through November-December. Sowing density may vary from 0.4 to 2.3 g/m² and, on average, 140-180 days after the plants are ready to be harvested. In particular, 126-157 days are necessary under European conditions, 131-200 in Peru and 110-190

in South America (Pulvento et al., 2010). Quinoa yield shows a wide range from 0.45 t/ha up to 5 t/ha directly linked to genotypes, pedoclimatic condition and fertilizing and irrigation procedures. The average quinoa yield is 0.84 to 3 t/ha, whereas it is reported yields for quinoa as low as 0.45 t/ha to a record of 5 t/ha. Traditional producers country (Bolivia Ecuador and Peru) have a low average annual yield equal to 0.6-1 t/ha in the period 1995-2012. In different regions yields are higher, in USA and Canada they are more than 1 t/ha, in Italy 1.50-3.60 t/ha and in Kenya 4 t/ha (Ahamed et al., 1998; Pulvento et al., 2010; FAO, 2011).

Nutritional value and antinutritional substances of Quinoa seeds

Quinoa seeds earned a renewed dietary importance for their high nutritional value both in origin area and in “new market” like United States of America and Europe. Moreover, and as already mentioned, FAO recognizes to this crop a strategic role in the international food supply. Quinoa seeds are an excellent source of starch about 60%, they have good protein content in the order of 12-16% and their fat content varies from 5 to 8%. The ashes, mainly consisting of potassium and phosphorus, crude fiber are respectively approximately equal to 3-4% and 2-10%. The energy value of 100 g of quinoa seeds (on dry weight) is approximately 330-430 kcal. Compared with traditional cereals, quinoa seeds have a good level of B and E vitamins. The last one is the most important fat-soluble antioxidant vitamin.

The following table (table n. 1) shows the proximate chemical composition of quinoa seeds and leaves. Even if leaves present a remarkable nutritional value (particularly mineral content) their use as food and fodder is limited to local production area. In this note we focus on quinoa seeds.

Table 1. Main nutritional information of quinoa seeds and leaves (% on dry weight basis)

	Seeds	Leaves
Protein	12-16	21-22
Fat	5-8	3-4
Ash	3-4	18-19
Carbohydrate	60-75	34-35
Crude fiber	2-10	13-14
kcal/100 g	330-400	250-

Source: personal elaboration by the authors on data Prakash et al., 1993; Schlick, Budenheim 1996; Culter, 1993.

Quinoa seeds protein

Quinoa seeds protein content is very interesting because of its quantity and quality. Depending on varieties, it ranges from 13% to 22% (on dry weight) with an average of 15%, and it is higher than true cereals such as barley (11%), maize (10%) and rice (7.5%), comparable with wheat (15%), and lower than legumes such as beans (28%), lentils (28%), lupines (39%) (Jancurova et al. 2009).

It is significant to bear in mind that the larger portion of quinoa seed proteins are included in the embryo (35-40% of the total seed proteins) so for instance, quinoa protein content substantially coincides before and after dehulled treatments. On the contrary, traditional cereals like wheat loose in protein content depending on refined grade.

Table 2. Amino acids composition of quinoa seed, barley, soybeans, wheat and skimmed milk

Amino acids	Quinoa seed	Barley pearled	Soybean raw	Wheat durum	Skimmed milk
	mg/g protein				
<i>Essential</i>					
Lysine	54	37	58	36	97
Isoleucine	36	36.5	44.5	43	46
Methionine	22	19	10,5	23.5	26
Phenylalanine	42	56	49	53.5	46
Threonine	30	34	39	36	40
Tryptophan	12	16,5	12	11.5	14
Tyrosine	19	29	36	33,5	50
Valine	42	49	47,5	61	61
<i>Non-essential</i>					
Arginine	77	50	70	83	35
Aspartic acid	80	62.5	136	94	73
Cystine	14.5	22	12	20.5	9
Glycine	49	36	39	45.5	17
Glutamic acid	132	261	151	195	213
Histidine	29	22.5	27	23.5	29
Leucine	59.5	98	72	83	82
Serine	40	42	50	52,5	58
Alanine	41,5	39	42	58	28

Source: personal elaboration by the authors on data Schlick, Budenheim 1996; Abugoch James, 2009.

Quinoa seeds protein nutritional quality is based on the portion of essential amino acids which must be provided with the diet. The amino acid

content of the quinoa seeds is well balanced and offer a wider amino acids range than other traditional cereals. It is equivalent to what is recommended for preschool and school children and adults by Food and Agriculture Organization of the United Nations (FAO), World Health Organization (WHO) and United Nations University (UNU) in their final report Human energy requirements: Report of a Joint FAO/WHO/UNU Expert Consultation (FAO, 2001). Quinoa proteins have a balanced composition of essential amino acids similar to the amino acid composition of casein. Table n. 2 shows the proximate amino acids composition of quinoa, other traditional cereals and skimmed milk.

Quinoa is rich in lysine (54 mg/g protein) the limiting amino acid in most traditional cereals and rich in sulfur amino acids (methionine + cystine 36.5 mg/g protein).

Also in quinoa seed albumins and globulins are the large protein portion (44-77%), consequently prolamines percentage is very low. This makes quinoa seeds a natural gluten-free seeds valuable for celiac diet (Jancurova et al., 2009).

Quinoa protein efficiency ratio¹ (PER) and protein digestibility are influenced by plant varieties and washing and/or cooked temperature. Quinoa flour obtained with washed seeds in hot water (approximately 85°C) shows a PER index ranging from 2.1 to 2.3, almost like casein PER (2.5). Digestibility or amino acids bioavailability calculated for quinoa raw washed seeds is lower than casein respectively equal to 83% and 91% (Abugoch James, 2009).

Quinoa seeds carbohydrates

Quinoa seeds are mainly carbohydrates (see table n. 1) and starch represents more than 50-65 % of their total. Quinoa starch size and amylase content ensure a good starch source useful in food and no food industries as a biodegradable filler in polymer packaging and also due to extraordinary freezing & melting stability, a perfect thickener for frozen foods and other applications requiring high retrogradation resistance.

Located in seed perisperm, it can be as single unit or spherical aggregates whose diameter range from 0.6 to 2 µm. Quinoa starch granules are smaller than other traditional cereals like rice (3-8 µm), barley (3-8 µm) or amaranth (1-2 µm). The amylase and amylopectin percentage (on dry basis) is respectively 3.5-22.5% and 75-77.5%.

Other carbohydrates in quinoa seeds are D-xylose, maltose, glucose and fructose, which allows its use in malted drink formulations. Also, its content

¹ PER indicates the increase in weight per 1 g of protein included in standardized diet.

of D-ribose and D-galactose and maltose would result in a low fructose glycemic index (Ahamed et al., 1998; Bhargava et al., 2006; Vega-Gálvez et al., 2010).

Quinoa seeds lipids

Quinoa seeds also contain 1.8-9.5% of high quality edible oil, more than maize (3-4% on dry mass basis) and lower than soy (20-21% on dry mass basis). Several studies have conducted on quinoa oil fatty acid composition and all different results are related to different plant species and different pedoclimatic conditions. An average fatty acid composition is recorded in table n. 3. The main fraction is represented by the unsaturated one approximately 65-75%, where linoleic (48-52 %), oleic (23-30 %) and linolenic (5-8%) acids have the highest percentage. The saturated fraction is characterized by the prevalent presence of palmitic acid (9-11%). Main properties of this high quality edible oil are its high degree of unsaturation with a low polyunsaturation index (3.9-4.7) and its stability, due to the presence of E vitamins (a natural antioxidant agent). The first index is higher than most edible oils, such as soya bean oil (3.92), corn oil (4.65), and olive oil (0.65). In addition quinoa oil has a significant squalene content ranging from 3.4 to 5.8 %. Squalene is a terpenoid and a precursor of cholesterol biosynthesis, the oldest and most abundant natural source of squalene is shark

Table 3. Fatty acid composition of crude fat from quinoa seed, corn, and soy oil

	Quinoa	Soy	Corn	Amaranth
Saturated (%)				
Myristic C14:0	0.1-2.4	traces	traces	Traces
Palmitic C16:0	9.2-11.1	10.7	10.7	12-25
Stearic C18:0	0.6-1.1	3.6	2.8	2-5
Unsaturated (%)				
Myristoleic C14:1	1	-	-	
Palmitoleic C16:1	0.2-1.2	0.2	traces	
Oleic C18:1	22.8-29.5	22	26.1	19-35
Polyunsaturated (PUFA)				
Linoleic C18:2 (ω -6)	48.1-52.3	56	57.7	37-62
Linolenic C18:3 (ω -3)	4.6-8	7	2.2	0.3-2

Source: personal elaboration by the authors on data Ahamed et al., 1998; Prakash et al., 1993; Amicarelli, Camaggio, 2012.

oil (40 - 80%) although a lower quantity is also found in vegetable oils such as olive (0.3-0.7%). Another good vegetable source of squalene is amaranth grain (6-8%) (Ahamed et al., 1998; Prakash et al., 1993; Amicarelli, Camaggio, 2012).

Quinoa seeds minerals and vitamins

Quinoa seeds have the highest content of minerals among cereals. Table n.4 shows the mineral composition of quinoa whole and dehulled seeds, quinoa flour, oat, wheat and barley.

Table 4. Mineral composition of whole and dehulled quinoa seeds, quinoa flour, oat, barley and wheat (mg/100 g)

	Quinoa seeds		Quinoa flour	Oat	Barley	Wheat	Amaranths
	Whole	Dehulled					
Calcium	86.3	55.1	70-86	58	29	50	25-389
Phosphorous	411	405	22-462	734	221	467	655
Potassium	732	656	714-855	566	280	578	290-580
Magnesium	502	468	161-232	235	79	169	232-363
Iron	15	14.2	2.6-6.3	5.4	2.5	3.8	7-18
Copper	//	//	0.7-7.6	0.4	0.4	0.7	1
Zinc	4	4	3.2-3.8	3.11	2.1	4.7	4
Sodium	//	//	2.7-93	4	9	8	7-100

Source: personal elaboration by the authors on data Abugoch James, 2009; Amicarelli, Camaggio, 2012.

Table 5. Vitamin composition of quinoa flour, oat, barley and wheat (mg/100 g)

	Quinoa flour	Oat	Barley	Wheat	Amaranth
Ascorbic acid	4-16.4	//	//	//	3.36-7.24
Thiamin	0.29-0.36	0.763	0.191	0.5	0.17
Riboflavin	0.30-0.32	0.139	0.114	0.16	0.2
Niacin	1.24-1.52	0.961	4.604	5.9	3.6
Pantothenic acid	0.487	0.119	0.260	//	//
Folate total	0.184	0.056	0.023	//	//

Source: personal elaboration by the authors on data Ahamed et al., 1998; Abugich James, 2009; Vega-Gálvez, 2010).

Quinoa minerals due to their high values and bioavailable forms could meet the daily mineral need of human being of any age. Calcium, magnesium, and potassium for instance, are enough for a balanced human diet. Plant varieties, fertilizers and soil typologies have an impact on mineral

composition. Seeds dehulling procedure finalized to saponins removal has a negative effect because same minerals, mainly calcium and potassium are located in the removed external layer (Abugoch James, 2009; Amicarelli, Camaggio, 2012).

Quinoa seed vitamins content is also remarkable. The higher value of vitamin B6 and total folate (table n. 5) are enough to meet children and adults daily needs. Vitamin E, ranging from 2.6 to 5.4 mg/100 g (on dry weight basis), is significant being an antioxidant (Prakash et al., 1993; Vega-Gálvez, 2010).

Quinoa seeds antinutritional substances

The limit of quinoa seeds nutritional value is represented by few antinutritional substances. The fraction of saponins is the largest group of them, these substances consist of different compounds characterized by diverse structures containing a steroidal or triterpenoid aglycone and one or more sugar chain with a distinctive foaming point. These substances grant a typical better-tasting and they may be toxic if highly concentrated. They can be found in many plants and also, in low concentration, in spinach, asparagus, alfalfa and soy beans. They have been named after the soapwort plant (*Saponaria*) discover, the root of which was historically used as a soap.

Saponins have been found in all parts of quinoa plant, leaves, flowers, fruits and mostly in seeds coats. As just mentioned before, quinoa plant can be classified upon saponins concentration, variable upon variety and pedoclimatic conditions, from 0.1 to 5% (on dry weight basis) (FAO, 2011; Bhargava et al., 2006). Saponins have the natural function to protect the plant from pests and diseases and prior to consuming quinoa seeds must be separated from saponins by rinsing them with cold alkaline water or by mechanical abrasion. Alkaline water solution, extrusion or roasting process could reduce the saponins effects on quinoa seeds and flour. Saponins extracted from quinoa seeds could also be a valuable antifungal treatment, to inhibit for instance the growth of *Candida albicans* (Kuljanabhadgavad, Wink, 2009). There is also evidence of saponins increasing membrane permeability, a precious help process for food and drug assimilation.

Phytic acid is considered one more antinutritional compound because of its ability to make unavailable minerals mainly like zinc and iron, and a smaller amount of calcium and magnesium. Phytic acid may be found not only in quinoa seeds out layers, as in the case of other traditional cereals such as wheat and rice, but also in the endosperm in the reason of 10-13 mg/g (on dry weight basis) (Rouales, Nair, 1993).

Quinoa seeds uses, production trends and new markets

Quinoa seeds uses

Besides quinoa seeds main nutritional and antinutritional peculiarities, it is important to analyze their real and potential uses. As food the whole grain of quinoa is used in daily diet to replace rice when it is combined with meat or fish, in soups alone or with other traditional cereals and/or legumes. It can be toasted, popped and extruded to make snacks and breakfast line products. Quinoa flour is used in tortillas, bread, pizza and noodles.

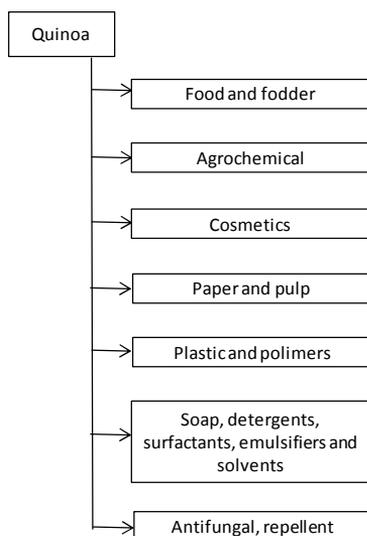


Figure 1 Quinoa traditional and potential uses

Source: Personal elaboration by the authors.

Adding quinoa flour to wheat one final products obtained improve their nutritional value (in terms of protein, oil, fiber and minerals) and it has also recorded a positive effect on rheological characteristic of dough. For instance, it is recorded an addition of 10-20% of quinoa flour in bread preparation, 30-40% in noodles and pasta and up to 60% in biscuits recipes. These types of products receive by panelists a good sensory evaluation (Jancurova et. al., 2009; Stikic et al., 2012). Quinoa seeds could be sprouted too to improve their nutritional quality increasing the bioavailability of vitamins and other biocompounds. Cereals and other seed sprouts reached greater popularity among consumers more and more consciousness about the strict relation between health status and dietary habits. In this field quinoa is an excellent option to enlarge the offer of these kinds of products on the market of

functional foods where, foods containing biologically active components with a potential in reducing the risk of disease are largely required. Quinoa ground seeds could be used to prepare drinks such as quinoa juice, mix with milk and quinoa fermented drinks . Other uses of this plant are linked to saponins and starch. Thanks to their properties this compound could be exploited in several industries such as agrofood, cosmetics, pharmaceutical and agrochemical. Figure n 1 summarizes traditional and potential utilizations of this ancient crop.

Quinoa seeds production trends and new markets

Bolivia and Peru have always been the main quinoa producers accounting, in the last fifteen years the 90% of world production. Table n. 6 shows the data related to area harvested, production, plant and seed yields from 1995 to 2012. Other traditional producers countries are Chile and Ecuador. There are no official figures on Chilean production before 2009, during the last three years, its contribution range from 1 to 2% of the whole quinoa production. Small quantity (approximately less than 2% of the total) is constantly produced in Ecuador.

Table 6. Quinoa area harvested, production, plant and seed yields from 1995 to 2012

	Area harvested (Ha)			Production (t)			Yield (t/ha)			Seed (t)		
	Bolivia	Ecuador	Peru	Bolivia	Ecuador	Peru	Bolivia	Ecuador	Peru	Bolivia	Ecuador	Peru
1995	36790	800	18729	18814	408	13773	0,51	0,51	0,74	562	47	485
1996	37493	1100	18704	23498	555	16070	0,63	0,50	0,86	580	31	547
1997	38680	600	27033	26366	304	23688	0,68	0,51	0,88	566	62	419
1998	37714	1800	30720	20291	938	28171	0,54	0,52	0,92	539	62	468
1999	35963	1800	28979	22498	938	28413	0,63	0,52	0,98	553	47	440
2000	36847	1300	28889	23785	650	28191	0,65	0,50	0,98	558	25	435
2001	37223	650	25601	23299	320	22267	0,63	0,49	0,87	567	23	425
2002	37817	600	27851	24179	294	30373	0,64	0,49	1,09	574	8	422
2003	38289	1000	28326	24936	519	30085	0,65	0,52	1,06	580	9	434
2004	38649	918	27676	24688	641	26997	0,64	0,70	0,98	590	33	431
2005	39302	929	28632	25201	652	32590	0,64	0,70	1,14	636	33	431
2006	42431	950	29947	26873	660	30429	0,63	0,69	1,02	682	33	449
2007	45454	980	30381	26601	690	31824	0,59	0,70	1,05	696	34	331
2008	46369	1000	31163	27169	741	29867	0,59	0,74	0,96	899	29	400
2009	59924	1100	34026	34156	800	39397	0,57	0,73	1,16	945	37	600
2010	63010	1176	35313	36106	897	41079	0,57	0,76	1,16	972	40	710
2011	64789	1277	35475	38257	816	41182	0,59	0,64	1,16	945	40	770
2012	63000	1250	38495	37500	800	44210	0,60	0,64	1,15	945	40	770

Source: personal elaboration by the authors on data FAO, 2014.

More recently, quinoa production expanded from native regions to new area. Quinoa was introduced to England in the 1970s, same years later in Denmark. Since the middle of 1980s United State of America (USA) cultivated quinoa firstly in Colorado Rockies and now in California, Oregon, Nevada and Washington. In 1993, the project “Quinoa—A multipurpose crop for EC’s agricultural diversification,” was sponsored by European Union, including England, Denmark, the Netherlands, and Italy, Scotland and France. Further countries, Poland, Czech Republic, Austria, and Greece have recently shown growing interest in this crop and Australia, Morocco, Kenya, China, Netherlands are organizing and testing the adaptability of this crop to their pedoclimatic conditions. Italy for instance, during a specific project had evaluated different yields of different genotypes planted in the Volturno river plain (Campania regions – South part of Italy) receiving good results in terms of annual yields and seeds quality (Jacobsen, 2004; Pulvento et al. 2010).

According to the figures included in table n. 6, the area dedicated to quinoa crop increase of 55% from 1995 to 2012. In the same period plant and seeds production rise respectively of 40 and 62%. This increment is due to the growing consume of quinoa out of the production area.

Quinoa global trade recorded a deep evolution from the year 2000, when less of 2000 t were exchanged, up today. World export rapidly expanded passing from 5000 t in 2005 to 40250 t in 2012 mainly supplied by Bolivia (64%) and Peru (27%). Quinoa export value has been increased passing from 13 to 79 million of USA dollars (USD). United State of America is the third exporter but its real role is a re-exporting countries. On a total USA quinoa export equal to 3350 t in 2012, only 728 t are domestic produced. In 2012, USA was the first quinoa importer (approximately 23000 t.) followed by Europe (less than 7000 t) and Canada (more than 2300 t.). Europe main importer countries are France (more than 2701 t), Netherlands (less than 1700 t) and Italy (less than 300 t). Actually, quinoa export average price is approximately equal to 3000 USD more than double compared with 2007 (Krivonos, 2013).

Conclusion

The proposed review of quinoa nutritional value and its botanical and agronomic characteristics explains and justifies the growing international interest in this ancient crop.

Quinoa seeds are rich in protein (14-15%) primarily lysine (50-54 mg/100 g), in minerals (calcium and magnesium) and in vitamins (B, C, E). Its oil fraction (1.8-9 %) has a high content of oleic (23-29%) and linoleic acid (48-52%) and also a remarkable % of squalene. Furthermore quinoa is also an excellent source of starch, with small granules and low amylose content,

useful in several food and no-food applications (cosmetics and pharmaceutical). Its grain is gluten free useful in celiac diet. The number of celiac enlarging year by years and the related food market need good and cheap gluten free products. In several European countries the interest in this cultivation is closely linked to this problem, in Italy for instance celiac is defined by low a social disease and affect 1 person out of 100-150 (GURI, 2005). Quinoa is also a good source of trimethylglycine, a substance with DNA protection, anti-aging and anti-cancer action. The presence of antinutritional substances like saponins and phytic acid do not limit quinoa utilization on the contrary offer new opportunities such as quinoa saponins application as phytotherapy and chemotherapy agents.

Quinoa great adaptability to different pedoclimatic, water, soil and fertilizers conditions, is making possible its diffusion outside the native area of South America. Annual yields in USA, Europe and Africa (1.5-4 t/ha) are higher than Bolivia, Peru and Ecuador (0.6-1 t/ha).

As just mentioned before, the United Nation General Assembly has declared 2013 the International Year of Quinoa. This has been a prestigious award for the Andean people who have continued to cultivate and protect this ancient high nutritional value crop. Furthermore, with this declaration UN has focused the world's attention on the role of this pseudocereal related to world food supply, poverty mitigation and biodiversity safeguard. The world population is growing, it is estimated that, in 2050, there will be 2 billion more people so quinoa – with other lost crops of ancient civilization - could be a good option for world food security. New projects are organized and sponsored by FAO to improve quinoa knowledge (select best genotype), quinoa yields (improve Andean annual yields) and quinoa cultivation economy in native and new world regions.

Quinoa increasing export remark its importance in developed countries where nutraceutical and functional food market segments are rapidly enlarging due to the changes in eating habits and the growing attention and consciousness of final consumers. This crop are changing its role from subsistence to high value crop. The consequence is that there is a lack of correspondence among quinoa export price, local Andean farmer incomes and retail prices. It is recorded that, in Oruro (Bolivia) from 2008 to 2013 the bread price is not change while quinoa grain double. Bolivian people lose in money and in high value nutritional intake. These would be the basic considerations of a new international agricultural policy by which decision makers have to be engaged to find the right equilibrium among domestic and international food supply and economic balance.

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This work is the result of the authors' commitment, starting from the idea and ending with its accomplishment. Particularly, introduction, conclusion and references collections are ascribed to G. Camaggio and sections 1,2,3 and references collection are ascribed to V. Amicarelli.

THE INNOVATIVE CHARACTER OF NANO PRODUCTS IN CONSUMER REVIEWS

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Introduction

Product innovations became a base ground for the expansion of modern market economy and with that commodities defined as the science of quality. Activities of this type are growing in popularity, as well as their understanding throughout the world. According to the generally applicable definition, innovations are all actions of scientific, technical, organizational, financial and commercial nature that are designed to achieve the implementation of innovation (Rudzewicz, Strychalska & Rudzewicz 2013).

New techniques and technologies are an integral part of an expanding consumer market by providing a basis for the design and introduction of innovative products. The definition of innovative products is perceived by the consumers in two ways. According to research by Zabrocki at the turn of 2011/2012 year, which included a group of around of 600 responders for an innovative product is considered:

- new product that had never appeared on the market (62%),
- the product known, but modified (23%),
- a product of a new manufacturing technology (approximately 10%) (Zabrocki, 2014).

Nanotechnology is recognized as one of the most innovative fields of science and technology. Its interdisciplinary nature appears to create a basis for development of previously impossible solutions to the functionality and utilization of the products. The final result can be obtained only through a combination of selected areas of chemistry, biology, physics, engineering, computer science, electronics, materials science and engineering (Szewczyk, 2011; Idzikowska, et al., 2012).

Most significant impact on the contemporary concept of nanotechnology has had the history of its development. It is believed that Richard Feynman was the precursor of nanotechnology. In his work he presented a vision of

possibilities in manipulating the molecular particles and atoms. Since 1959 Feynman was introducing an idea of the miniaturized computers, specialty wires and other materials that could be used by medical science[5]. Others seem to believe that the father of nanotechnology is Albert Einstein, who in his PHD thesis calculated the size of a sugar molecule, giving it a size 1 nanometer. In fact, this small size is 10^{-9} meters. This unimaginably small size can be illustrated as 10 hydrogen atoms arranged one behind another (Sobczak, 2003, Sokół 2012).

According to one of the widely accepted definitions, a nanomaterial is a material in which at least one structural element has a size less than 100 nanometers. As a result of operating in the nanoscale created materials can obtain unique properties that are impossible at the macro level (eg, better chemical, electrical, mechanical or optical qualities). For example, the reason for such change may be an increase of the external surface of the material as compared to the same materials on the macroscale (Świątek - Prokop 2012; Sokół 2012; Szewczyk 2011; Świdorski, Waszkiewicz & Robak 2006)

The introduction of innovation in nano products gives plenty of opportunities and benefits for many branches of the modern market. The process of development of new technologies is also accompanied by legal and social problems. A very important aspect is to ensure the safety of human life and health, as well as protection of the natural environment. Therefore, one of the most important issues is definition of clear legal regulations. Current lack of precise legal regulations concerning the use of nanomaterials is very evident. While in European Union legislation introduced specific rules governing the use of nanomaterials in cosmetics, biocides and food, but it still lacks legislation defining nano products as a distinct group of products. Decision of the Council of Europe on December 3, 2013 has been derived from the framework program for research and innovation for 2014-2020. The result of the work will be overcoming the ambiguities associated with the safe use of the products generated by the capabilities of nanoscale, and same develop precise and clear laws. This document clearly indicates the areas in which required intensive research on the management of nanotechnologies, provide validated scientific tools and methods relating to hazard, exposure and risk assessment, and management of nanomaterials throughout the life cycle of the product (Idzikowska, et al. 2012; Jurewicz 2013; Dz. U. L 347 z 20/12/2013, str. 965—1041).

Actions of individuals interested in the development of products at the nanoscale, resulted in the fact that consumer can find more than 1000 identified nano products on the current market. With innovative properties derived from nanoscale began to form products with new properties and applications (Table 1.). In 2011, the market value of nano products was 4.18 billion dollar. It is estimated that in 2025 its value will be 100 billion dollar. For example, one of Australia's bakery produced bread with tuna nanocapsules oil (rich in omega 3), which resulted in overcoming the

persistent smell of fish (Błaszczak, Jasiczak 2013; Nowak 2008; Świątek - Prokop 2012).

Table 1. Examples of possibilities to use the nano materials and nano products

Industry or science	Application	Additional features as a result of use of nanoscale
Medicine	Dressings, bandages, plasters on the wound with disinfecting layer of silver nanoparticles.	The antibacterial effect may last more than a week, due to the controlled release of silver nanoparticles.
	Screens, curtains, clothing.	Made of fibers and materials impregnated with nanoparticles of silver and copper. Exhibit biocidal properties; helps to limit the possibility of hospital-acquired infections through physical contact with the such materials as well as patients.
	Nanotubes covering implants.	Increasing the surface of the congruence.
Cosmetology	Soap enriched with silver nanoparticles.	Elimination of bacteria.
	Facial creams enriched with nanostructures such as gold, silver, copper, and platinum.	Exhibits a greater efficiency thanks to the active substances that get to where they need to go, for example, into the deeper layers of the skin
Construction Industry	Antibacterial paint with silver nanoparticles.	Used to protect the walls against the growth of microorganisms.
	Glass with self-cleaning or fog-free properties.	Used eg for fireplaces.
Textile Industry	Socks made of fibers coated with silver nanoparticles.	Limitation of unpleasant odors.
	The nanoparticles of soot can form an anti - static membrane.	The water-proof biproduct that allows steam to go thru, but is efficient in diffusion of static electricity.
Food Industry	Deposition of nano silver onto the filters placed in air conditioners working in meat processing plants.	Air purification from microorganisms that could possibly contaminate production halls.
	Packaging of food products.	By using nanoparticles of silver and copper in modification of polymers which are the basic of raw material for the packaging materials production. This allows significant extension of their storage life and thru that reassures safety of their consumption. Silver nanoparticles fill the whole mass of plastic packaging and assist in eliminating any bacterial flora, mold and other fungi.
Electronic	Graphene battery in a flexible, electrical -resistant coatings.	The most effective natural conductor of electricity; considered the most durable material in nature.
Motorization	Extreme lightweight and durable materials (polymers, nano fibers).	Enables pretty much any of modern construction solutions.
	Car care products with addition of nanoparticles.	Resistance to dirt and shine for a long time.

Source: Own research by: Pulit et al., 2012; Nowacka & Niemczuk, 2012; Szymański, 2012; Sokół 2012; Szewczyk, 2011; Schlecht & Schroeder, 2010

The scope of possibilities for application of nanoscale is not only food production, but also in packaging technology, chemical industry, electronics, aeronautics, medicine, nursing, cosmetology, construction, automotive, and environmental protection (The Project on Emerging Nanotechnologies).

Very fast growing market of nano products makes the demand for the materials also increases. A specific need for nano materials, for the electronics and medicine (Table 2.).

Table 2. Projected demand for the use of nanoscale achievements in 2011 and 2025

Expected market	Sector demand for nano materials in 2011	Sector demand for nano materials in 2025
Medicine	50%	35%
Electronics	22%	37%
Structural parts	21%	21%
Other	7%	7%

Source: Świątek- Prokop, 2012;

The discussion about price competitiveness is pointless while introducing nano products to the consumer market. These products require higher production costs compared to traditional products. The only chance to obtain a market advantage is to persuade consumers to the functionality of these products. Consequently, the market growth rate will be determined by the attitude of the consumers to nano products (Błaszczuk, Jasiczak, 2013).

The following study is an attempt to assess consumer attitudes toward placement of nano products on the market and the factors determining their attitudes, as well as purchasing decisions.

Materials and methods

The study was conducted in the form of a diagnostic survey with the use of a questionnaire survey consisting of 14 questions.

The questionnaire form has been arranged in four parts assembled with closed questions once and multiple-choice. The scope of research was to determine:

- the level of respondents' knowledge about the basic concepts of nanotechnology and nano products,
- consumers' attitudes toward nano products,
- the frequency of purchase and use of nano products by consumers,

- the source of knowledge and the willingness of respondents to its expanding.

It was assumed that the end result of research would be definition of the knowledge level and awareness of the respondents regarding nanotechnology and nano products. Part of the research findings includes definition of the determinants and consumer behaviors.

The study group consisted of 300 randomly selected consumers from the area of the Tri-City and surrounding areas.

Results and Discussion

As a result of the survey, it was found that Polish consumers correctly indicate the meaning of the term nanotechnology. Almost 73% of them indicated that nanotechnology is a technology based on particle sizes millionths of a millimeter. One quarter of the respondents indicated that nanotechnology is the science dedicated to the development of innovative products. Only 3 respondents (women) have linked the given definition of science dedicated to the development of nuclear weapons.

Consumers rated their level of knowledge about nanotechnology and nano products as low. To complete lack of knowledge admitted 15% respondents. More than half of those surveyed believe that they know little about nanotechnology and nano products. A high level of knowledge was declared by 15% of all respondents (Figure 1.).

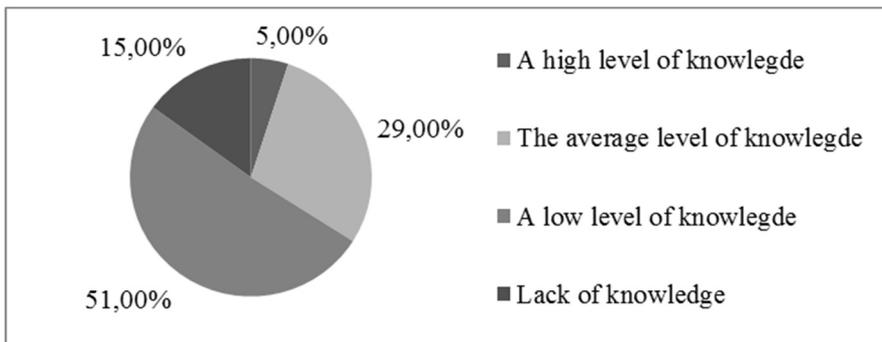


Figure 1. Self-assessment of the level of knowledge nanotechnology and nano product

Source: Own research

The primary objective of the survey was to investigate consumers' attitudes toward nanotechnology and nano products. A positive attitude toward innovative technology declares 42% of respondents, including the far greater number of men. Their positive attitude motivate consumers primarily

interested in the novelty (80%). Almost 90% of respondents believe that nanotechnology, by interacting with all sectors of the economy, creates the potential to produce a range of materials with new properties. The same percentage of people indicated, that the development of nanotechnology - based products simplified their daily life. Only 9 respondents indicated their negative attitude toward technology based on nanoscale. This attitude was motivated by negative impact of nano products on environment (17%) and fear of contributing into innovations in weapons technology (45%). Only 6 respondents explained their negative attitude with the fear of deterioration of their lives by the use of nano-products. Surprising was the fact that despite the high level of positive attitude towards nano products (Figure 2.), about 42% of respondents believe that nano products are not fully examined, so their use puts the consumer at risk.

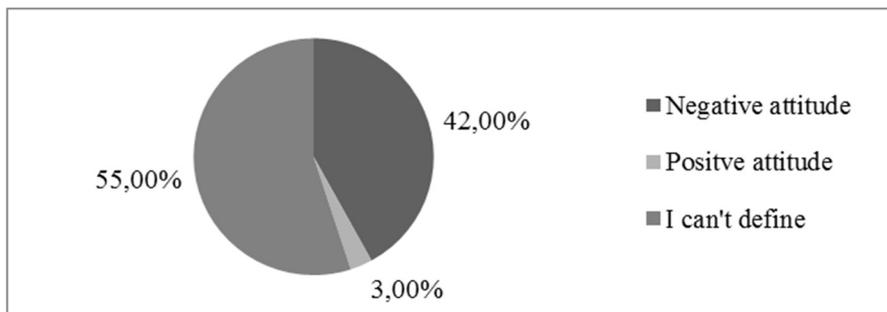


Figure 2. Attitude toward nano products

Source: Own research

In the next stage of the research, consumers were asked to declare their attitude in case of real contact with nano product in the store. Almost 90% of respondents declared interest in nano products, taking into account their possible purchase. Despite the relatively high level of positive attitude towards nano products (Figure 2.) consumers declared that they are making their purchases very carefully, only 5% of them would buy nano products consciously (Figure 3.). Fear of the news was confirmed by 21 respondents.

Among the study group of respondents, only 30% of them consciously often buy nano products. It may be noted that a significant group of people buying these products are often consumers with a better financial situation from the major cities. Quarter of respondents motivate their choice by the fact that they are at a convenient price and have better properties than for conventional nano products counterparts (Figure 4). For occasional purchase of goods produced by nanoscale admits 35% of respondents. The same percentage of people will never knowingly buy nano products. This may be due to the fact that one quarter of respondents do not believe in properties resulting from nanoscale (Figure 4). Almost half of the respondents believe that nano products are expensive.

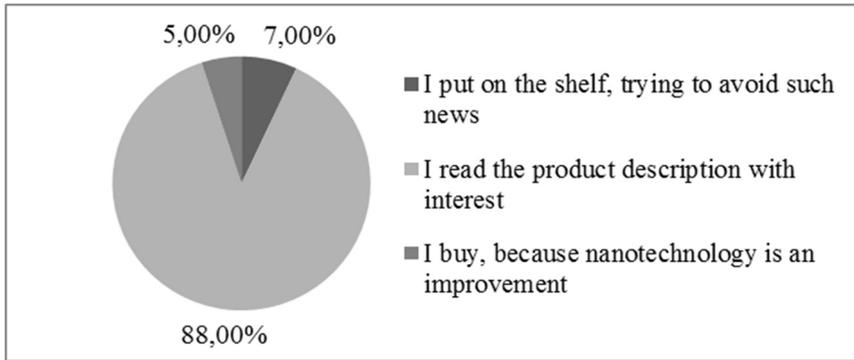


Figure 3. The reaction of the respondents in the case of real contact with the nano product

Source: Own research

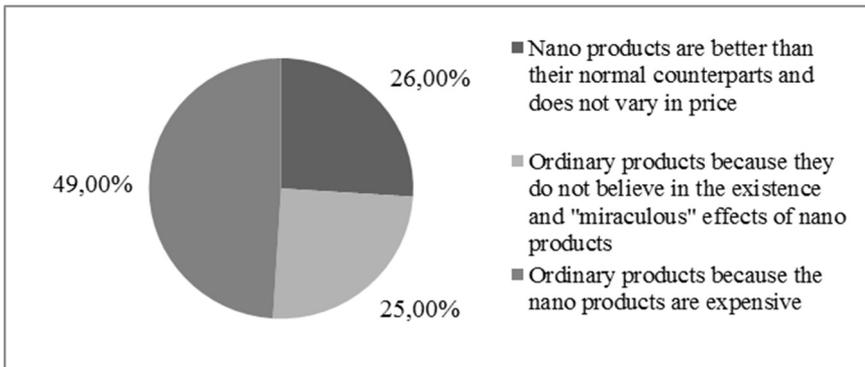


Figure 4. Purchasing motivation of the respondents

Source: Own research

Respondents declare a fairly high degree of knowledge about nano products currently available on the market. About 60 respondents know nano products, such as car wax/paste, cleaning cloths and mops containing nanofibers and nanoparticles. Significant number of people from the test group indicated that the idea of cosmetics based on nanosilver antibacterial patches of silver nanoparticles, creams for muscle and joint pain, and easily washable fabric with nanoparticles is not foreign to them.

The majority of respondents (80%) asked about the likelihood for the use of nano products, most want to use mobile phone with high quality and clothing with high resistance to stains. It may be noted that people with a higher level of knowledge and higher income were more likely to apply examples of nano products than people with lower levels of knowledge in the field of nanotechnology and nano products. The least useful by respondents

nano products turned out to be the soil of nanofibers eliminating disease-causing pathogens from the environment. Respondents indicate a clear interest in ecological solutions. For example, 60% of them are very willing to use bus, which are loaded batteries through nanotechnology within 20 seconds the vehicle.

Consumers participating in the survey used the information from sources such as the internet (83%), TV (37%), university (36%), newspaper (31%).

The analysis of responses showed that consumers have a basic level of knowledge in the field of nanotechnology and nano products. The knowledge that they use comes mostly from unreliable sources of information. Respondents indicate the negative attitude toward nano products and nanotechnology is an evidence inconsistent knowledge base about this matter. However, respondents indicate quite open attitude toward knowledge about the topic of nanotechnology and nano products. Evidenced by the fact that more than half of consumers eager to broaden their knowledge about nanotechnology. Lack of decisiveness in this aspect was shown by more than 30% of respondents.

Analysis of the literature references confirmed the above statement. A clear trend of increased knowledge and interest in nanomaterials can be noticed at the level of the global product markets. Studies conducted in the United States and Europe confirm that consumers have little knowledge about nanotechnology and products manufactured by using nanoscale. Respondents from more developed countries such as the United States have more open attitude to innovative technology while seeking to facilitate everyday life (Łopacka, Póltorak, 2013; Siegrist, 2008).

Conclusions

1. Consumers have a basic knowledge of nano products and nanotechnology and show a high level of willingness to broaden that knowledge.
2. Respondents are characterized by a seemingly positive attitude to nano products and nanotechnology. Despite the declaration of a positive attitude, their purchasing choices are cautious, thus demonstrating social resistance to innovation. It can be assumed that this position will change with the increase of knowledge in their areas and the introduction of public information campaign.
3. Respondents indicate the existence of a clear economic barrier that restricts the purchase of nano products. Adjustment of their prices to the financial capability of consumers could lead to increased interest in these products and overcoming of the barrier of fear of innovation.

Summation

The interdisciplinary nature of nanotechnology makes innovative products pose potential for the development of global markets. Fairly quick pace of the new generation of novel functional properties and functional, however, faces a number of barriers to the legal, economic and social. The next few years will be significant for the development of this product sector. For this reason, a very important role to play to ensure the proper transmission of reliable information to potential consumers and users, which can largely overcome widespread fear of innovation.

The consumers are seen as an important element influencing the increase in demand and should be treated as a key element in the development of innovative products. In the absence of reliable information sources regarding possible use of nanotechnology, we may experience a phenomenon of general lack of acceptance of nano products. The same effect also accompanied introduction of genetically modified foods (GMO) to the market. The fear of the new as well as their fear of potential negative effects of the use of nano products may affect their popularity and consequently the development of this sector of the market considered as innovative (Babicz – Zielińska & Zabrocki, 2007).

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FLAVOUR PROFILES AS INDICATORS OF BOTANICAL ORIGIN OF HONEY

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Introduction

In the face of continuing production globalization, the issue of food product authenticity constitutes one of the most dynamically developing food analysis research trends. It is a difficult, time-consuming, and expensive process to confirm the authenticity or to detect adulteration of food products.

As for honey, authenticity verification is linked with their varietal identification; therefore, this research study is an attempt to determine the varietal authenticity and origin of honey that would allow identifying the botanical origin of honey with the use of a gas chromatography

For many years, the varietal identification of honey has been a research subject in many scientific centres (Bogdanov 2007; Bogdanov & Gallmann 2008).

Pollen analysis has been the most often applied method when identifying varietal honey. It is a traditional method used to confirm the biological origin of honey (Louveaux et al. 1970; Sawyer 1988; Feller-Demalsy & Parent 1989; Bambara 1991; Faegri & Iversen 1993; von der Ohe et al. 2004; ; Skubida et al. 2005; Bogdanov & Gallmann 2008).

Alas, this method is highly time - consuming and depends strongly on the skill of professional experts. Over recent years, this method has also been applied together with specialist statistical software. However, since the content of various pollens in honey is very large, this method is now used as a supporting one, collaterally with the sensory and physical-chemical analyses. Honey bees collect floral pollen from various plants, thus, pure mono-pollen honey are very rarely encountered (Bogdanov & Gallmann 2008).

Sensory analysis is also used in the varietal identification of honey (Serra Bonvehi & Gomez Pajuelo 1988; Kerlvliet 1992). But this method is considered as subjective, so, many researchers endeavoured to use the analysis

of physical-chemical parameters of honey in the varietal and geographical identification of honey (Sanz et al. 1995; Marzec 1998; Popek 2001).

Amidst all the physical-chemical parameters of honey quality, specific electrical conductivity appeared to be the most effective when identifying varietal honey. This parameter is mainly used to distinguish some varietal nectar honey from nectar poly-pollen and honeydew honey (Kubišova & Mastny 1976; Sancho et al. 1991; Salinas et al. 1994; Salinas et al. 1994a; Popek 1998).

No anticipated results are reported as regards researches comprising honey identification based on pigments in honey, chiefly flavonoids (Tisse et al. 1994; Bogdanov 1989; Meda et al. 2005), or on colour parameter measurements in L a* b* and X Y Z systems (Ortiz Valbuena & Silva Losada 1990; Piazza et al. 1992; Giemza 1999).

Another approach points to a chemometric analysis of results obtained from searching into physical-chemical parameters such as: content of saccharides, concentration ratio of glucose to fructose, contents of nitrogen, sucrose, 5-hydroxymethylfurfural, ash, and water, or from determining the pH level, total acidity, specific rotation, or from the analysis of aromatic acids and amino acids (Krauze 1991; Pirini et al. 1992; Pena Crecente & Herrero Latorre 1993; Persano Oddo et al. 1995; Ojeda de Rodríguez et al. 2004; Serrano 2004; Sanz et al. 2005). The combination of the above named parameters makes it possible to recognize some mono-floral honey (Pena Crecente & Herrero Latorre 1993; Persano Oddo et al. 1995; Bogdanov 1997; Piro et al. 2002; Terrab et al. 2004). Yet the results of those analyses are not satisfactory, because it is not possible, based on those results, to classify all honey according to their individual types and varieties (Bogdanov 1997; Piro et al. 2002; Terrab et al. 2004).

Some of the methods to analyse volatile fractions in honey facilitate the discrimination of honey of different botanical origin. One of those methods is the extraction of volatile compounds with the use of n-pentane by ultrasound (Alissandrakis et al. 2003). Presently, markers are tested in order to choose those that might be applied to identify honey using gas chromatography coupled with mass spectrometry. The search results obtained seem to point to the existence of some compounds that could be helpful when identifying the origin of honey (Radovic et al. 2001; Agric et al. 2007; Glory-Cuevas 2007). The results of analyzing honey originating from regions in Greece suggest that the developed discrimination and classification methodology is useful for the botanical classification of honey samples and for the assessment of differences in their composition (Aliferis et al. 2010).

The application of near-infrared and mid-infrared spectroscopy (Ruoff et al. 2006; Ruoff et al. 2006a). includes basic analysis of spectra that makes it possible to distinguish honeydew honey from floral honey; additionally, with

the use of specialized software (TQ Analyst), varieties of floral honey are determined (Benson 2003; Bogdanov et al. 2004; Persano Oddo et al. 2004; Sadlej 2004).

The research carried out by Irudayaraj proved that FT-MIR spectroscopy could be successfully applied when assessing the authenticity of food products including honey (Sivakesava & Irudayaraj 2002; Tewari & Irudayaraj 2004).

A further method is isotope analysis known as IRMS (Isotope ratio mass spectroscopy). The content of isotopes in honey is correlated with the climate in a given region, but it can also be correlated with the composition of rainfall (Kelly 2003; Piasentier et al. 2003). IRMS is based on the knowledge of the ratio of isotopes that are characteristic for individual plant species; it consists in determining the quantity or the ratios of isotopes of one of the three basic elements occurring in living matter: $^{13}\text{C}/^{12}\text{C}$, $^{18}\text{O}/^{16}\text{O}$, $^2\text{H}/^1\text{H}$, and in comparing them to standard values (Przetaczek-Rożnowska & Rosiak 2011).

However, many times the results of those often complex analyses, which, also, require expensive equipment, do not permit that the authenticity of honey be confirmed or contested. Thus, other methods are suggested. They involve measurements of a couple or a dozen physical-chemical characteristics of honey and mathematical-statistical analyses of the parameters measured (for example: variation analysis, canonical analysis, analysis of key components, multidimensional analysis, taxonomic analysis, and discriminant analysis) in order to choose a few characteristics of honey. The selected characteristics will be then considered for an optimal varietal or geographic distinguishing feature (Krauze & Zalewski 1991; Pena Crecenta & Herrero Latorre 1993; Salinas et al. 1994; Salinas et al. 1994a; Sanz et al. 1995; Anklam 1998; Persano Oddo et al. 1988; Poppek 2001; Piro et al. 2002; Devillers et al. 2004; Terrab et al. 2004).

Upon reviewing the reference literature, it can be concluded that a complex evaluation of varietal authenticity of honey is an outcome of the analysis and interpretation of the measured organoleptic and physical-chemical parameters as well as of the pollen analysis results (Persano Oddo & Bogdanov 2004).

The analysis of the reference literature available confirms one fact: although so many diverse honey identifying methods are applied, it is still necessary to improve them or to develop a more effective method to better and more successfully identify the type and variety of honey, and this issue is the objective of the present research study. To achieve the objective set, an postulation by Anklam was assumed, i.e.: for the purpose of developing and increasing the potential of applying authenticity evaluation techniques and of rising their effectiveness, it is indispensable to bridge various scientific fields and to expand joint co-operation in interdisciplinary fields (Anklam 1998).

Materials and methods

The experimental material in the present research study consisted of 72 samples of honey belonging to the variety types (nectar (from rape, acacia, heather, linden, buckwheat, and multi-floral nectar from various plants) honeydew, and nectar-honeydew). The honey samples analyzed were produced in apiaries located throughout Poland, and each sample was from a different apiary.

The botanical origin and the purity of honey samples were monitored using a savouriness profiling method developed by Cairnocris and Sjöström and modified by Tilgner (Tilgner 1962), and a pollen analysis (Szczęsna & Rybak-Chmielewska 1993).

Results of chromatographic studies and analysis thereof

Sensory quality, and, in particular flavour and taste, is a very significant factor from the point of view of the consumer making a purchase decision. Flavour is a significant distinguishing feature of quality; also, it can be a characteristic of a variety/type. Flavour is a feature of volatile substances (or substances containing volatile substances) perceived by the sense of olfaction. Every food product is characterized by an individual and unique set of volatile compounds, which make up its flavour (Czapski et al. 1999). The character of flavour that is typical for an individual foodstuff depends on many factors, as well as on the reactions ongoing between the components thereof. Volatile compounds in foods are usually a mixture of several tens or hundreds of compounds present therein in different concentrations (from ppt to ppm). From a vast number of volatile compounds identified in various foodstuffs, only some of them determine the flavour of a food product. It is a complex process of how a person perceives a particular flavour. For example, depending on his/her initial connotation of a given flavour, the same person may perceive it differently when he/she repeats the assessment of the same flavour. Most frequently, flavours were, and still are evaluated by experts. However, new methods of assessing flavours have appeared along with the developing science. Instrumental analysis has become an excellent addition to the assessment of organoleptic quality of foods because, under the flavour analysis, a quantitative analysis of flavour-active compounds is as important as the identification of flavours. It is possible to directly identify flavour compounds and other volatile substances on the basis of gas chromatography coupled with mass spectrometry. In the past, it happened that the components, which impacted the flavour, could not be determined, because they were present only in trace amounts. As for the present day, there are special sensory systems designed for this purpose and the creation thereof was possible with

the new feats of flavour perception technology and the development of measuring technology including computer techniques utilized to assess the results obtained (Baca et al. 2011; Jeleń & Zawirska-Wojtasik 2010).

Honey has a very distinctive flavour; its general character depends on reactions induced by volatile substances contained in honey, which originate either from plants (nectar or honeydew) or from animals (produced in the organisms of bees or aphids). The formation of a general taste-flavour profile of honey is a process that also continues while honey matures, is extracted from honeycomb, and is stored and transported. Honey is capable of absorbing odoriferous compounds from the environment; therefore, its flavour can be changed by, for instance, loose capping or improper storing.

It was attempted to accomplish the assumed objective of the study by: - answering the question: what compounds occur in the volatile fraction of varietal honey; - separating flavour profiles from the experimental materials; and – finding differences in the composition of odoriferous volatile compounds contained in honey types of various botanical origin. Flavour profiles were developed using a capillary gas chromatography coupled with mass spectrometry (GC-MS). Gas chromatography is a very effective method used to separate substances analyzed. Mixtures of compounds characterized by different volatility can be successfully separated in different stationary phases, and the chromatographic column and temperature are suggested in general. With a gas chromatograph and mass spectrometer employed as a detector, it is possible to analyze also the separated compounds. The GC-MS technique is applied to identify compounds and to develop qualitative methods for determining trace amounts of substances in complex mixtures. Volatile compounds were extracted by a headspace solid phase microextraction technique (HS-SPME). The HS-SPME technique is characterized by a high responsiveness and repeatability. Moreover, it does not require any solvents and it combines the process of extraction and condensation in one phase. The HS-SPME technique involves an analysis of headspace; thus, while determining the volatile substances, it is possible to selectively isolate compounds present in the gaseous phase above the sample's surface.

Each assay consisted of eight honey samples of various varieties. Samples were prepared in 15 ml vials; each sample was a mixture of honey and de-ionized water, their by weight proportion was 5: 1 (2.5 g of honey and 0.5 ml of water).

In the samples analyzed, over 300 different compounds were determined in total and ca. 200 of the 300 determined could be identified. Some of them are those reported in the literature referring to the composition of the volatile fraction of honey. Organic acids and essential oils are those compounds that significantly impact the flavour and taste of honey. Honey also contains higher alcohols, aldehydes, ketones, esters, and polyphenols. Furthermore, there is a small amount of nitrogen compounds (e.g. enzymes produced by salivary

glands of honey bee workers; those enzymes play an important biological role) (Soria 2009; Witkiewicz & Heper 2009; Baca et al. 2011).

For individual varieties of honey, there were selected characteristic fragment ions, for which the areas of peaks in a given variety were higher than in all other samples of other varieties. In Tab. 1, there are listed those detected odoriferous compounds that were described as characteristic for each individual type of honey.

Table 1. Odoriferous compounds characteristic for individual types of honey

Honey					
Acacia	linden	buckwheat	heather	rape	honeydew
2-butenal octanal nonanal 4-methoxy phenytoetanol	α-felandren (isomer I) α-terpinene (isomer I) 3 felandren (isomer II) 2,6-dimethyl-1,3,5,7- octatetraene (isomer I) p- cymene dimethylstyrene 3,5- dimethyl-2- octanone 4,7- dimethyl benzofuran p- methyl acetophenone 2-methyl-3- phenyl -2- propenal Cymenol (isomer I) Cymenol (isomer II) Cymenol (isomer III) Cymenol (isomer IV) Geranium acid 4- metoksyfenyloethanol 2-karen-10-al p-menta-1,4(8)-diene 1,2,4,- tri (methylene)- cyclohexane Methylstyrene rose oxide (isomer I) Rose oxide (isomer II) 1,3,8-p-mentatrien NID Terpinene (isomer II)	Furfural Pentanal 3-methylbutanoic acid + acid 2-methyl butanoic pentanoic acid Butyric acid Butanal Dihydro-3- methyl -2(3H)- furanone Dihydro-5- methyl-2(3H)- furanone Dihydro-4- methyl-2(3H)- furanone 2- methyl propanal 3-methyl-2- butenal 2-methyl-2- butenal 2-methyl Butanal 3-methyl butanal p-anizaldehyde 4-methylfenol 3- amino acetophenone	3,4,5-trimethylo phenol Phenyl acetic acid Benzoic acid β-damascenone isophorone (3,5,5-trimethyl- 2-cyclohexen-1- on)	Acetoin Acetol 1-hexanol 1-(2- furanyl)- ethanone Butanoic acid NID Benzyl alcohol Benzoic acid Hydrocinnamic acid 3,5- dimethoxy benzaldehyde 3,4,5- trimeth oxybenzoate methyl 3,5-dimeth oxybenzyl alcohol	2,5- dimethyl furan 2,3- dimethyl- 2-norbornene 6-methyl-5- hepten-2-ol 1-octanol 4,7- dimethyl benzofuran 1-decanol

Source: own research

Owing to the application of the HS-SPME/GC-MS method to analyze the honey studied, it was possible to identify ca. 150 volatile compounds from the groups of chemicals such as: aliphatic and flavourtic acids, aldehydes and

ketones, alcohols and phenols, terpenoids, derivatives of furan and pyran. The HS-SPME/GC-MS method enables the checking of the quality of honey and the detecting of honey adulteration. Based on the comparison of varietal honey samples by the HS-SPME/GC-MS method, it was also possible to determine compounds that could be utilized as discriminators of honey types for the purpose of classifying honey types as regards their variety and quality.

The results obtained confirm that the above described method can be applied to construct profiles of odoriferous volatile compounds contained in honey and to find correlations between the composition of the volatile fraction of the honey product and its quality and between the instrumental and sensory analysis. The method under discussion can be also utilized as a handy tool when checking the botanical origin of honey. Consequently, the conclusion is drawn that the performed research study confirmed the fact that the developed method could be a supportive and effective addition to other contemporarily utilized methods of classification and assessment of honey quality (sensory and pollen analysis) since it brings more comprehensive quantitative and qualitative results in a relatively short time period.

Summary

Within the scope of the research study, a hypothesis was verified, which assumed that chromatographic analyses constituted a basis for varietal discrimination of honey. In order to verify this hypothesis, volatile compounds present in honey were analyzed. The assays performed using a gas chromatograph made it possible to construct profiles of compounds coincident with the botanical origin of honey. Therefore, based on this criterion, it was possible to identify them. The most characteristic profiles of volatile compounds could be made for the acacia, linden, buckwheat, heather, rapeseed, and honeydew honey.

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THE WORLD MARKET OF FUNCTIONAL FOODS

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Introduction

Since the early 21st century, our society has been witnessing a continuous increase in life expectancy and, at the same time, greater attention to quality. Consumers are increasingly concerned about their health and pay more attention to their lifestyle and the healthiness of their diet (Szakály, Szente, Kövér, Polereczki, & Szigeti 2012).

The link between diet and health has been recognized since ancient times: as Hippocrates said, “Let food be thy medicine and medicine be thy food”. This “connection” is increasingly the focus of attention of the modern consumer, for whom, nutrition must not only meet basic nutritional needs but also provide an additional physiological benefit (Hasler 1998). In this context, functional foods (FFs) play a key role. The increase in demand for such foods can be explained by the increasing cost of healthcare, the steady increase in life expectancy, and the desire to improve their quality (Siró, Kápolna, Kápolna & Lugasi 2008).

The concept of FF was born in Japan in the early 80s, used by the industry to describe foods fortified with specific ingredients to impart some health benefits. In 1993, concerned about the aging population and the high cost of health care, the Japanese Ministry of Health and Welfare instituted a policy of “Foods for Specified Health Uses” (FOSHU) by which health claims of some selected functional foods were legally permitted. Up to now (November 22, 1999), 167 FOSHU products have been born (Arai 2000). Later, they have also been introduced in the USA, Europe and the rest of the world (Verhagen, Vos, Francl, Heinonen & Loveren 2010).

Functional foods are the first to bear health claims. According to the Codex Alimentarius, a claim means “any representation which states, suggests or implies that a food has particular characteristics relating to its origin, nutritional properties, nature, production, processing, composition or any other quality”. The two main types of claims regard: (a) what the food

contains, i.e. nutrient content claims and comparative claims; and (b) what the product does in terms of health, well-being and performance, i.e. health claims (Richardson, Affertsholt, Asp, Bruce, Grossklaus, Howlett, Pannemans, Ross, Verhagen & Viechtbauer 2003). These health claims have become a means to communicate to consumers the health benefits of foods that contain specific formulations, conveying relevant information that would otherwise remain unknown.

Communication has a great impact on consumers' knowledge and attitudes (Verbeke 2008). Health claims are key factors for the development of the functional food market. They play a central role in driving purchase decisions, and help consumers make more informed food choices, especially if the product is made of new ingredients or performs actions beneficial to health that are poorly understood (Annunziata & Vecchio 2012). Consumers' purchasing decisions are influenced by many factors. Some studies have shown, for example, life satisfaction and age influence the choice of functional foods (Urala & Lahteenmaki 2007; Carrillo, Prado-Gascó, Fiszman & Varela 2013). These new trends have dramatically changed the industry. Food companies are investing in this sector, with new marketing and communication strategies, and changing their food innovation process. The market for functional foods is dynamic and growing. Japan, the homeland of the FFs, is a leader in the field, followed by the United States and Europe (Bleiel 2010).

The aim of this paper is to identify which areas are more prone to the development of this market, paying more attention to those countries that contribute significantly to the growth of this sector.

Nutrition and health claims

The history of functional foods is not very old. In 1980, a Japanese company, was always more interested in the prevention of diseases related to a healthy lifestyle, out of its awareness of the rapid aging process. This gave a strong impetus to food science and to the politicians in the food sector (Arai 2001). Later, in 1991, these foods were defined by the acronym FOSHU, Foods for Specified Health Use, namely foods having beneficial effects on human health due to the presence of particular elements or the lack of allergenic constituents. The first FOSHU product was approved in 1993. Over 500 have been approved since then (Jones & Jew 2007).

Most food industries have paid attention to the concept of functional food introduced by the research projects of the MESCC (Ministry of Education, Science and Culture), started in 1984. In the US, special attention was given to claims that accompany the products with a functional role. Three important changes occurred in 1990, 1994 and 1997. They affected the dissemination of

information to consumers about the relationship between diet and health in food regulations. The first of these is the Nutrition Labeling and Education Act of 1990 (NLEA). The NLEA allows statements on food labels that characterize the relationship of any food or food component to a disease or health-related condition. Such “health claims” must be pre-approved by the FDA before their use.

The second and probably most important (and controversial) change in food regulations was the passage of the Dietary Supplement Health and Education Act of 1994 (DSHEA). This act regulates dietary supplements as foods, not food additives, defining them as “vitamins, minerals, herbs or other botanicals, amino acids, or other dietary substances for use by man to supplement the diet by increasing the total dietary intake, including concentrates, metabolites, constituents, extracts, or any combination of the above”.

To expedite the health claims approval process and thus hasten the availability of health messages to consumers, Congress enacted the FDA Modernization Act (FDAMA) in 1997. This legislation streamlines the FDA pre-approval process by enabling the use of so-called “authoritative statements” on food labels as health claims. Historically, companies that have attempted to launch a functional food in Europe have faced a variety of legislation regulating the approval of products, the kinds of nutrition information required on labels, and the types of functional and health claims that were allowed in connection with a product (Bech-Larsen & Scholderer 2007). After a first attempt at harmonization, which has prohibited all product-related communications from attributing properties for prevention, treatment or cure of human diseases to food (European Parliament & Council of Europe 2001), the situation changed. On July 2003, the European Commission proposed a harmonized regulation COM/2003/0424 on nutrition and health claims made on foods, including dietary supplements (FAO 2007). In December 2006, the Council and Parliament adopted the Regulation 1924/2006 on nutrition and health claims made on foods. For the first time, this Regulation lays down harmonized rules across the European Union for the use of nutrition claims such as “low fat”, “high fibre” or health claims such as “reducing blood cholesterol” (European Parliament & Council of Europe 2006). This regulation ensures clear and accurate information based on evidence accepted by the whole scientific community. In order to bear claims, foods will need to have appropriate nutrient profiles, which will be set. This Regulation introduces a new category of claims, i.e., “Reduction of disease risk claims”. This means any health claim that states, suggests or implies that the consumption of a food category, a food or one of its constituents significantly reduces a risk factor in the development of a human disease. Europe instead displays requirements for a high level of scientific substantiation. The European Food Safety Authority (EFSA) is involved in

implementing the new regulation. This Authority has published a guidance to help companies who want to submit health claims for authorization (EFSA 2007). The assessment of a health claim by EFSA is the first step in the authorization process. Only those claims, which are scientifically substantiated, will finally be authorized for use. The final approval of a health claim is the responsibility of the European Commission and Member States, based on the scientific assessment expressed in the opinion of EFSA's Panel.

World market of functional foods

The market of functional foods is growing rapidly and is highly dynamic. In many ways it may even be characterized as an experimental environment (Bech-Larsen & Scholderer 2007).

The increase in life expectancy, resulting in an increase in the number of the elderly and the desire for an improved quality of life, as well as the increasing costs of health care, has stimulated governments, researchers, health professionals and the food industry to study how such changes can be managed more effectively.

It is difficult to have a precise estimate of the data on the total turnover and the volume of functional foods sold.

There are three regions where sales of functional foods are concentrated: Japan, United States and Europe (Datamonitor 2004; Menrad 2003). Williams et al., (Williams, Pehu & Ragasa 2006), which indicates that the demand for functional foods within developing countries is growing, presenting a lucrative opportunity to develop domestic markets. The demand for functional foods appears to be heterogeneous throughout different countries. The industry growth is affected by the selling price and, consequently, the average per capita expenditure globally. In 2013 Euromonitor estimated that the average per capita expenditure is around US\$ 36 from country to country, reaching a maximum value of approximately US\$ 272 with a worldwide turnover in the sector of approximate US\$ 252 billion (Euromonitor 2013). Figure 1 shows the global distribution of functional foods market shares.

As can be inferred from figure 1, the main market for functional foods is Asia Pacific. Revenues for the field of functional foods in Asia and the Pacific islands constitute as much as 34% of total revenue worldwide. This is not surprising, given that Japan alone is one of the main markets for functional foods. The aggregate of Asia Pacific covers a vast territory, which includes the various countries where the market for functional foods is growing rapidly, as, for example, China, South Korea and Malaysia. For this reason, the revenues for the field of functional foods in the Asian territory are so crucial in the world. The second largest market in the world is the North American one, substantially consisting of the United States and Canada. These two

countries alone reach 25% of total revenue worldwide. Particularly in the United States, the field of functional foods is a real business. Their marketing is favored by a legislative approach as well as a very permissive advertising. In addition to that, the growth of the sector is boosted by the size of the territory and the population. These factors contribute to the high revenue.

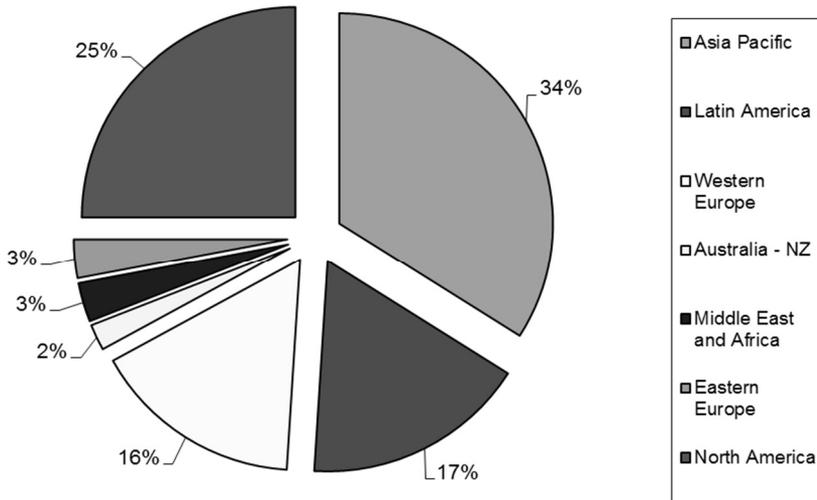


Figure 1. Percentage breakdown of total revenues worldwide.

Source: Euromonitor 2013

In Latin America, the functional foods and food production are relatively “new”, but very promising. In fact, as can be seen from figure 1, the revenues for this sector are really high. Whereas the marketing of functional foods in South America started very recently and revenues for the field of functional foods currently make up 17% of total revenue worldwide, it can be said that Latin America is a market on which investment in functional foods should continue. From the point of view of the percentage breakdown of functional foods revenues worldwide, the markets of Australia, New Zealand (NZ), Africa and the Middle East and Eastern Europe can be considered negligible, since all together they make up just 8% of total revenue worldwide. Regarding Australia, the low revenue for functional foods is certainly due to the small size of the population and, therefore, the small number of potential consumers. As far as Africa and the Middle East are concerned, the market for functional foods is hampered by extreme poverty and socio-cultural backwardness of

most of the territory, as well as by the uprisings and conflicts which unfortunately involve many countries of this vast geographic area. Let's not forget that for most of the African population there is no need to eat healthy, rather to just eat. Therefore, apart from a few rare exceptions, the market for functional foods in Africa and the Middle East is not favourable.

The demand for functional foods in Europe varies considerably from country to country, being affected by culture and culinary tradition (Castellini, Canavari, & Pirazzoli 2002). The market of Western Europe, has been relatively stable over time, with a 16 % of total revenue worldwide. The main source of revenue for the area of functional foods is the United Kingdom with 20 %, followed by Germany with 14%, which corresponds to more than four million Euros. Follows France, with 13% of total revenues, amounting to about four million Euros. Spain and Italy account for 12 and 11% of total revenues respectively, which correspond to more than three million Euros.

In Eastern Europe the functional food market accounts for approximately 3% of total revenues worldwide, and it includes some interesting countries from the viewpoint the development of this sector. Russia alone, for example, constitutes 51% of total revenues, followed by Poland, which has 17% of total revenues and the Czech Republic with approximately 9% (Liberatore & Vicentini 2014). On the other hand, for most of the other countries that make up the vast east European territory, the marketing of functional foods has just timidly begun or is even non-existent. So, the market for functional foods in the East European region is very fragmented and in need of a better organization and an adequate promotion in order to foster development.

Table 1. Annual percentage growth rate in the functional foods sector worldwide.

	2007-08	2008-09	2009-10	2010-11	2011-12
Asia Pacific	6.2	6.3	8.9	8.1	10
North America	4.8	- 4.2	1.4	5.5	1.4
Latin America	11.1	8.5	12.8	12.9	12.4
Middle East and Africa	14.9	6.6	9.1	9.8	9.3
Australia-NZ	6.7	6.6	7.9	8.1	5.2
Western Europe	5.4	2.4	1.5	2.3	2.5
Eastern Europe	20.6	1.6	7.7	11	9.2

Source: Euromonitor 2013.

For what concerns the global trend of functional foods the situation varies from country to country. In order to have a more complete view of the world market, table 1 shows the performance of the sector in terms of annual growth rate.

This boom not only underlines the dominance and maturity of the Asian market, which in 2012 recorded as many as 10 percentage points. As shown in table 1, the market has been consistently strong, only in 2010-11 did it register a small decline but this has not destabilized its growth.

Asia is a vast territory, the most important countries in the market for functional foods are China and Japan, let's not forget the Pacific Islands, which contribute to the growth of the market, despite the small percentages.

The economic and social context influences this development, it was found that the increase in income guides the growth of the country. Several studies claim that the market for functional foods in China is expected to expand further in the near future, becoming increasingly important globally (Cranfield 2011).

Despite being the second largest market, North America sees a decidedly unbalanced trend. From 2007-08 to 2008-09 the retail value of functional foods decreased by less than 4 percentage points. The reason may depend on the economic recession that hit the United States, which has crippled the global economy. Despite this, America has recorded positive values, reaching about 6% in 2011, but then suffered a further decline.

Latin America is considered a promising region. In 2008-09, there was a slight decrease of 4 percentage points probably due to the economic crisis. From 2009 to 2011-12, the trend was positive and this does nothing but emphasize how this turns out to be potentially attractive in terms of growth and investment.

Finally, the Middle East and Africa. The economic and social context make the growth of these markets difficult and unstable. However, the situation is not entirely negative. There are also areas with high growth potential such as Saudi Arabia, South Africa and the State of Israel. These aggregate with Australia and New Zealand, with minimum percentage impact on the industry.

The growth trend in Eastern and Western Europe is varied over the years. Definitely better for Western Europe with a market that has established itself over time even if the growth value is lower compared to Eastern Europe (emerging market). Indeed, the latter recorded a sales boom in 2007-2008 before suffering a collapse in the ensuing year, which coincided with the global economic crisis, and a sharp recovery as early as 2010.

Although the annual growth rate in Eastern Europe is higher than that of Western Europe, it must be emphasized instead as the average annual consumptions per capita are higher in this area (Euromonitor 2013).

Conclusions

The agribusiness sector has undergone a profound change in the last two decades. The deepening global crisis that has hit the economy has considerably changed consumers' habits and as a result, companies have been forced to revise their action plan. Increased attention to consumers' health, has given the scientific research and business, a stimulus to develop products with therapeutic features that go beyond the role played by traditional foods. Research in this field has given a strong contribution and made sure that the food market can evolve.

These products have had an immediate success in most of the industrialized countries and in the developing ones, therefore we can say that they can actually be an opportunity both for the industry in economic terms, and for the consumer in terms of health.

Looking at the situation of the world market, the leading exponent in the field of FFs is Asia Pacific, followed by North America, Latin America, and finally Western Europe. The field of "functional" food has a potential that still has to expand, but it calls for the solution of the main issues related to regulations and information aspects. Therefore, investments in the field of functional foods will prove beneficial only if specific legislation, an international collaborative approach and a direct and effective strategy for communication between producer and consumer will be developed.

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HELICICULTURE AND SNAIL CAVIAR: NEW TRENDS IN THE FOOD SECTOR

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Introduction

The snail farming is an emerging economic activity with intense diffusion in the last decade, but the available information on domestic and foreign markets, economic actors and their involvement are still unclear and often contradictory. On economic crisis contexts and prospects characterized by a globalized and highly competitive market, we are witnessing the growth of interest for productive activities that create added value. This is the case of companies related to land snails. Many initiatives, including industrial ones, see the snails production and their food products as a way of feeding the economic outlooks, employment and gastronomic interest growth. On the other hand, the gastronomy quest of new subtle and delicate flavors has been leading to a new food product, an exotic quality of “white caviar”: the *Helix aspersa* snail eggs. Land snails are recognized as healthy food due to their low fat content and presence of nutrients required for a well-balanced diet, in addition they are a great vehicle for flavour. Snails have been farmed for food for hundred years and are a rich source of proteins. Indeed snail preserves were established in Rome in 50 B.C. so the food consumption and snail farming date back to ancient Romans. Gaius Pliny the Elder" (23 - 79 A.D.) in his "Naturalis historia" treatise refers to the edible species. Among the known snail species, 116 are considered edible (Abdulmawjood, A. and Bülte, M. 2001a). The most popular edible snails are *Helix aspersa*, *Helix pomatia*, *Cornu aspersum* and *Eobania vermiculata*. Another less valuable edible snail, consumed in Europe, is *Helix lucorum* from northern Italy and Turkey. *Cooked snail meat is highly appreciated in France, Italy, Spain, Germany, Austria. It is also gaining popularity in Britain and in the United States, particularly in areas populated by French, Italian, and German people, but it is already eaten in numerous countries such as Philippines, Morocco, Nigeria, Algeria, Portugal, Greece, Belgium, Vietnam, Laos, Cyprus, Cambodia, Ghana, Malta, Terai of Nepal, Japan, several regions of India and*

Southwestern China (Perma Culture Science 2014). Moreover, we have to consider the new producing countries such as Poland, Bulgaria, Turkey, North Africa and Chile; this last is one of the best producers of snail caviar. One of the most used species for caviar production is the North African Great Grey Snail, a domesticated form of *Cornu aspersum*. The consumption rise has increased the interest for breeding, which has slowly gained a space in the market, through the study of the most updated techniques and which has given the opportunity to obtain more expensive and quality products, with undoubtedly superior characteristics compared to the product collected in the wild.

Breeding, production and marketing of snails are usually subject to the general rules related to animal health, agriculture and food consumption, but most of the countries do not have specific legislation, as it happens in producer countries as Argentina, Belgium and France, where a quality label has been also established. The lack of legislation facilitates the commercialization of land snails for consumption without a prior check by administrations, as well as the possibility for some companies to buy product from not registered stock farms or small rural collectors.

If snails provide an easily harvested source of protein to many people around the world, it is true they are also considered as one of the worst causes of food allergy. In fact, some people may develop severe episodes of asthma after ingestion of snails and a connection with house-dust-mite allergy is firmly established (de la Cuesta C.G. and others 1989); more recent studies have identified an allergen in snails which is responsible for cross-reactions with foods and inhalants (Martins L.M.L. and others 2005). Snail allergy is rare but it can cause fatal and almost fatal anaphylactic reactions, so a notice explaining the hazards of eating snails should be included in the label together with other relevant information for consumers.

Characteristics of edible snails products

Snails have appeared on Earth, in an aqueous environment, million years ago. Their name comes from the Byzantine period and it is the diminutive of the word “saliagas” (slug = Saligari in Greek), from the burr (Salio in Greek) released during their movement. Snails are among the most known animal species in the world and belong to the second largest category among the groups of animals that inhabit the Earth (after insects), called Gastropods Pneumonofori Moluschi. They have a special adaptive capacity to different environmental conditions with a significant logarithmic reproduction rate. In ancient times, the snail's role was instrumental for the development of many sciences. The father of Medicine Hippocrates was the first who studied and appreciated the special properties of foamy slime that the snail secretes for its self-protection and the protection of its eggs. Pitagora from Samos studied

shell's structure and formulated his famous "golden rule". Archimedes conceived the principle of "lifting of the screw" in Mechanics. Snail's role has been important as food and gastronomy since ancient times. Many times it was mentioned that snails were part of the daily diet either of the poor or the rich Greeks. Since the end of the 19th century, their consumption has greatly increased due to the advertising based on their nutritional value and culinary uses.

There are many species of snails on the market, but the one which dominates the world market and the "gourmet cuisine" is the "*Helix aspersa Müller*", which owes its name to the Danish naturalist Otto Friedrich Müller (1974). The *Helix aspersa Müller* (*Cornum aspersum*) name is found in many scientific references and especially at zootechnic and commercial level. Other common names of the species are: "escargot petite gris" in French, "brown garden snail" in English, "cretan koxlios" in Greek (SALIPAP C.O. 2014)

Snails, improperly called land snails, are mollusks belonging to the *Order Stylommatophora*. They are equipped with shell and this feature distinguishes them from so-called snails, which belong to Families: Arionidae and Limacidae. They have got edible and excellent meat, extremely lean and rich in proteins of high biological value, which has been used for food in some European and extra-European countries. The available references about the nutritional values refer to the species *Helix pomatia* and are rather discordant. The meat however contains low levels of cholesterol or fat (0.5 to 0.8%) and contains vitamins A and C, plus 1.5% minerals (zinc, copper, manganese, potassium, calcium and iodine). As for protein (a total of 12 to 16%), the meat contains 9 of the 10 essential amino acids important for humans (Abdulmawjood, A. and Bülte, M. 2001b). With regard to identified fatty acids, results show that snails, although being terrestrial animals with lung breathing, presents a fatty acid composition similar to that of fishery products. Indeed, the breakdown of fatty acids per 100 grams of edible portion is equal to 0.26 g of monounsaturated fatty acids, 0.25 g of polyunsaturated fatty acids and 0.36 g for saturated fatty acids. The edible land snails have got a very low energy value (Table 1) and, if boiled, they could be easily included into a low-calorie diet against overweight. In any case they aren't easily digestible food; the maximum recommended serving is about 10-12 mollusks per person, while the average serving size includes between 5 and 6 pieces for adults.

The real value of the snail meat is given by amino acids, and in particular by the presence of the essential ones. It is also rich in minerals, especially calcium and phosphorus, due to the genesis and the development process of the shell. Among other mineral elements is definitely worthy of note the presence of iron, potassium and magnesium.

Table 1. Chemical composition and energy value of *Helix pomatia* snails meat per 100g of edible portion

COMPONENT	Value	Unit
PROXIMALS		
Alcohol (ethanol)	0	g
Energy, Total	86	kcal
Fat, total (total lipids)	1.4	g
Proteins, Total	16.1	g
Water (moisture)	79.2	g
CARBOHYDRATES		
Carbohydrates	2	g
FATS		
Fatty acids, total monounsaturated	0.26	g
Fatty acids, total polyunsaturated	0.25	g
Fatty acids, total saturated	0.36	g
Cholesterol	50	mg
VITAMINS		
Vitamin A, retinol equivalents retinoschisis activities and carotenoids	30	µg
Vitamin E, alpha tocopherol equivalents vitamers activities E	5	mg
niacin equivalents, Total	1.4	mg
riboflavin	0.12	mg
thiamine	0.01	mg
Vitamin B-12	0.5	µg
Vitamin B-6, Total	0.13	mg
MINERALS		
Calcium	10	mg
Iron, total	3.5	mg
Potassium	382	mg
Magnesium	250	mg
Sodium	70	mg
Phosphorus	272	mg
Selenium, total	27.4	µg
Zinc	1	mg
PERCENTAGE DISTRIBUTION OF ENERGY		
COMPOSITION	percentage	
Proteins	76%	
Lipids	14%	
Carbohydrates	9%	
Alcohol	0%	

Source: Elaborated from “Consortio BEDCA y Agencia Española de Seguridad Alimentaria y Nutrición, 2010”

Besides the meat, snail eggs are also consumed, a highly prized "French caviar", while their shell, especially of the *Helix lucorum* species, finds

numerous applications for dining preparations. The caviar consists of snail eggs looking like small white beads of 1-2 mm. It is the result of a very difficult job of selection by snails producers, through induced coupling thanks to recreation of the necessary habitat and the microclimate, the broods and individual eggs selection. Each snail lays hundred eggs per year (about 4 g) between March and April, therefore, to have an adequate production it is necessary to seasonally adjust the spawning, so snails don't hibernate and lay four times a year instead of two as they would do in nature. Once harvested, the eggs are cleaned and tidied, by means of very hard manual work. Then the product is usually pasteurized, with added sea salt, citric acid, potassium sorbate, sodium benzoate and flavored with rosemary and transformed in to product that will last for at least three months, to be sold as "Pearls of Aphrodite." The first manufacturers were the French in 2004, but have been joined by other countries such as Chile, Italy and Poland. The caviar production is well underway and there are producers who are working to understand where, how and when to harvest the eggs. On the other hand there is the food and wine sector that begins to exploit the potential of an "elite" product like caviar. For this product no scientific studies have been published on the nutritional value and antioxidant properties and the effects on intestine, while there are some evidences of allergic reactions (Food Science Lab 2014). Only one reference in literature (Panus 2014) reports nutrition information on *Helix aspersa Müller* caviar (Table 2).

Table 2. Caviar's nutritional value of *Helix aspersa Müller* snail

NUTRITION INFORMATION		
Serving Size: 1 tea spoon (10g)		
Portions per package: 5		
	100g	1 portion
Energy (kcal)	46	4.6
Protein (g)	4.5	0.45
Total Fat (g)	0.3	0.03
Carbohydrate (g)	6.4	0.64
Sodium (mg)	1400	140

Source: Elaborated from "Panus", 2014

There is another trend that has centralized further interest on heliciculture. For cosmetics, there has always been a particular interest in the use of snails, due to the healing that derives from the *Helix aspersa* snail. The mucus they leave behind is claimed to contain hyularonic acid, powerful proteins and antioxidants, all of which can help to repair and revitalize the skin. Snails have been reported in the literature since ancient times as a

therapeutic remedy. Since the ancient Greek age to the Middle Ages, snails have traditionally been used as a medicine for stomach problems such as peptic ulcers and gastritis, to heal wounds and to stop bleeding and, in the form of syrup, to calm cough, loosen up the phlegm and let its easier elimination. Back in 19th and early 20th century snails were used in England as a tuberculosis cure, once dissolved in salt and mixed with cream and sugar (Canada Free Press 2013).

Snail production and market

Drawing out a general framework on snails farming is complex. Production mainly consists of natural harvesting and artificial breeding. The available data on production are still incomplete and not exhaustive and refer very often to private institutions and industry associations. The global character of the phenomenon is evident considering the interest of stakeholders who still face a booming market. The market, basically, is constituted of harvesting countries or producers and consumer countries. Then, it is important to notice that there are producer countries which cannot cover domestic demand as for France and Italy.

The snails market for gastronomy of *Helix* species has suffered in the last 20 years deep and wide-ranging changes in quantities, commercial lines and prices. In the '70s in large part of Western Europe specific laws were enacted that regulated the natural harvest of *Helix*, mollusks, but later the harvesting practice has been prohibited in Europe. In the '70s heliciculture was developed, in order to compensate, through a managed and controlled production, the decline and lack of product, previously retrieved in nature. Recently in the world, especially in the Western countries, a significant increase in consumption due to the entry of snails in the world of industrial processing and canning has been recorded. In the early 2000s the total number of snails placed on the market, including alive, frozen and preserved, stood at 320,000 tons per year worldwide (Figure 1).

Product harvesting has shifted from Western Europe to Eastern Europe, where intense and organized harvesting was encouraged in all countries of the area from Poland to Bulgaria, in order to increase the European currency intake to help their fragile economy. The total consumption has come to touch, at the beginning of the new century, the share of 420,000 tons, purchased for almost 50% by processing industries in France which, with their factories in Turkey and especially in North Africa, exports worldwide canned snails with the famous name "escargot".

Post-Chernobyl radioactivity has encouraged the French to purchase intensively grown snails, in order to avoid the consumption of wild snails collected in Eastern Europe. The global business of snails in the past ten years,

referring to the wholesale prices touched and exceeded 10 billion euro (Istituto Internazionale di Elicicoltura, 2014a).,

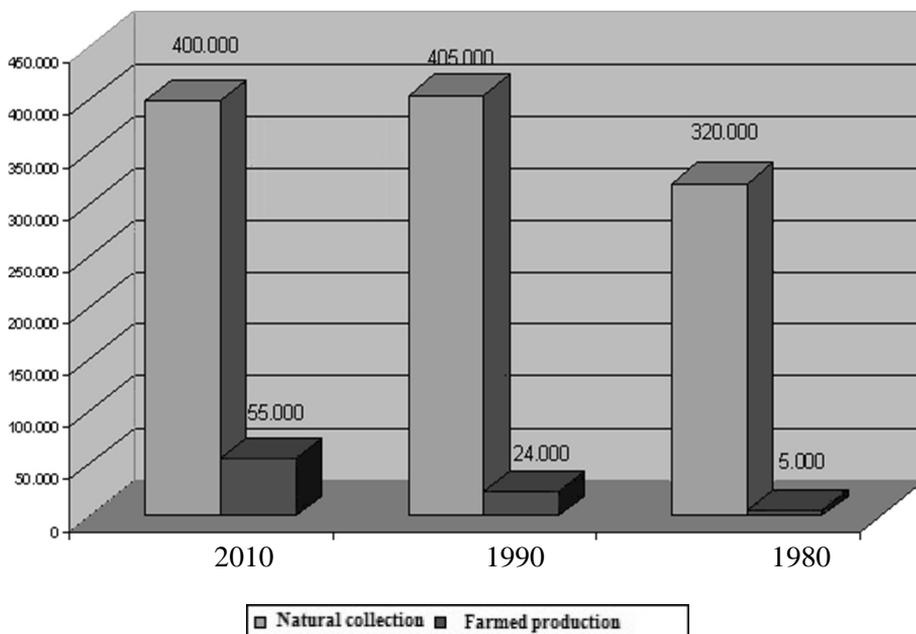


Figure 1. Global Snails Consumption: natural collection and farmed production

Source: International Institute of Heliciculture, 2014

For example, Italy in 2010, despite the economic crisis, has exceeded 38,000 metric tons of consumption, an increase of about 2% over the previous year. Imports of live and preserved snails by producer countries have touched again in 2010, 225,000 tons, about 65% of the total product. The total value of the entire supply chain (farmed production, imports, industrial processing and gastronomic consumption) was more than 210 million euro.

The international prices of live and semi-finished product exported from the producer countries are due to low labour costs of people who ran the business of wild harvesting. Prices have changed and increased in the early '90s when the events that followed the fall of the Berlin Wall have changed the global policy framework and the approach of these countries towards the European Economic Community. Along with the consumption growth, the interest in the industrial production has increased, bringing to more expensive and superior quality products, in respect to the wild harvested ones. Anyway the snails breeding has not achieved its original purpose yet, that is to fully replace the wild harvest. The industrial production covers in fact only a small

part of the whole consumption. In 2010 it was estimated that globally the snails breeding covered only 15% of the total sales (Istituto Internazionale di Elicicoltura, 2014b).

Poland is one of the harvesting and producer snail countries which exports to France, Spain and Italy most of its slimy mollusks: *Helix aspersa*, *Helix pomatia* or "winniczek" in Polish, also known as Burgundy snail in the French region. The French import 95% of the national snail consumption, 9% of which comes from Poland, according to the trade figures diffused by the Polish embassy in Paris. Poland also offers canned meat, pearly white snail eggs caviar and slime facial cream. According to the Polish Central Statistical Office, in 2011, Poland exported 282 tons of snails, corresponding to an economic value close to 1.1 million euro (AFP, 2013).

Heliculture legislation in European Union countries

In Europe, even if there isn't a specific regulation for snails production, there are countries such as Belgium and France that are endowed with a quality label. In Italy there is "The International Institute of Helicoltura" which buys *Helix aspersa* Müller and *Maxima* snails from national companies which have signed a partnership business contract with it, which allows the use of all the technical information provided by the Institute and sell it snails, but not in exclusive way. The Institute also gives producers the origin brand "Italian Snails". A quality brand is also given in Belgium where, following the issue of the decree of 1998 which regulates the production requirements and quality, the brand "Escargot Fermier" or "Snail Farm" is released to snails produced in farms that practice biological heliculture (Sicurezza–alimentare, 2013a). In addition to it, a sale label is applied on products, reporting type of preparation and/or performed treatment, as well as the physical conditions of the snail, the total net mass of consumption, the number of parts or units contained in each package, the scientific name of the snail, the list of ingredients and the expiration date.

In Spain, the lack of legislation and of a quality label facilitates the commercialization of land snails without a prior check by administrations, so some companies can buy the product from not registered individuals. Snail breeders are subject to the same legal conditions as for any farmer or producer. The only existing European standard is related to "the conservation of natural areas and wildlife" which strictly prohibits the indiscriminate harvest. In Italy, in Spain as in other countries, the lack of precise legislation forces companies to equate the production of snails to that of ruminants or other animals; however, when the demand is covered by other countries, the European Union legislation imposes specific requirements regarding compliance with the minimum conditions for the health of the animals (Sicurezza-alimentare,

2013b). The purpose of the standard is to prevent that these products constitute a risk for human consumption. It is difficult to import products without the required health certificate. In this why, the market is better regularized outside than inside. The sector therefore requires specific rules governing the breeding of snails, in order to avoid the consumption of wild land snails. Wild snails, in fact, are exposed to massive use of toxic products used in agriculture and can cause serious damage to the health of consumers.

The current policy of the European Union aims at protecting human health and consumers' interests. This policy has been reformed since 2000 to ensure a high level of food safety at all stages of processing and distribution. The Hygiene Package of European Union has introduced the extension of self-control HACCP (Hazard Analysis Critical Control Point) for the primary production and the application of the ISO 22000:2005 "Traceability in food chains" standard (De Leo, F. and Pastore, S., 2012). Through the Hygiene Package all Member States have the same criteria regarding the hygiene of food production and therefore health checks are carried out according to the same standards throughout the European Community. The adoption of this practice is for all food producers, but also for all people involved in the subsequent and intermediate steps in the food chain. Among the regulations included in the hygiene package there is the EC Regulation 852/2004 laying down the principles and requirements of food law, establishing the European Food Safety Authority and the procedures in matter of security. The same regulation affects all activities of the food supply chain, either animal or vegetable, including primary production. This refers to the breeding and cultivation of raw materials, including harvesting, milking and processing prior to slaughter, and also it includes hunting, fishing and gathering of wild products (mushrooms, berries, snails, etc.).

Snail species are usually identified and differentiated by morphological characteristics; however, specific snail species are unrecognizable after cooking and canning. This could represent an issue because snails are imported as canned products too and it can happen that cans are incorrectly labeled just to increase profits. So there is the need for a suitable technique to identify canned snail meat, as for fish, crustaceans, and caviar (Carrera and others 1996, 1998, 1999). Moreover snails carry protozoa and many forms of salmonellae, therefore proper health practices should be used when feeding them. So the preparation of snails or snail eggs for preservation or consumption should be subject to inspections for public health and specific information for consumers should be provided in relation to mild symptoms such as oral allergy syndrome, urticaria (hives) and severe symptoms such as asthma and anaphylactic shock that can occur after snails consumption (InformAll. Allergenic Food Database 2014).

Conclusion

Snails' meat was highly appreciated as food since Roman times and the art of fattening snails is said to have been discovered by a Roman named Fulvius Lupinus (Larousse Gastronomique, 1961). Snails were also eaten at Minoan Akrotiri, perhaps imported there from Crete as a luxury item. According to Galen, they also represented a common food for Greeks of the Roman period. Snails were fattened on emmer meal mixed with grape syrup (Food in the Ancient World from A to Z, 2003). In France snails have always been very popular. Snails have been eaten cooked in large part of European Countries, but they are also gaining popularity in Britain and in the United States. From a nutritional point of view they are excellent food. Their meat is very lean and rich in proteins of high biological value, with an excellent flavor.

On economic crisis contexts with a globalized and highly competitive market, the breeding of snails is an emerging productive sector. The farming process of edible terrestrial snails (heliculture) was developed in the '70s when specific laws were enacted to prohibit the harvest of *Helix* mollusk in large part of Western Europe. The heliculture sector has recorded in the last 20 years a consumption rise and major interest from breeders in European and extra-European countries.

The market launch of snail caviar and the use of snail mucus in cosmetics are increased the interest in the snails farming. The implementation of this activity in some of the world's poorest communities gives a valuable source of income and may even help to meet the ever-increasing demand for food. Moreover, the environmental footprint of snails as food is much smaller than other types of meat.

"Escargot" is the French name which most commonly refers to either *Helix aspersa* or to *Helix pomatia* species, but other varieties of snails are eaten. Physical features are expressed by variations in size, colour and banding of the shell. Some species have got higher potential for adaptation and, if introduced in non-native habitats without control, they could possibly dominate the local snails. Because of its dominating characteristics, *Helix* group snails are banned in Canada; only *Otella lactia*, *Otella emulata* and *Cedpaea nemoralis* species are allowed (Ministry of Agriculture, Food and Fisheries Canada, 1994). Snails farming, if done in the proper way, could also help to protect biodiversity.

The lack of common regulation for production and trade, as well as an European quality certification that identifies the product, together with the limited data available, make the analysis of an emerging phenomenon, characterized by strong interests, very difficult. The application of the Hygiene Package in the European Union to snails farming doesn't represent a complete protection for consumers. So the creation of a Community quality

certification is necessary which takes into account also the existing national brands as well as a label with health information, containing warnings in relation to the hazards of eating snails, like mild symptoms such as oral allergy syndrome, urticaria (hives) and severe symptoms such as asthma and near-fatal anaphylactic reactions.

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INSECTS FOR FOOD: A CRITICAL REVIEW

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Introduction

Well-being of people and economic growth have always attracted the attention of economists and governments. The Physiocrats first and the Classical economists later, put in evidence that the natural resources, and in particular the availability of arable land, were not unlimited and that this prospect, in the long term, would have caused recession, economic crisis, social conflicts and poverty.

A couple of centuries after the publication of “*An Essay of the Principle of the Population as it Affects the Future Improvement of Society*”, the catastrophic theories of Malthus have been, thankfully, largely disregarded for many reasons. However, a fifth of the world population is still suffering from hunger and the state of the environment has become increasingly worse. The modern societies, especially the most technologically advanced, have developed and continue to grow along ethically, environmentally and economically unsustainable pathways.

The law of diminishing marginal returns of David Ricardo is, in fact, still valid today, especially with regard to the availability of food resources, for the achievement of which it is necessary to massively increase the use of inputs with high environmental impacts, such as, for example, fertilizers and pesticides. In addition, it is expected that, by 2030, the world population will be near 9 billion people (United Nations 2013a). To the need of food for these people, the need of land, water and resources to feed animals also must be added.

Nowadays, there is the widespread concern that, in front of a steady growth of the world population and a constantly increase of the consumption of meat, our planet could not sustain new intensive livestock farming (Alexandratos & Bruinsma 2012). There will be the need to have a “second

planet” in order to properly meet, in the future, the current standard of consumption: the Earth’s resources could not be enough for all the people.

In this context, the use of insects for human consumption “could” represent a reliable source of important nutrients and a good eco-friendly choice, having a remarkable nutritional and environmental potential. We have used the verb “could” because the aim of this paper is to evaluate the advantages and disadvantages associated with entomophagy and eventually review the current certainties that support the opinion that, already today, this alternative food choice is already mature for the market.

Materials and methods

Currently, according to FAO, about 1,900 species of insects – beetles (31% of the total), caterpillars (18%), ants, bees and wasps (14%), grasshoppers, crickets and locusts (13%) etc. – are consumed in the world and there are about two billion people, concentrated in South-America, Africa and Southeast Asia, who regularly use insects (FAO 2013; Van Huis 2013). Even if Westerners already consume and appreciate, often without awareness, products coming from the world of insects – for example, the “red cochineal” or carmine, known as E120, obtained by aphids – we are still very reluctant to accept this potential new source of food.

In fact, it seems that psychological and cultural factors represent the main obstacle and attitudinal barriers to the consumers’ acceptability of this source of food and for the diffusion of entomophagy among people in the richest countries. However, because these are topics that are beyond our competences, these aspects, as well as those related to socio-cultural, anthropological, historical and geographical aspects will not be analysed in this paper.

We have investigated, instead, the nutritional, environmental and economic issues associated with the consumption of insects, comparing them to livestock. We have also omitted the discussion about the massive use of insects in traditional and, in part, modern medicine and pharmacopoeia of many countries, such as China, Korea, Brazil and India (Alves & Alves 2011) and also the analysis of the farming practices of insects for animal feed.

A large number of references have been examined and in particular the most significant and most recent papers, highlighting the positive and negative aspects related to the use of insects for human consumption. These critical evaluations have enabled us to express an opinion related to the sustainability of medium to long-term consumption of these “new” alternative food resources.

Results and discussion

The institutions and reports which support the entomophagy as a secure source of food and nutrients, to the entire world population, are very numerous and promote their important nutritional and environmental benefits. In particular, many authors think that, in this way, it is possible to achieve three of the eight Millennium Development Goals (MDGs) set by the United Nations (2013b) as priority objectives to be met by 2015, namely: 1) combating poverty and hunger in the world; 2) reduce the current high rates of infant mortality due to malnutrition, and 3) orientate the social development towards environmental sustainability (Shockley & Dossey 2014).

However, in this paper, we would need to assess, by an objective and scientific manner, the related advantages and disadvantages and to assess whether the former are more influential than the latter. For example, in many papers it is stated that the consumption of insects, undeniably, lead to an improvement of environmental conditions. However, it is not measured, by the same authors who claim that, what would be the consequences of a massive consumption of insects for the entire ecosystem, especially if this consumption is conducted through the use of massive and indiscriminate capture techniques. It could occur as a consequence, for example, the drastic decline or even the complete disappearance of beneficial insects in some areas, such as pollinators, or the lack of insects that are at the base of the food pyramid in a specific geographic area. Let's examine some of the most important issues.

Advantages

Insects have been consumed in significant quantities for a long time by large segments of the world population; in addition the nutrients contained in hundred species of edible insects (the most edible species, in descending order of importance, belong to the orders of Lepidoptera, Hemiptera, Coleoptera) are often of undoubted biological value (FAO 2013).

Regarding the potential contribution that insects could provide to integrate the diet of the poorest populations and remove, in this way, the risk of malnutrition, many authors believe that this is possible using locally available species of insects with the highest nutritional properties (Premalatha et al. 2011).

In fact, Rumpold & Schlüter (2013) have confirmed that insects have a level of proteins, in particular those with essential amino acids, and fats, especially of the omega-6 and omega-3 groups (linoleic and linolenic fatty acids), comparable to the proteins of milk and meat, which are the foods that currently largely satisfy the world's food needs.

DeFoliart, already in 1992, showed that insects, having a high content of proteins, composed of essential amino acids, such as lysine and threonine, are able to compensate the deficiencies of the cereal protein, notoriously poor in this pattern of amino acids.

Many species of edible insects contain important substances, such as metabolites, enzymes, antioxidants, etc. which, if ingested through the diet, have beneficial effects on human health. The insects are also very rich in fibres and micronutrients (like magnesium, iron, selenium, copper, manganese, phosphorus, zinc, etc.). For example, while beef has an iron content of 6 mg/kg, the value for some locusts is between 8 and 20 mg /kg (Finke & Oonincx 2014).

Still very few and of low scientific level are the studies that have investigated the vitamin content of edible insects. However, Bukkens (2005) has shown that many insects have a good content of various vitamins (A, B1, B2, B12 and D).

With the same energy output provided (caloric content), it seems that insects require fewer feed inputs and a significantly lower availability of land than those needed for farmed animals. In this way, we could potentially increase the current rates of animal production and the availability of cheap proteins.

The mechanized trapping of those insects that live free in the environment, to allocate them for food, can lead, according to some authors, to environmental and economic advantages: for example, limiting, in this way, the use of pesticides, increasing crops yields and integrating the techniques of organic farming (FAO 2010).

Some studies, carried out to measure the energy efficiency of biotransformation of insects compared to vertebrate feedstock, have found that the former have a smaller ecological footprint than the latter (FAO 2013). They have shown, in fact, that insects, being poikilothermic or “cold-blooded” animals, have a great feed conversion ratio (or “efficiency of conversion of ingested food index” or ECI), turning it into edible biomass for humans (Spang 2013). In fact, on average, an insect is able to convert 2 kg of feed into 1 kg of meat, while for a cow it takes about 8 kg (Halloran & Vantomme 2014).

Another point of strength is the wide variety of species that potentially are of interest for human nutrition. It means almost two thousand edible species (Jongema 2012). This great biodiversity offers the possibility to breed, for food purposes, those species with high levels of ECI and low rearing costs.

Moreover insects, as it is well known, reproduce themselves very quickly and easily, even in difficult and not optimal conditions. This involves the availability of a large mass of precious food in a short time.

In terms of environmental protection, in particular, for the purpose of limiting the greenhouse effect, another important benefit is the peculiarity of edible insects to produce, during their metabolic processes, lower levels than cattle of greenhouse gases (GHGs), such as carbon dioxide, methane, ammonia and nitrous oxide.

Oonincx et al. (2010), in fact, have experimentally demonstrated this by measuring the amount of GHGs emitted by insects, being animals that do not need to regulate their body temperature. The same authors have also found that the edible insects, forming part of the panel subjected to analysis, had a relative growth rate or average daily gain much higher than that of the livestock. The insects have, in fact, a greater food production level than cattle, for the same foodstuff consumed.

Van Huis (2013) has also found that insects have a better water footprint because, in addition to consuming less water compared to farm animals, related to the edible substance produced, obtain the water that they need directly from the food and thus have no need for additional amount of drinking water.

Insects, it is easy to understand, do not require to have arable land for the production of their feed, as occurs, for example, to farm animals, since they can consume what remains from agricultural crops: like agro-industrial wastes such as straw, agricultural crop residues, twigs, organic fraction, and so on. In this way, insects could be a realistic “biological converters”, able to transform waste biomass into foods with a high biological value.

From the point of view of environmental efficiency, everyone knows that the cattle breeding is among the major causes of the greenhouse effect, as a result of their emissions of CO₂, SO₂ and CH₄. Well, with the breeding of insects, these problems would disappear, as only cockroaches and termites produce methane.

Disadvantages

The disadvantages, or, at least, the aspects that suggest caution, seem to compensate, in equal measure, the benefits outlined above.

First, while it is relatively easy to obtain the insects needed to feed a small community, by outdoor hunting (harvested from the wild), it is not easy and cheap satisfying, with this method, the food needs of larger human groups, for which there should be the provision of effective industrial methods of breeding (farming) and transformation. In fact, the harvest of insects, especially if conducted as a form of control/prevention of infestations of crops, it requires, as stated by Van Huis (2013), a massive use of labour, preferably cheap, to be able to keep prices low.

Moreover, even if the wild-harvesting of insects could lead to, theoretically, environmental benefits, however, an intense activity of collecting, lasted for long periods and repeated for the same areas, could lead to counterproductive ecological problems, i.e. determining an overharvesting and undermining the balance of the ecosystem. Moreover, it could also result in the collection of insects already contaminated with pesticides or other pathogens, causing, thus, serious health risks to consumers.

Currently, most of the insects used for food are caught or farmed at a family-farm level and sold in small niche markets. The rearing of insects, according to many authors, even if conducted on a large scale, implies higher prices of sale compared to meat from the conventional farms. In the Netherlands, for example, Meuwissen, in 2011, has demonstrated that the breeding cost of mealworms, as animal feed, is 4.8 times greater than that of other chicken feed (Veldkamp et al. 2012).

However, obviously, if we considered, in the calculation of production costs, also the avoided external diseconomies (GHGs emissions, waste, water, etc.), these costs would be, obviously, very competitive. Increased mechanization/automation of rearing processes may cause more remarkable reduction of production and processing costs.

Another important point to consider, in the perspective of predisposition of facilities for insect breeding, is to understand exactly the particular type of feed, which is more suitable from a nutritional point of view and which must be given to insects by the farmer (FAO 2013). There are very few researches at this regard due to the lack of interest about this topic.

An environmental risk, linked to the farming of insects, is the realistic possibility of introducing, deliberately or accidentally, alien species of insects that can be very invasive and competitive against native species, causing destabilization of particular ecosystems.

In addition, the current and most commonly used methods of cooking (roasting and frying) could remove or destroy many nutrients, denaturing fats and proteins (Ekpo 2011). Further studies are therefore required to avoid this and to preserve the nutritional value of these foods.

It will also be necessary to identify – particularly by biologists, entomologists, nutritionists, psychologists and marketing experts – the best way to present these particular foods to the public; for example, transforming them into liquid, solid, pasty, dry or dust forms, in order to preserve the best nutritional value and enhance the perceptual and sensorial characteristics. This will involve the need, from the big brands of food sector, to invest much money in scientific and market research, processing technologies and product packaging, advertising campaigns, etc.

Another problem is the substance of which the exoskeleton consists: the chitin, a polysaccharide. It is therefore an undesirable component, that has to

be eliminated, since its presence, tolerable at very low doses as dietary fibre, can represent an anti-nutritional factor for humans, because we don't have chitinase enzymes like other primates and it can potentially limit the absorption of other nutrients (Raubenheimer & Rothman 2012). It will be required, even in this case, to identify the most correct and effective way to remove the chitin from other edible components, at acceptable costs of processing.

From a safety and health point of view, it is necessary to provide – during the preparation, processing and packaging – proper pasteurization and disinfection processes, in order to eliminate bacteria and spores that can cause health problems to consumers. Obviously, because these diets are “new” for the western industrialized reality, it is necessary to study the most appropriate sterilization technologies in relation to the specific bacterial strains present in these foods. Since it is the terminal part of the intestine of insects the preferred location where they can nest bacteria and other pathogens to humans, sophisticated preparation technology to remove, in addition to chitin, also this part of the body of the insects have to be provided (Shackleton & Shackleton 2004).

Beside the predisposition of transformation and preparation technologies, there is the need to develop unique and clear regulations to determine, in relation to this particular type of food, a set of hygiene characteristics and transformation protocols, methods and techniques of good practice. Before the mass consumption of these sources of food, it is therefore necessary to fill, as soon as possible, this legislative gap, both at national and European level, because the current rules, in their general principles, are limited only to recall general principles of health and hygiene of “traditional” food (Regulations EC No. 178/2002, 179/2002 and 853/2004).

Conclusions

Placing insects into diet of populations and cultures which, like ours, have been alien to these eating habits, would like to say to act on several fronts: first, by investing in promotional and educational campaigns for the proper use of these new foods and then in expensive and sophisticated processing technologies.

Even if it is undoubtedly true that insects have a protein content quantitatively comparable to that of animal feedstock (some species of insects have a protein content equal to 60% of their weight), there is the problem to place systems which eliminate the chitin from other edible proteins. This involves, in addition to a technical complication on the production line, slower process times and higher costs. However, we have to consider that also other

animals, while not having chitin, have many other waste parts which cannot be consumed, such as skin, hair, bones, cartilages, some organs, etc.

Surely these new food choices will be desirable, at least, to diversify our current eating patterns focused on a too small number of food sources.

Another problem, as we have mentioned, is that concerning the food safety. In fact, there is the need to prepare laborious and detailed regulations to protect consumers against possible health risks and to ensure the biological value of the food, for example by updating and developing voluntary regulatory codes such as the Codex Alimentarius.

It is necessary to acquire, especially during the preparation of standards of good hygiene practices, relating to the handling and processing of these elements, a multidisciplinary approach, in order to acquire, especially by the decision-maker, the greatest number of information and to obtain, in this way, a large and correct overview of the situation.

To reach these goals, everyone must be involved: governments, research institutions, universities, corporations, private companies, scientists with different skills, etc. It is necessary, in fact, to provide accurate information and ensure good products, from a sensory and health point of view, to consumers. In this research and dissemination work, the experts of commodity science can also give their important contribution.

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FACTORS THAT INFLUENCE ACRYLAMIDE CONTENT IN CEREAL-BASED BABY FOODS

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Introduction

Thermal processes used in food manufacturing can lead to changes in their composition. The Maillard reaction is considered as one of the most important chemical reaction taking place during food processing. The loss of thermo labile compounds and the formation of compounds that are not naturally present in foods is the most significant consequence of this reaction. Some of Maillard reaction products may have mutagenic, carcinogenic and other high toxicological potential. Well-known toxic compounds formed are heterocyclic amines, nitrosamines and polycyclic aromatic hydrocarbons. Recently, also 5-hydroxymethylfurfural (HMF) and acrylamide (AA) (Claeys et al. 2005; Claus et al. 2008; Bent et al. 2012).

Acrylamide has been added to the list of food-borne toxicants since 2002. The major pathway for acrylamide formation in foods is Maillard reaction with free asparagine and reducing sugars. The acrylamide formation in foods is largely associated with the presence of non-reactive matrix, such as starch or protein substances. Moreover, high temperature (usually above 120 °C) during thermal processing of food, has a decisive impact on AA level in the final product. Other processing parameters, which also influence acrylamide formation are, i.e. the type of raw materials, the product composition, pH, moisture. The highest levels of this compound are formed in foods due to following thermal processes: potatoes frying and browning, cocoa beans and coffee roasting, bread and cakes baking, thermal processing of cereals (Claeys et al. 2005; Zhang et al. 2005; Bent et al. 2012; IFST 2012).

Acrylamide was detected at high concentration exceeding 2000 µg/kg in heat-processed, carbohydrate-rich foods. Most of these foods are particular popular among children. Cereal-based baby foods are an important source of

nutrition for children around the world. However high consumption of these products by infants and children may be a significant source of daily exposure to acrylamide (Fohgelberg et al. 2005; Hilbig et al. 2004; Jiao et al. 2005). Therefore, it is important to understand the reactions leading to acrylamide formation and degradation during heating in cereal-based foods for infants. It is necessary in order to develop methods for decreasing AA content in baby foods.

This study was aimed to evaluate the effects of certain additives, pH, water activity and contents of cereal-based baby foods components on the acrylamide formation. In particular, the effects of natural components (sugars, amino acids, ascorbic acid) were evaluated. The possible role of iron and calcium ions in the formation of acrylamide, were also examined. The study was conducted in order to design strategies to prevent or decrease the extent of AA formation in baby foods.

Material and methods

Chemicals. The acrylamide standard ($\geq 99.8\%$, catalogue no. 23701) was obtained from Sigma-Aldrich (St. Louis, MO, USA) and d_3 -acrylamide standard from Polymer Source Inc., Dorval, Quebec, Canada. Acrylamide and d_3 -acrylamide standard solutions were prepared in 80% methanol. L-Asparagine, D-glucose, D-fructose, sucrose, ascorbic acid, L-lysine and CaCl_2 , $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$, all of analytical grade and for HPLC, were purchased from Sigma-Aldrich (St. Louis, MO, USA). Acetonitrile, methanol, hexane and all other chemicals and solvents used in this study were of HPLC analytical grade and were obtained from Sigma-Aldrich (St. Louis, MO, USA) and from Merck (Darmstadt, Germany).

Preparation of model system. The preparation of the model systems was based on the method described by Becalski et al. (2003). For modeling studies data on nutrient concentrations in cereal-based baby foods was used. Initially, a mixtures of following eleven major amino acid: asparagine, aspartic acid, glutamine, glutamic acid, valine, lysine, serine, alanine, glycine, cysteine, methionine and six sugars: galactose, fructose, glucose, maltose, lactose, sucrose present in cereal-based baby foods was used. Most of these was shown to be the most active in the Maillard reaction. Therefore the model was simplified and consisted only of asparagine and glucose, fructose and sucrose. Three model systems were prepared by weighing and mixing different levels of asparagine monohydrate and sugar powders (asparagine-glucose, asparagine-fructose and asparagine-sucrose). The independent variables were heating temperature (heat treatment was performed at 100, 120, 140, 160, 180, 200 °C in oil bath with thermostat), heating time (1, 5, 10, 20, 30, 60 min),

certain additives (iron and calcium ions, ascorbic acid and lysine, 0.1 – 5%, 0.1 - 5%, 0.2 – 15% and 10% were added, respectively, before the start of reaction to test their inhibition effect on acrylamide production.), pH (pH of the model systems was adjusted by addition of NaOH or HNO₃ in a pH range between 3 and 9. The pH was measured by using a CyberScan PH 510 from Eutech Instruments Europe B.V., Nijkerk, Netherlands.), water activity (The model systems were lyophilized using Christ Alpha 1-2 LD plus from Osterode am Herz, Germany. Direct addition of increasing volumes of water to the lyophilized model was performed and the water activity varied from 0.2 to 0.9. The lyophilized model systems was stored in vacuum jar to avoid any water adsorption. The measurement of water activity at 25 °C before heat treatment was performed using LabMaster-a_w from Novatron, London, England.), asparagine monohydrate and sugar (fructose, glucose monohydrate and sucrose) according to molar ratio (1:1, 1:2, 2:1, 1:4, 4:1), and their interactions between each other were also taken into consideration. Each sample was prepared in triplicate. The independent variables were important factors which significantly contribute to the generation of acrylamide, such as heating temperature, heating time, molar quantity of asparagine monohydrate, level of the selected additives, pH level and water activity, presence of particular amino acids and sugars in the mixture and their interactions between each other.

Analysis of acrylamide. Prior to analysis, samples were diluted in 80% methanol in water (1:5 - 1:10) and filtered on a 0.2 µm nylon syringe filter (Millipore from Merck, Darmstadt, Germany). The determination of acrylamide concentration was performed using the ion-pair RP-HPLC method developed by Michalak, Gujska and Kuncewicz (2013). The following instrumentation was used: a Shimadzu LC-10A series HPLC (Kyoto, Japan) equipped with a UV-Vis 190–600 nm diode array detector (DAD model SPD-M20A, Shimadzu) set at 200 nm. The column used was a Synergi Hydro-RP 80 A 4 mm 250 x 4.6 mm (from Phenomenex, Torrance, CA, USA). The operating conditions were as follows: the mobile phase was an isocratic mixture of 5 mM sodium 1-heptanesulfonate in water/acetonitrile (97/3, v/v); flow rate, 1.0 mL/min; injection volume, 100 µL; column temperature, 25 °C. Peak identification was based on the retention time using a comparison of the ratio of UV spectra with that of a standard and quantified by the external standard procedure, using a calibration curve.

Statistical analysis. Data analyses were performed by using Student's t-test with the Statistica 9.0 software package and Duncan multiple range using SPSS program package. Analysis of variance (ANOVA) was tested on a significance level of p=0.05.

Results and discussion

The role of amino acids and sugars in acrylamide formation. In our studies data on nutrient concentrations in cereal-based baby foods was used. The results showed (data not shown) that acrylamide was not detected in mixtures of amino acids and sugars heated when asparagine was not present in this mixtures. Therefore, the presence of asparagine was the main factor for acrylamide formation in foods. Acrylamide was produced only when asparagine and glucose, fructose or sucrose were used. Therefore the model was simplified and consisted only of asparagine (Asn), and glucose (Glu), fructose (Fru) and sucrose (Suc). Three equimolar model systems were prepared by weighing and mixing of asparagine monohydrate and sugar powders (asparagine-glucose, asparagine-fructose and asparagine-sucrose) and were heated at 100, 120, 140, 160, 180, 200 °C in glass tube for 1, 5, 10, 20, 30, 60 min and the formation of acrylamide was monitored. The yield of acrylamide from different model systems is shown in **Figure 1**. Our studies also showed fructose to be much more efficient than glucose in generating acrylamide. This means, that acrylamide is formed from fructose at lower temperatures. Mixtures with fructose generated acrylamide at lower temperature (120 - 200°C) then mixtures with glucose, in which acrylamide was formed at higher temperatures (140 - 200°C). Acrylamide was not detected in samples with fructose heated at 100 °C and in samples with glucose heated at 100 and 120 °C. This may be related to the difference in these sugars structures. It is well known that the structural features of the carbonyl reactants are important for the Maillard reaction. In our studies, glucose, which is considered to be more reactive in Maillard chemistry, leads to the formation of relatively lower levels of acrylamide than fructose. This may be related to the fact that fructose has two α - hydroxyl groups and glucose only one, which might be relevant for the reactions between the reducing sugars and asparagine in the Maillard reactions. However, also other parameters than chemical reactivity may play a role to explain this phenomenon. Although many researchers have found that acrylamide is mainly formed when a specific amino acid reacts with a reducing sugar in the presence of heat, in our study the reaction of sucrose and an amino acid in the presence of heat resulted in acrylamide formation comparable to the levels formed by fructose and glucose. Asparagine needs sucrose at relatively higher temperature to increase the amount of acrylamide.

Our studies showed that acrylamide was also formed in systems which sucrose was present at 140 – 180 °C. Sucrose is a non-reducing sugar and should not participate in the acrylamide formation. However, sugar such as sucrose, can easily hydrolyse upon heating at high-temperature and at slightly acidic pH, resulting in the formation of monosaccharaides. In theory, one sucrose molecule could give rise to two reducing hexoses resulting in a molar ration of 2:1 sugar to amino acid. If sufficient reactants are present to react with the α -NH₂ group of asparagine in the Maillard reaction pathway,

acrylamide can be formed. As to the reactivity, Stadler et al. (2002) found fructose, galactose, lactose and sucrose to release AA with comparable yields. On the other hand, Becalski et al. (2003) observed a lower relative yield of AA from a dry asparagine-sugar system when sucrose was used compared to glucose or fructose. Also the addition of 40% sucrose to wheat flour did not result in increased AA levels (Biedermann et al. 2003). Comparing glucose and fructose, fructose appeared to result in a higher AA yield than glucose in an asparagine-sugar system (Biedermann, et al. 2002a; Biedermann & Grob, 2003; Becalski et al. 2003).

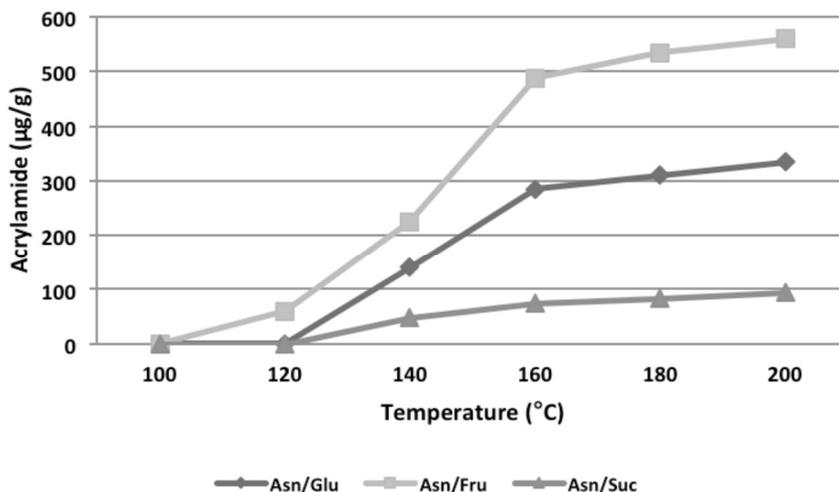


Figure 1. Formation of acrylamide by heating equimolar mixtures Asp/Glu, Asp/Fru, Asp/Suc for 5 min

Source: own research.

The effect of molar ratio of asparagine to sugars. Mixtures of asparagine/sugars (Asn/Glu, Fru and Scu) were prepared according to molar ratio: 1:1, 1:2, 2:1, 1:4, 4:1. To study the effect of initial reactant concentration on the kinetics of acrylamide formation and to evaluate the effect of an excess of one of two main reactants on the course of acrylamide concentration, the initial reactants molar ratio in mixtures was changed from 1:1 to 1:2, 2:1 and 1:4 or 4:1. All mixtures were heated at 140, 160, 180, 200 °C for 20, 30, 60 min. The acrylamide yield depended on the initial reactant ratio. In the model systems with an excess of asparagine, the yield was the highest. Whereas the acrylamide yield in the model system with an excess of glucose, fructose and sucrose, was higher than in the equimolar model systems. In all model systems tested, the initial rate of acrylamide formation increased with increasing temperature, and the maximal acrylamide concentration was attained in all samples with different molar ratios heated at 180 °C for 30 min. But in the

samples in which molar ratio was 4:1 (asparagine/sugars) the acrylamide concentration was the highest. When the molar ratio of asparagine to sugars was 4:1 and heated at 180 °C for 30 min the average yield of acrylamide was fivefold and threefold higher than in the samples at molar ratios of 1:1, 2:1 respectively was four times higher than in samples in which molar ratio was 1:2, 1:4 as shown in **Figure 2**. The results showed that the availability of asparagine in the reaction medium may be the main limiting factor for the formation of acrylamide in food. ANOVA analysis of variance of the asparagine–sugar type of model showed that the molar quantity of asparagine monohydrate became the most important parameter contributing to the formation of acrylamide. By changing the reactant ratio in models system, the rate constant for acrylamide formation remained within the 95% confidence interval. The coefficient of correlation between amount of Asn and the formed amounts of AA in models was 0.731, demonstrating that participation of Asn in the model plays a role in AA formation. The asparagine content represents 14% and 18% of the total free amino acids of wheat flour and high-protein rye, respectively (Mottram et al. 2002). Some authors suggest that even though amino acids in general and asparagine in particular play a primary role in the formation of AA in heat-treated food, the AA content seems to be little or not at all correlated with the total amino acid content or with the concentration of asparagine in products (Amrein et al. 2003). However, in wheat and rye flour, a high correlation between the AA yield and the level of asparagine was observed (Surdyk et al. 2004).

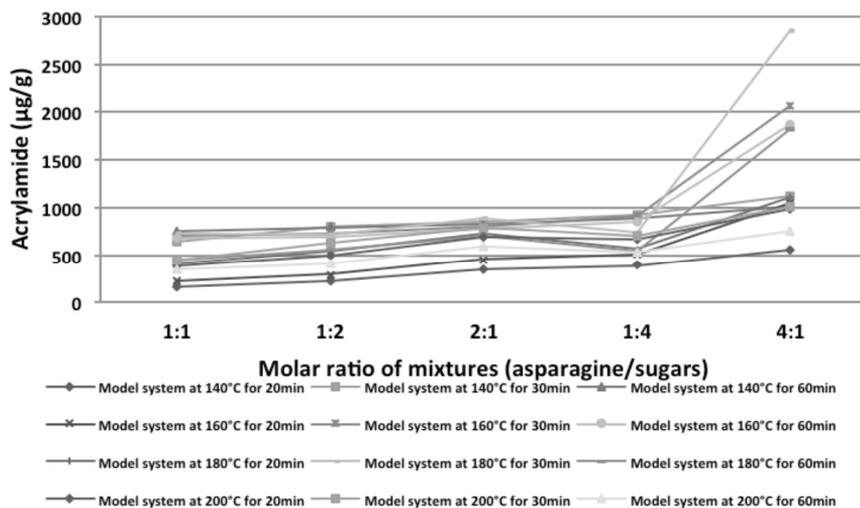


Figure 2. The acrylamide yields from mixtures (asparagine/sugars) with variable reactant ratio after heat treatment at different temperatures and times

Source: own research.

The effect of reaction time and temperature. Heat treatment was performed at 100, 120, 140, 160, 180, 200 °C for 1, 5, 10, 20, 30, 60 min and the formation of acrylamide was monitored. As shown in **Figure 3** and **Figure 4**, these parameters strongly influenced acrylamide formation. Acrylamide was not detected in unheated control samples. Acrylamide was also not detected in samples heated at 100 °C for 1-60 min. Study showed that acrylamide is formed at about 120 °C at long reaction times. The yield of acrylamide from model systems depends on the chemical reactivity of sugars. In samples with fructose acrylamide was generated earlier than in those containing glucose or sucrose. It means that acrylamide is formed at lower temperature, about 120 °C. In case of glucose and sucrose, about 140 °C. Sucrose generates very low amounts of acrylamide at 140 °C compared to fructose and glucose. The highest yield of acrylamide in the samples was obtained by heating at 160 and 180 °C for which the maximum yield was reached after 30 min. In all model systems tested, the initial rate of acrylamide formation increased with increasing temperature, and the maximal acrylamide concentration was obtained in the samples heated at 180 °C for 30 min. In the samples heated at 140, 160 °C content of acrylamide was 31 and 13% lower than when heated at 180 °C. In case of samples heated at 200 °C it was also lower of about 10% than when heated at 180 °C.

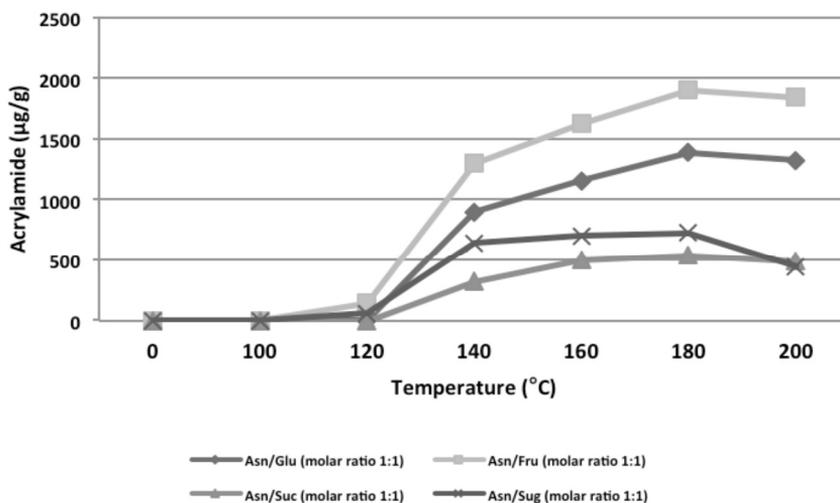


Figure 3. Acrylamide formation as a function of temperature in mixtures heated for 30 min

Source: own research.

In the present work, the graph of acrylamide amount generated in the model systems showed a decline tendency in the combined condition of

elevated temperature and extended time. The decrease of acrylamide concentration, observed in all model systems tested at temperatures above 180 °C and particularly in temperature at 200 °C after prolonged heating (60 min). It is probably a consequence of reaction of acrylamide with other components formed within the model systems. Higher temperature (200 °C) combined with prolonged heating time produced reduced levels of acrylamide, due to elimination/degradation processes. In our study, we found a strong correlation between heating temperature from 0 to 180 °C and acrylamide ($r = 0.696$, $p < 0.05$) and a lower but significant correlation between heating time from 0 to 30 min and acrylamide ($r = 0.404$, $p < 0.05$). The influence of temperature and time on the formation of acrylamide has been repeatedly demonstrated. Eriksson (2005) reported that time and temperature is of great importance in the formation of acrylamide in heat-treatment. Prolonged heating time decreases the acrylamide content.

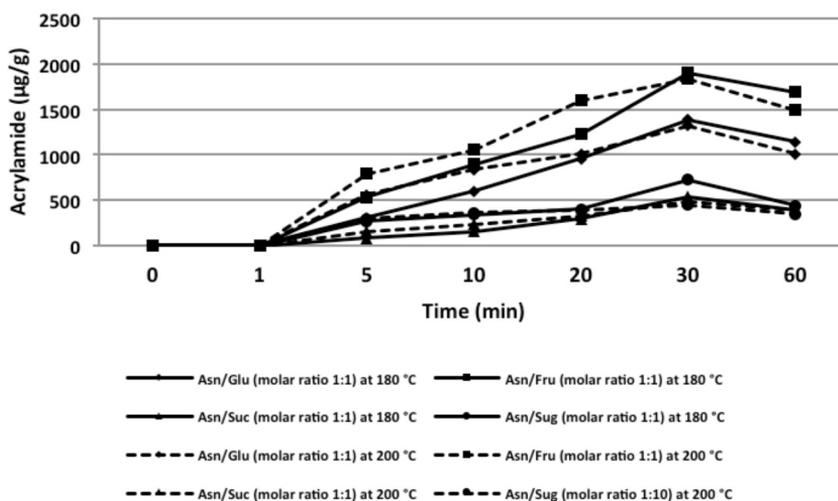


Figure 4. Acrylamide formation as a function of time in mixtures heated at 180 °C and 200 °C

Source: own research.

The influence of lysine on acrylamide formation. Subsequently, the impact on acrylamide formation of L-lysine was investigated. Many studies have shown that the reactivity among the diamino acids increased with the length of the carbon chain. Among the amino acids studied lysine was most reactive in Maillard reaction (Lingnert et al. 2002). But this amino acid might also reduce acrylamide formation by competing with asparagine to react with reducing sugars in the Maillard reaction. Our results indicate that the efficiency of free amino acids to decrease the acrylamide content may depend

on the experimental set-up such as, for example, the heating conditions. As shown in **Figure 5**, the concentrations of acrylamide in samples in which was the addition of lysine, decreased with heating temperature. In the samples heated at 140, 160, 180 and 200 °C the losses of acrylamide were 11, 21, 21 and 28%, respectively. The study showed that when lysine was applied at the amount of 10%, acrylamide decreased significantly (t-test) on average on 23% compared to the blank. In conclusion, lysine applied proved to be a suitable and effective tool for acrylamide minimization in products. Rydberg et al. (2003) reported that the addition of other free amino acids or a protein-rich food component strongly reduced the acrylamide content, probably by promoting competing reactions and/or covalently binding of formed acrylamide.

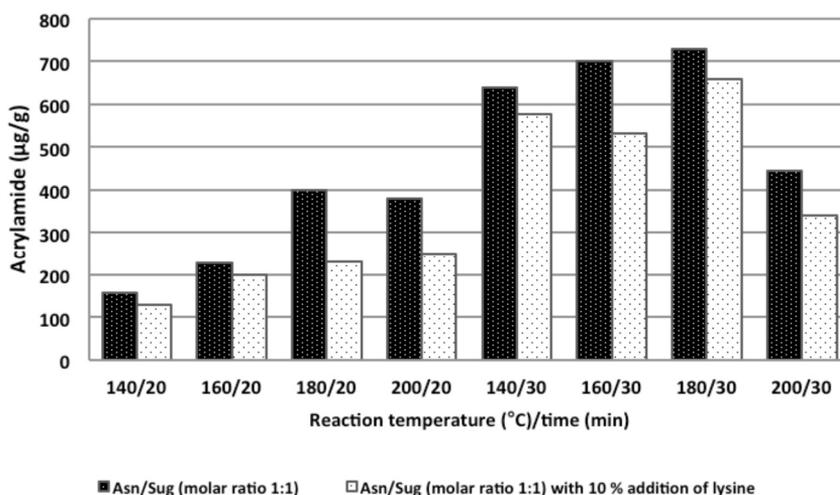


Figure 5. Acrylamide contents with 10% addition of lysine in the model systems

Source: own research.

The influence of pH on acrylamide formation. Formation of AA is like the Maillard reaction dependent on the pH of the system. The pH will influence the reactivity of both the sugar and the amino group. A higher pH favors the open chain form of the sugar and the unprotonated form of the amino group to reactivity. The formation of acrylamide was investigated as a function of pH by heating mixture with different initial pH in a closed glass tube. Three equimolar model systems were prepared by weighing and mixing of asparagine monohydrate and sugar powders (asparagine-glucose, asparagine-fructose and asparagine-sucrose). The pH of the model systems was adjusted by addition of NaOH or HNO₃ to pH between 3 and 9. Prior to

heating, the pH was measured by placing the pH electrode directly into the mixture. Subsequently, 1 g of the mixture in glass tube was heated. Heating was performed for 30 min at 180 °C. With these heating conditions, acrylamide could be quantified over the whole range of investigated pH values. After heating, a quick cooling was established, submerging the glass tube in the ice bath for 5 min. Finally, the 1 g mixture was analyzed for acrylamide content. All reported acrylamide levels are the average of at least three heating experiments. The acrylamide contents of the mixtures, heated at 180 °C for 30 min, are shown in **Figure 6**. This investigation showed a significant impact of the mixtures pH on the acrylamide formation, based on 95% confidence interval. These results indicate that acrylamide formation in mixtures with glucose and fructose showed an optimum between pH 7 and 8.

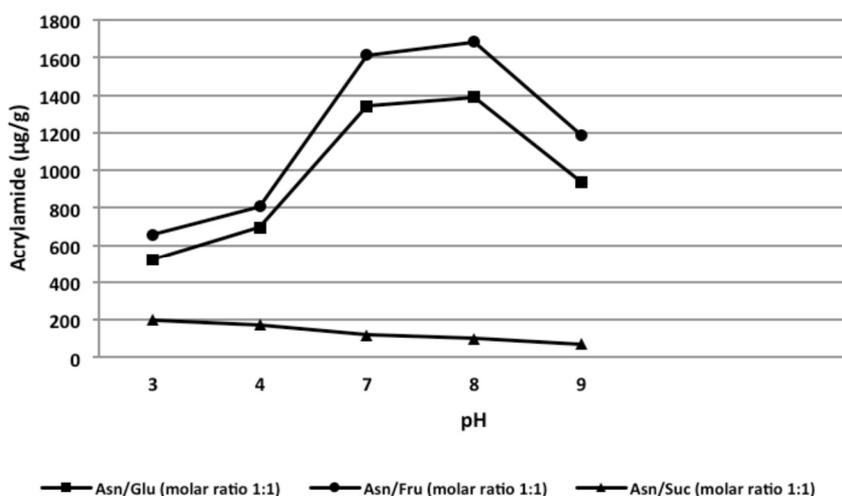


Figure 6. Acrylamide formation as a function of pH in the model systems, heated at 180 °C for 30 min

Source: own research.

The pH-dependence of acrylamide formation shows a maximum around pH 8; higher and lower pH enhanced elimination and decelerated formation of acrylamide. The clear decrease in acrylamide content was noted upon acidification of the samples. The acrylamide content decreased of about 60%, 50% when the pH was adjusted to 3, 4 respectively, compared to the mixtures in which pH was 7 and 8. The acrylamide content also decreased of about 30% in mixtures when the pH was adjusted to 9, compared to the mixtures in which pH was 7 and 8. In addition, it was demonstrated that sucrose generates highest amounts of acrylamide at pH 3 - 4. In this case, the increase of acrylamide content (about 40%) was noted with acidification of the mixtures.

This may result from the fact, that sucrose can easily hydrolyse upon heating at high-temperature and at acidic pH. This results in the formation of monosaccharides, which are considered to be more reactive in Maillard reaction, leading to the increased formation of acrylamide than sucrose, which is a non-reducing sugar. Correlation analysis was performed to investigate the strength and direction of the relationship among the pH of the model systems with fructose, glucose or sucrose and acrylamide concentration. A positive correlation was found between pH of 3 - 8 and acrylamide formation in the mixtures with fructose and glucose ($r = +0.894$ and $r = +0.886$ respectively, $p < 0.05$), but a negative correlation at the same pH and acrylamide formation in the mixtures with sucrose ($r = -0.654$, $p < 0.05$). The results of these experiment emphasize the importance of controlling sample pH. The optimum pH for AA formation of suggested by many researchers is around 7–8 (Broda & Alexe 2011; Rydberg et al. 2003). One of the factors altering the pH is heating. Heating can cause a decrease in pH because of increased water dissociation (Gertz & Klostermann 2002).

The role of water activity (a_w) in acrylamide formation. In this study, the effect of initial water activity on acrylamide formation was evaluated. Water activity and temperature effect are being linked ($T_{sat}=f(a_w)$). Thus, it is difficult to separate the influence of each parameter in acrylamide formation/degradation reactions during heat treatment. Besides, the effect of water activity on acrylamide formation remains unclear and sometimes contradictory, because the experimental procedures and the physical state of the reactants reported in the literature vary considerably. In our study the formation of acrylamide was investigated as a function of the water activity of the heating mixture with different initial water activity in a closed glass tube. Three equimolar model systems were prepared by weighing and mixing of asparagine monohydrate and sugar powders (asparagine-glucose, asparagine-fructose and asparagine-sucrose). The model systems were lyophilized. Direct addition of increased volumes of water to the lyophilized model was performed and the water activity varied from 0.2 to 0.9. The lyophilized model systems was stored in vacuum jar to avoid any water adsorption. Their water activity was measured at 25 °C and then, immediately, 1 g of the mixture in glass tube was heated. Heating experiments were performed for 30 min at 180 °C. After heating, the glass tubes filled with the final reaction products, were taken out from the oil bath and immediately cooled in prepared ice water to stop any further reaction. The whole cooling process was performed in a room with stable air temperature and relative humidity. **Figure 7** shows the changes in acrylamide formation affected by initial water activity. This investigation showed that water has both an inhibitory and an accelerating impact on the acrylamide formation. Water may act partly as a reactant and partly as a solvent and transporting medium of reactants (reactant mobility). The results from our studies in model systems

showed the a_w between 0.4 and 0.7, to be favourable to acrylamide formation. It was also found that acrylamide formation did not vary significantly at the water activity of 0.4 – 0.7 and that the activation energies did not vary significantly within the initial and final water activity (0.2 and 0.9), based on 95% confidence interval. In all model systems it was found, that the constant rate of acrylamide formation showed a clear minimum at 0.9 and 0.2 a_w . Probably the main explanation for an optimum reaction rate at an intermediate a_w , is that the reactants are diluted at the higher a_w , while at a lower a_w the mobility of reactants is limited, despite their presence at increased concentrations. At high a_w the reactivity of reactants is also limited due to high dilution of the reactants.

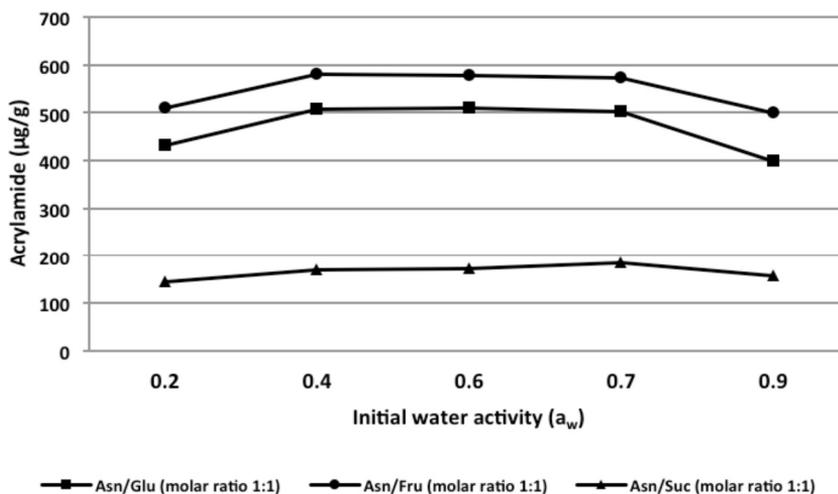


Figure 7. Acrylamide formation as a function of initial water activity (a_w) in the model systems, heated at 180 °C for 30 min

Source: own research.

On the basis of different studies AA formation seems to be mainly a surface phenomenon. It was found that in real food (cake) acrylamide formation did not occur in a large extent when moisture content was higher than 5% (Elmore et al. 2005). It was also found that an increase in acrylamide formation/elimination rate constants when initial water activity decreased (Bassama et al. 2011). Robert et al. (2004) suggested that the formation of acrylamide cannot be correlated with water activity.

The effect of ascorbic acid on acrylamide formation. The purpose of this investigation was to determine the influence of the addition of ascorbic acid solution at different concentrations on AA formation in asparagine/fructose, asparagine/glucose and asparagine/sucrose equimolar

model system. Natural antioxidants may be another important way to reduce AA content in Maillard model system and in food matrix. However, there were some contradictory reports on this topic from different researchers. Some researchers found that addition of antioxidants including ascorbic acid, significantly reduced acrylamide formation in the equimolar asparagine–glucose model system. While other researchers observed no correlation between acrylamide formation and antioxidants including ascorbic acid in a thermally processing model system of asparagine and glucose. To clarify the disagreement among reported observations, addition ascorbic acid was used to investigate its effect on elimination and formation of acrylamide in model reaction systems. In our study, the model systems (asparagine/sugars: Asn/Glu, Fru and Suc to molar ratio: 1:1) were prepared with the addition of ascorbic acid solution at different concentrations of 0.2%, 0.5%, 1.5% and 15%. In order to eliminate the effect of different initial water activity on acrylamide formation the model systems were lyophilized. After this, the water activity was 0.7 in the test groups and identical was in the control group. The reactant mixtures in hermetically sealed glass tubes were all heated in the oil bath at 180 °C for 30 min. After heating, the glass tubes filled with the final reaction products were taken out from the oil bath and immediately cooled in prepared ice water to stop any further reaction. The whole cooling process was performed in a room with stable air temperature and relative humidity. The acrylamide contents were determined in the cooled reaction products. Based on the quantitative results of acrylamide contents in mixtures, the relationship between the reduction of acrylamide contents affected by the addition of different levels of ascorbic acid solutions could be observed. The effect of ascorbic acid on AA formation was shown in **Figure 8**.

In Asn/Glu model system, the reduction rates were 0%, 1%, 3%, 8% and 11% after the addition of ascorbic acid solution at the concentrations of 0.2%, 0.5%, 1%, 1.5% and 15%, respectively. Similar results were obtained in the Asn/Fru model system. In Asn/Suc model system, ascorbic acid had no influence on AA formation. When the concentration of ascorbic acid was 15%, the highest reduction rate of AA formation was found, and the reduction rate was 4%. In our study, ascorbic acid showed no effective reduction effect on AA formation in all model systems, especially when the addition of ascorbic acid was lower than 1.5%. Significant difference could be only found between the addition of high concentration of ascorbic acid solution (1.5% and 15%) compared with the model systems without the addition of ascorbic acid ($p < 0.05$). Addition of the ascorbic acid caused a decrease of acrylamide content which could be explained by lowering the pH or increased binding water. Ascorbic acid is well-known antioxidant. However, in our study the ascorbic acid showed no high effective reduction effect on AA. The mixtures containing ascorbic acid had a lower pH (3.70 to 6.50) compared to the control mixtures (6.50). It was found that the addition of ascorbic acid lowered the pH

of the mixtures and accordingly the acrylamide content, but no significant reduction, apart from the lower pH, was assessed. This suggested that ascorbic acid may cause an inhibiting effect on acrylamide formation, primarily through the pH effect. Contradictory results were published on the impact of different types of antioxidants on acrylamide formation. Biedermann et al. (2002b) reported a weak decrease of AA formation by the addition of ascorbic acid (1%) to a potato model. Ascorbic acid seemed neither to inhibit AA formation, nor to influence AA elimination significantly. Rydberg et al. (2003) observed a significant reduction in AA content when 1.7% ascorbic acid was added to a potato sample.

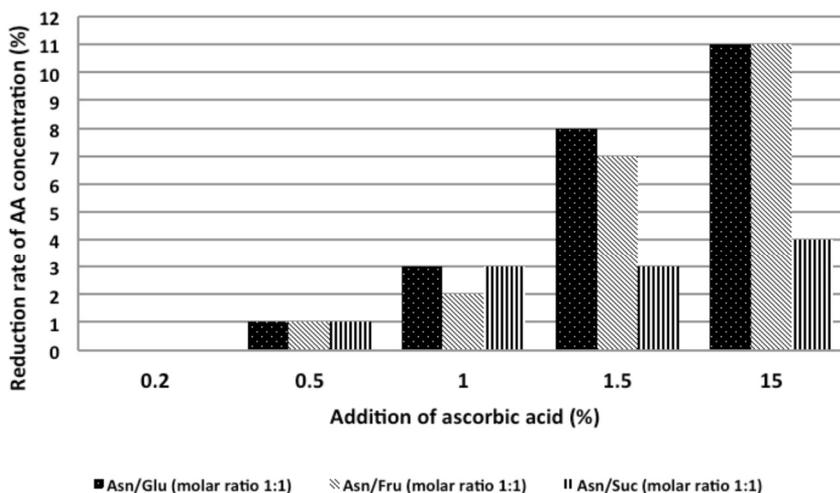


Figure 8. AA contents with different addition concentrations of ascorbic acid in model systems, heated at 180 °C for 30 min

Source: own research.

The effect of some cations on acrylamide formation. A recent patent application also showed that polyvalent cations are able to reduce some compounds formation during heating. For this reason, this study was aimed at investigating the effects of divalent and trivalent cations on acrylamide formation in an asparagine-glucose, asparagine-fructose and asparagine-sucrose model systems. In our study, the model systems (asparagine/sugars: Asn/Glu, Fru and Suc to molar ratio: 1:1) were prepared with the addition of CaCl_2 and $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ to mixtures at 0.1%, 0.5%, 1%, 1.5% and 5%. Then, as indicated above.

As shown in **Figure 9 and 10**, addition of cations significantly influenced the formation of acrylamide. The presence of Ca^{2+} and Fe^{3+} cations in the reaction mixture decreased acrylamide formation. The cations decreased the

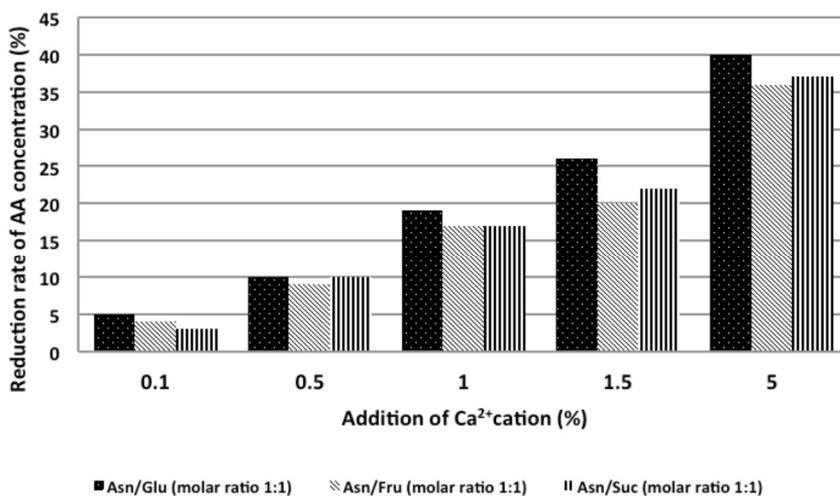


Figure 9. AA contents with different addition concentrations of Ca²⁺ cation in model systems, heated at 180 °C for 30 min

Source: own research.

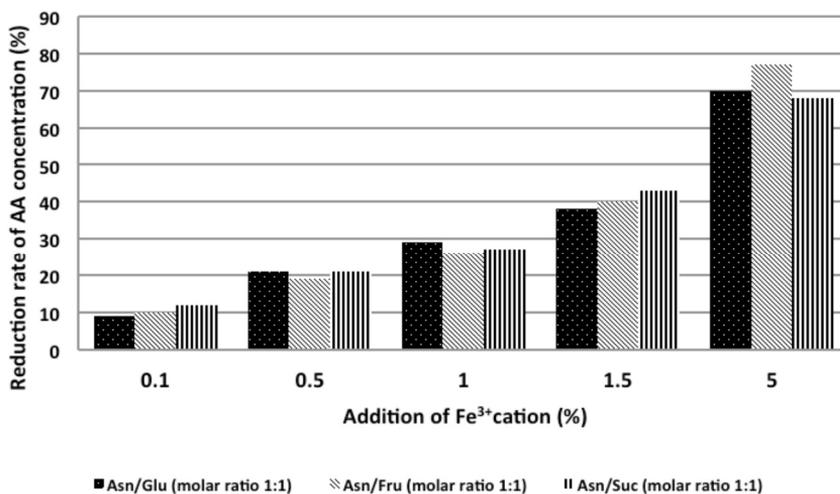


Figure 10. AA contents with different addition concentrations of Fe³⁺ cation in model systems, heated at 180 °C for 30 min

Source: own research.

amount of acrylamide formed in the reaction mixture when their concentration increased from 0.1% to 5%. Increasing the amount of Ca^{2+} and Fe^{3+} ions from 0.1 to 5% decreased acrylamide formation during heating of the reaction mixtures by 36 - 40% and 68 - 77%, respectively. The significant differences with the control mixture was observed, confirming the efficient acrylamide reducing effect of calcium and iron cations ($p < 0.05$). Ca^{2+} and Fe^{3+} ions acrylamide lowering effect may be due to the binding of ions to asparagine in order to prevent the Schiff base formation, which is a key step in acrylamide formation. Adding divalent cations such as Ca^{2+} or Mg^{2+} to dough had a strong effect on acrylamide content. Elder et al. (2004) reported an almost 20% reduction in acrylamide when these ions were added and a decrease of 50% when slightly acidic conditions ($\text{pH} = 5.5$) were provided. Gökmen and Senyuva (2007) reported a negative correlation between acrylamide formation and added Ca^{2+} concentration in potato products.

Conclusions

In our study we demonstrated that acrylamide occurs in heated food products for baby, with unexpectedly high levels in certain cereal-based baby foods. Our results are in agreement with earlier findings from model systems in which acrylamide formation was strongly influenced by the presence of sugars and asparagine.

The acrylamide content of cereal-based baby foods is the result of complex reactions leading to both, the formation and degradation of this molecule. The study showed, that it is possible to reduce acrylamide content even up to 70 - 80% in model systems. We showed that by selecting material low in sugars and low in asparagine, acrylamide content can be substantially reduced when using an industry standard procedure and a heating temperature of 180 °C.

Pilot acrylamide reduction projects should be introduced especially for foods for particular nutritional uses (products for infants and children). Currently, there is no practical and effective method for eliminating acrylamide from food products and even if the occurrence of acrylamide can be reduced, there will not be one solution applicable to each food type. Moreover, many measures capable of reducing the acrylamide content may have an adverse effect on the taste, color or consistency of products. Consequently, it is important to develop and introduce methods for reduction of the occurrence of acrylamide in foods while preserving their sensory characteristics. Therefore, all modifications connected with the raw material or process conditions should also be considered from the consumers' perspective and laboratory tests should be combined with research covering both consumer and commercial aspects. Even if the full extent of the reduction

could not be achieved for commercial reasons (e.g., organoleptic properties), our study shows the importance of controlling some precursors of acrylamide.

Acknowledgments

This work was supported by the Polish Committee for Scientific Research (project no. N N312 227236, 2272/B/P01/2009/36).

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LABELLING OF GENETICALLY MODIFIED FOOD PRODUCTS

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Introduction

Genetically modified food is foodstuffs' category that increases quickly worldwide and is widely launched into the market. Every year Poland imports from the USA, Argentina and other countries of the world about 2.5 million tons of grain (soy, corn, canola). In more than 90% they are genetically modified plants. Currently, high-protein products such as soybean meal are the basis of animals' feed (de Jong 2010, Anioł, Pruszyński & Twardowski 2007).

The source of GMOs may be vegetable oils (corn, soybean, canola), chips, vegetarian burgers, meat substitutes, including hot dogs for kids, ice cream, yogurt, tofu, soy sauce, soy cheese, tomato sauce, protein, cereals, burgers, hotdogs, margarine, mayonnaise, cereals, crackers biscuits, chocolate, candy, baking powder, alcohol, vanilla, powdered sugar, peanut butter, enriched flour, pasta. Soy protein concentrates are added to the majority of highly processed foodstuffs, e.g. to sausage or yoghurt, and most of soybean which use Polish processing plants is transgenic. Thus, the probability of consuming products containing GMOs, especially GM soy is very high (de Jong 2010). This raises a lot of controversy not only among scientists in biotechnology, but also among the average consumers, the more that European consumers have shown and still show a clear preference for non-GMO products (Cardwell 2010, Badora 2013). The same trend even stronger can be noticed in Poland. In addition to the concerns about the consequences of GMOs on human health, the impact of GMOs on the environment is a concern, in particular their effect on biodiversity and existing species (Badora 2013, Twardowski 2012, Pytka, Kordowska-Wiater & Stój 2014). It happens even though the European Commission, based on a number of opinions of experts, has stated that „products GMOs do not pose any other threat to our society and the environment than conventionally sourced materials, and strict evaluation criteria lead to the conclusion that these

products are actually safer than classical ones“ (Anioł, Pruszyński & Twardowski 2007, de Jong 2010).

Studies of the acceptance of food products produced with the use of GMO tend to divide buyers into two groups: those that accept or are indifferent to the presence of GMOs and those who reject it. Some buyers care about the presence of GMOs but also care about other quality attributes such as environmental impact, nutritional quality, and convenience of use. They will buy a GMO products if safety is assured (Caswell 2000, Anioł, Pruszyński & Twardowski 2007, Badora 2013).

Generally the Poles have a great distrust of the food produced on the basis of genetic engineering achievements. Despite the lack of clear evidence about the dangers of consuming genetically modified organisms, short history of the use of these modifications, popularization too general knowledge, as well as known from the past various types of adverse effects of use of inventions such as insecticides and medicines, seem to explain this attitude. The belief that the Polish accession to the European Union made the food in our country less safe than it used to be before favours them also (Pułaska-Borowicz 2009, Badora 2013, Twardowski 2012, Pytka, Kordowska-Wiater & Stój 2014).

GMO labelling was introduced to give consumers the freedom to choose between GMOs and conventional products. Since 1997 Community legislation has made labelling of GM food mandatory for products that consist of GMO or contain GMO and for products derived from GMO but no longer containing GMO if there is still DNA or protein resulting from the genetic modification present. Due to the EU Regulation 1829/2003 on genetically modified food and feed all food (including processed food) or feed that contains greater than 0.9 percent of approved GMOs must be labelled. It is especially important in the context of the fact that the great majority of European consumers has shown and still shows a clear preference for non-GMO products (Heslop 2006, Wrześniewska-Wal 2013, Viganì, Raimondi & Olper 2010)

Mandatory GMO labels are helpful to buyers who wish to avoid or find products produced with the use of biotechnology. For these buyers, GMO presence or absence equate with low quality and some with high quality. Mandatory labels help these consumers find the product that they consider “high quality.” The drawback with mandatory labels is that everyone, even those who do not care, bears the cost of testing, certification, and labeling (Caswell 2000).

Main aim of the paper was to present the basic EU requirements on labelling of food products with GMO. In the experimental part of the paper the packages of 75 food products available in Malopolska Region, especially in Cracow hypermarkets potentially containing GMO or produced with GM engineering techniques were analysed in the context of presence of the

appropriate label. Special attention was paid to soybean products as soy protein play very important role in the foodstuffs manufacturing and the EU imports 35 to 40 million tones of soy annually, while about 90% of it is GM. The analysis covered the packages of: baked goods, breakfast cereals, pasta, beverages and toppings, meat, poultry and fish products, diary products as well as soybean meat substitutes. Consumers' attitude towards foodstuffs with the label "with GMO" and "GMO free" was also determined on the basis of the survey carried out in Malopolska Region on February 2014.

Labelling of genetically modified food products in the EU

The EU recognizes the consumers' right to information and labelling as a tool for making an aware choice between GMOs and conventional products. Essentially, if a foodstuff is produced using genetic engineering, this must be indicated on its label. Actual labeling practice, however, is far more complicated. It must be planned and regulated taking into account issues such as feasibility, legal responsibilities, coherence and standardization (Premanandh 2011, Gruère 2006, Gruère, Carter & Farzin 2009).

In 1997, the EU introduced GMO labeling policies with the purpose of guaranteeing 'the consumer's right to know', but labels carry indications other than just the presence of GM ingredients, they also give information on health factors and product diversification (Veysièrè & Giannakas, 2004). Labeling has also met environmental issues, playing a role in consumption decisions of consumers concerned by environmental factors associated with GM products. Indeed, a label can act as a warning, indicating potential hazards and thus affecting the demand for GM and non-GM products, particularly reducing the demand for the ones including GMOs (Gruère, 2006).

According to article 12 paragraph 2 of the EU Regulation No 1829/2003 labeling requirement specifying that the product contains GMOs applies to foods containing material which contains, consists of or is produced from GMOs in a proportion greater than 0.9 percent of the food ingredients considered individually or food consisting of one component, provided that this presence is adventitious or technically unavoidable (European Commission 2003a, European Commission 2003b). It clearly means that all food (including processed food) or feed that contains greater than 0.9 percent of approved GMOs must be labeled (Davison 2010). It includes:

- Food which is a genetically modified organism (GMO) or which consists of GMOs - it can be GM plants, GM animals or GM microorganisms.
- Food, ingredients or additives, which are produced from GMOs.

For labeling, it is irrelevant if the GMOs used are detectable in the end product.

At present, possible products include those made from GM soy beans and GM maize:

- oil from GM soy beans;
- margarine from GM soy bean oil;
- oil from GM rapeseed/canola;
- cornflakes from GM corn;
- starch from GM corn;
- bread with GM soy protein or GM soy flour;
- glucose (dextrose), glucose syrup and other ingredients with GM corn starch.
- peanut puff snacks oder tacos containing GM corn starch.

Additives which are produced from GM plants also must be labeled, and include:

- sugar from GM sugar beet;
 - lecithin from GM soy beans;
 - vitamin E (tocopherol) from GM soy beans; and
 - cellulose from GM cotton, used as thickening agents and binder.
- Food, ingredients and additives, which contain genetically modified organisms. This category applies primarily to food produced with GM microorganisms, and includes:
 - wheat beer with GM yeast;
 - yeast extract from GM yeast;
 - yoghurt with GM lactobacilli (lactic acid bacteria);
 - salami (raw sausages) with GM lactobacilli (lactic acid bacteria);
 - blue cheese with GM moulds;
 - Quorn (protein from protazoa) from GM fungi.

Under certain conditions, numerous products are exempt from labelling obligations. These exemptions primarily concern additives and processing aids, but also apply to meat, milk and eggs. Not all applications of genetic engineering oblige the manufacturers to label the ingredients on the end product.

Food, which is produced with the aid of GMOs does not have to be labeled. Legislatively, no labeling is required when food ingredients or additives have not resulted directly from a GMO. In this context, labeling is dependent upon food or additives containing material made from genetically modified organisms.

Labeling is not required for i.e.:

- meat, eggs, milk, and dairy products obtained from animals fed with genetically modified feed.
- additives, flavours and vitamins produced with the help of GM microorganisms, which are defined as “processing aids” if foods or additives do not contain any of these microorganisms or their residues
- food containing GMOs up to a threshold of 0.9 percent:
 - if the producer or importer of a product is in a position to supply evidence that appropriate steps have been taken to avoid the presence of such material and that the presence is adventitious or technically unavoidable,
 - if the GMOs are authorised in the European Union and therewith classified to be safe.
- honey containing pollen or nectar from genetically modified plants,
- products containing the GMOs substances that must not be declared on the list of ingredients, such as enzymes, substrates for microorganisms, at present, most often GM corn or GM soy, carrier substances, which may be derived from starch, dextrans or glucose, which all may consist of GMO raw materials (GMO Labelling. Guidelines 2007).

Soybean food products produced with GMO ingredients

Each year, the EU imports approximately 40 million tones of raw soy products, primarily from Brazil, the United States and Argentina, which are the worlds’ leaders of GMO soy producers. Imported soy is predominately used to feed livestock (cattle, swine, and chicken) - without the soy protein Europe would not be able to maintain its current level of livestock productivity. During processing, soybeans are pressed in oil mills, and the derived oil is extracted and refined for food use. In addition, soybeans are used to produce numerous food ingredients and additives.

Many food manufacturers recognize soy protein as a versatile food ingredient with functional and nutritional properties that greatly enhance the value of finished foods in every consumer category. It can be present in:

- baked goods - soy protein is used in the manufacturing of breads, cookies, crackers and other baked goods to improve texture, hold moisture, create cake richness, whiten bread, extend shelf-life, reduce breakage and crumbling, enhance nutrition, improve manufacturing, handling and machine ability. It also improves mouth feel and overall quality as perceived by the consumer.

Table 1. Requirements for labeling of genetically modified products

Genetically modified product	An example of a food or food ingredient	Required labeling
Foodstuffs	Corn flour	YES
Products derived from GM plants	Corn oil Soybean oil Rapeseed oil	YES
Food derived from animals fed with GM feed	Eggs Meat Milk	NO
Food produced with the participation of enzymes GM	Bakery products obtained with the participation of amylase	NO
Additives and flavorings produced from GMOs	Lecithin extracted from soybeans used in the production of chocolate	YES
GM feed	Corn	YES
Feed produced from GMOs	Corn gluten Soybean feed	YES
Additives to feed derived from GMOs	Vitamin B 12 (riboflavin))	YES

Source: own elaboration based on GMO labelling. Guidelines, 2007

- breakfast cereals - soy protein is used extensively as an ingredient in hot cereal mixes and breakfast bars to boost protein value and quantity.
- pasta – fortification with soy protein increases nutritional value, especially protein content.
- beverages and toppings - soy isolates are used in coffee whiteners, liquid whipped toppings and pre-whipped toppings. They also are used in sour cream dressings to emulsify fat, control viscosity and provide textural characteristics. Instant beverages used as meal replacements often contain soy concentrates and soy isolates as a source of protein.
- meat, poultry and fish products - processed and whole meat products can be improved by adding soy protein, which provides the product flexibility and cost stability consumers demand. Adding soy protein to meat and poultry products can enhance moisture holding, texture, binding and cohesion, product yield, juiciness, protein quality, appetizing color and appearance, longer shelf-life, palatability and total nutrition.

- dairy products – a number of dairy analog products have been developed with soy protein, including imitation milk, imitation cheese, non-dairy frozen desserts, coffee whiteners, yogurt and others. Soy protein lowers cost, improves nutrition and reduces allergenic response.
- milk blends - many companies produce soy and milk protein blends for food manufacturing, combining the two to offer protein content similar to milk in a non-fat dry milk form. The different blends are used as a complete or partial replacement for non-fat dry milk in baked goods, sauces, meat products and other foods (Gelder, Kammeraat & Kroes 2008).

In the context of the labeling of foods potentially containing GMOs or produced based on genetic engineering in Table 1 there are presented examples of regulations on the labeling of foodstuffs and animal feed based on the European Commission Regulations 1829/2003 and 1831/2003.

Materials and method

The aim of the research carried out in March and April 2014, was verification of the presence on the Polish market food products, the packaging of which have contained the information about the genetic modification of the product or its ingredients. Inspected food came from retail shops and supermarkets located in Krakow. The analysis covered the packages of: baked goods, breakfast cereals, pasta, beverages and toppings, meat, poultry and fish products as well as dairy products. Altogether 75 batches of food products were analysed across 8 business units, such as: Alma, Carrefour, Tesco, Biedronka, Lidl, Polo Market, World's Kitchen and Organic Health Farm. The method used in this research was uncategorised observation, although information was collected in a deliberate and planned way. The complementation of the market research was the survey conducted in order to determine the general knowledge of the inhabitants of Malopolska Region on GM foods and their attitude towards products labeled with such an information. Special attention was paid to soy products potentially containing GM soy in their composition. 150 adult respondents (95 women and 55 men) living in Malopolska Region took part in a survey conducted by telematic. It consisted of 22 questions, including 3 open and 19 closed ones.

Conducted surveys, due to the relatively small number of respondents and the way to reach them (electronic survey) can not be regarded as representative, but they may be an important contribution to the knowledge of Poles, especially the younger market participants, on food products containing GMOs. Therefore, they are not merely cognitive, but may also have the meaning of application.

Results and discussion

Polish consumers rarely have the possibility to find labels/information indicating the use of genetic engineering on food products' packages. Only 4 out of 75 checked in this scope products packages had a label 'contains GMO'. It was soy cutlets, chocolate, soybean oil and salad vegetable oil.

Other controlled parties of the foodstuffs were not labeled as a food that contains any element of genetic modifications. Similar results obtained the study conducted by IJHARS - none of the analyzed samples potentially including GM soybean contained such an indication. Additionally laboratory tests performed on these products have confirmed that the modification content of the samples did not exceed 0.9% of the genetic modification, and thus the threshold at which food is to be marked (IJHARS raportuje o GMO 2009). In the present research it can be assumed that the reason for the lack of information on the GMO content on the other foodstuffs' labels was similar.

To get to know the Malopolska inhabitants' attitude to GM food and the level of their interest of such products' labeling, respondents were asked whether GM foods in their view is currently present on the market. According to 42% of questioned people it is, 7% were not able to clearly answer this question, and 51% of respondents claimed that it is not. When asked about their subjective attitude to this food category the majority explained that they are afraid of transgenic foodstuffs, 22% is free from these concerns, and 33% had no opinion on this point.

To the question of whether respondents personally encountered with products containing GMOs 25% of them answered affirmatively, 59% negatively and 16% said that it is difficult to say. The most often difficulties to answer this question had people over 50 years old.

When making a purchase decision of food 84% of the respondents never check product labels for the presence of information about genetic modification, 11% sometimes do that, and only 5% often and very often.

Although the respondents claimed to have seen products labeled with GMO on the market they have a considerable difficulty in identifying specific examples. The most often mentioned were the products which are analogues of meat – soy burgers (34%), soy hot dogs (30%), as well as soy milk (30%), cornflakes (23), animal feed (20%) and products containing modified starches (18%). The last indication generally demonstrates a lack of reliable knowledge about GMO foods and erroneous classification of products containing modified starch to this group. None of the respondents mentioned bread, chocolate, oil or pasta as a product suspected of the presence of GMOs.

The factor that affects the ability to identify on the foodstuffs' market the food products containing GMOs is the level of education of the respondents. In general, people with higher education were more interested in GMO foods

and more aware of its presence on the market, although not always their attitude to these products was generally positive. The results confirm studies carried out on a representative group of Poles in 2012 by CBOS (Badora 2013).

Representatives of the Malopolska inhabitants asked about the legal regulations concerning the labeling of food products containing GMOs or produced by genetic engineering were convinced that in Poland it is compulsory to label food products with an appropriate information. The vast majority of respondents claimed that this applies to both plant products containing GM organisms (95%) and animal products from animals fed with feed containing GMOs (93%). These opinions dominate in all socio-demographic groups, but most people who can not take a position on this issue are among respondents over 50 years old and have at most primary education. It is also worth noting that respondents slightly greater emphasis put on the issue of the need to label products directly derived from genetically modified organisms, than products from animals fed with feed containing GMOs.

As is widely known one of the most important determinants of food choices on the Polish market is the price of products. Therefore, respondents were asked about their preferences in the choice between cheaper products containing GMOs, and significantly more expensive (min. 30%), but GMO free. It turned out that the vast majority of respondents - 75% of them when having the opportunity to make an informed choice prefers the more expensive products, but free from genetic modification treatments. 7% of respondents had difficulty in making a choice, while 18% admitted that their previous knowledge about GMOs allows them to make a choice based on lower prices of these products. The survey's results indicate a high distrust of Poles to the achievements of genetic engineering.

Respondents were asked about their attitude to food in the packaging of which there is an information about genetic modification of its ingredients. The vast majority of them - 72% treat it as a warning against genetic modification and knowingly waives the purchase of such a product. With this perception in mind it is not surprising that even if the manufacturers want to use the component containing GMOs do everything not to exceed the amount of it above 0.9% as it is the amount above which the product must be labeled as including GMOs. For the remainder of the surveyed information that the product contains GMOs is a specific assurance of a high quality of such a product. Its modified ingredients have to undergo multiple tests to be allowed on the market. Some respondents ignore this information, because their knowledge is insufficient to be able to consciously take a stand on this issue. Wisely prefer to choose a product labeled GMO-free, or without any information in this regard. Only a small proportion of respondents - 8 of 150 person indicated that in this case, you have to deal with the foodstuff in the

production of which a much greater amount of pesticides, herbicides and so on were used than in the case of plant products including GM soybean for instance.

Majority of the inhabitants of Malopolska Region are not aware that the application of genetic engineering is very common and often unavoidable in nowadays food production. What is more most of these cases are not covered by the labelling directive. Milk, meat, eggs, and other animal products from animals fed GM plants can be the best examples of it. Another example may be enzymes produced with the help of GM microorganisms used in the production of cheese, juices, wine or glucose syrup.

To sum up it has to be stated that even supermarkets with no products with GMOs labels may not be free from all types of genetic engineering.

Conclusions

To conclude the results of conducted analysis it has to be stated that producers avoid labelling of food products with GMO on the Polish market. Only 4 products out of 75 with the label indicating the use of genetic modification were found. The average consumer interprets such a label as a warning rather than as information about the application of genetic engineering. Many consumers prefer to choose products without GMO labels as they are meant to notify of health hazards. In order to avoid risks of losses in sales and damage to their image many producers have changed the composition of their products – trying to avoid GMO ingredients, or are spending extra money for the ones with written guarantee that GM content does not exceed 0.9%. This for example allows the producer to use soy and forgo the GM label. On the other hand GMO-free labels are more and more popular on the food products, however the rules of labelling products with them should be harmonized within the EU. We should keep in mind that genetic engineering is a very broad field, and even when organisations, producers, and retailers use the term "GMO-free", genetic engineering often is involved nonetheless. Therefore, even supermarkets with no products labelled as GMOs may not be free from all types of genetic engineering.

It is believed that consumers have the right to know not only which products contain GMO, but also which ones are GMO-free, however the mandatory labelling can be misleading. The EU should adopt harmonised rules for GMO-free labelling to ensure consistency in the EU market and that the threshold for GMO-free labelling should be harmonised at EU level at 0.1%. This percentage is a reasonable detection limit, significantly better than the threshold of 0.9% for GMO labeling, and representative of GMO-free soy beans available on the world market. This will ensure that soyfood producers who invest in traceability and certification of GMO-free soy beans and even

pay an extra premium to their suppliers, are able to label their products as GMO-free which is also relevant information for the consumer.

It is worth noting that mandatory labeling programs for GMOs have limitations as methods of communicating about product quality, in that the label is one-dimensional (presence or absence of GMOs). For consumers, the full informational content of a GMO label will depend on all the additional information, indicators, and cues they have regarding the quality attributes of products produced with or without biotechnology.

Consumers should be educated about GMO food products. Objective campaign is needed including the composition of the products they purchase and consume just to not blindly follow the most prominent slogans appearing in the media not always backed up by reliable scientific research.

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ANTIOXIDANT CAPACITY OF SELECTED COMMERCIAL SPICES

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Introduction

Spices are food ingredients that can be used in food products, not only because of its flavouring properties - taste, flavour, aroma and color. Also important are the antioxidant properties, which allow for replacement of synthetic antioxidants such as BHA, BHT commonly used in food technology. These antioxidants are volatile and easily decompose at high temperatures. Additionally, it is still unclear whether chronic consumption can lead to health risks (Martinez-Tome et al. 2001).

Furthermore spices, seasoning, blends or its extracts are widely used in commonly each meal, and they can constitute in the daily diet source of antioxidants.

The scavenging action of plant constituents has been found to be associated with phenolic compounds (Madsen et al. 1996).

It is well known that the antioxidant activity of plant extracts containing polyphenol components is due to their capacity to be donors of hydrogen atoms or electrons and to capture the free radicals. DPPH• analysis is one of the tests used to prove the ability of the components of the spices extracts to act as donors of hydrogen atoms (Stoilova et al. 2007).

Many factors can affect the content of polyphenols, the most important seem to be those associated with the botanical origin of spices components and antioxidant content. The research of literature show that the amount of identified polyphenols also depend on the type of extraction solvent (Su et al. 2007).

Different specific antioxidants accept basic ones, are present in spices e.g. piperine in black pepper, gingerol and zingerone in ginger, thymol and carvacrol in marjoram.

The research material consisted of the different dried spices from different parts of plant: black pepper (*Piper nigrum* L.), cinnamon

(*Cinnamomumverum*), ginger (*Zingiberofficinale* L. Roscoe), marjoram (*OriganumMajorana* L.).

Black pepper is the most popular and most widely used spice, cinnamon is the most popular spice used to sweet dishes, marjoram is the most popular, traditional domestic spice and ginger very good known oriental spice.

Table 1 shows the part of plant from which they comes and botanical family for all investigated samples.

Table 1. Characterization of the plant material

Part of plant	Spice	Botanical family
fruit	Black pepper (<i>Piper nigrum</i> L.)	<i>Piperaceae</i>
roots	Ginger (<i>Zingiberofficinale</i> L.)	<i>Zingiberaceae</i>
leaves	Marjoram (<i>OriganumMajorana</i> L.)	<i>Lamiaceae</i>
bark	Cinnamon(<i>Cinnamomumverum</i>)	<i>Lauraeceae</i>

Source: Drygas & Śniegocki 1971

The aim of this study was short commodity evaluation of selected spices, then determination the antioxidant activity of them, measured by their abilities to scavenge free radical using DPPH• reagent, and at least determination of total phenolic content measured using the Folin-Ciocalteu method.

Now more and more often, it is believed that the method of using the FC reagent is used to determine total antioxidant activity rather than the polyphenols content (Everette et al. 2010).

Commodity evaluation of selected spices

Black pepper(*Piper nigrum*L.)

All spices, particularly pepper, must be used with consummate skill. Even the most insipid dishes can be improved by taking advantage of the pungent taste and spicy aroma of pepper to produce savoury dishes; that is why pepper is a universal favourite, the most popular spice in the world. About 60% of *Piper* species occur in central and northern South America. Pepper is now grown in tropical zones such as the Asia Pacific region, mainly India, Indonesia, Malaysia, Sri Lanka, Thailand, China, Vietnam and Cambodia. Outside the Asia Pacific region the crop is distributed in Brazil, Mexico, Guatemala, etc.(Ravindram, Kalluparackal 2001).

Black pepper is obtained from the still immature fruit, colored green to yellowish. During drying, the surface shrinks and wrinkles, color changes to black. The main alkaloid present in the pepper, responsible for its pungent, burning taste, is piperine, it forms about 98% of the overall impression of the crisp pepper aroma(Melchior, Kastner 1976).

Parmarlisted the following flavonols from pepper: quercetin, isoquercetin, isorhamnetin 3- β -D-rutinoside, kaempferol 3-arabinoside, kaempferol-3-o- β -galactoside, quercetin-3-o- β -D rutinoside(Parmar et al. 1997).

Cinnamon(*Cinnamomumverum*)

The genus *Cinnamomum* belonging to the family *Lauraceae* comprises about 250 species which are distributed in India, China, Sri Lanka and Australia. Cinnamon (*Cinnamomumverum*) are called aromatic cinnamon bark, which have a specific odor and sweet-bitter taste. Cinnamon is the oldest spices in the world, after black pepper is one of the most frequently sold one. The sweet taste of cinnamon is due to the presence of cinnamaldehyde. It is reported that, when combined with sweet food, the sweet sensation of the food is enhanced because of the synergetic effect between the sweet taste of sugar and sweet aroma of cinnamon (Kumar et al. 1997).

On the domestic and foreign markets are two species: *Cinnamomumzeylanicum* Breyne (*Cinnamomumceylanicum* Ness) i*Cinnamomum cassia* Blume from *Lauraceae* family.

Ceylon Cinnamon outside Ceylon (Sri Lanka) is cultivated in Java, Sumatra, the Seychelles and Brazil, Guyana and the West Indies. Chinese cinnamon plantations outside of China are in Cambodia, Vietnam, Japan, Indonesia and Sumatra.

In trade cinnamon occurs most often in two types: *Cassia vera* – bark from thin branches and *Cassia lignea* – bark with the thickness of above 3 mm, obtained from trunk and thick branches. (Melchior, Kastner 1978, Newerli-Guz, Śmiechowska 2008) The components of cinnamon of particular importance in food processing are oil, coumarin and cinnamon aldehyde, which owes its distinctive cinnamon flavour (Lee, Balic 2005).

Ginger (*Zingiberofficinale* L.)

Ginger is a spice plant not issuing fruit, is reproduced by division of rhizomes. Spices are thickened, fleshy rhizomes, sometimes bent and branched. Some of them may gradually pass the root.

Native to Southern Asia, although extensively cultivated at tropics, principal India, Sri Lanka, Jamaica, Africa (Sierra Leone, Nigeria), China and Australia (Arthur 1996).

Raw ginger is a beige-gray rhizome with bark in pieces. Cross-section of the raw material is pale yellow, grainy, mealy, and fibrous due to the presence of tufts of bast fibers. The smell of ginger root is strong, aromatic, flavor strongly spicy. (Drygas Śniegocki 1971, Melchior, Kastner 1978)

The most valued is very aromatic and sweet ginger from Jamaica - the highest quality type, rhizome scraped from all sides, not treated with lime and not bleached (Melchior, Kastner 1978).

The characteristic organoleptic properties of ginger are due to steam volatile oil and non-volatile solvent extractable pungent components. Its roots and the obtained extracts contain polyphenol compounds (6-gingerol and its derivatives) which have a high antioxidant activity (Herrmann 1994).

Marjoram (*Origanum Majorana* L. syn. *Majorana hortensis*)

Marjoram is a herbaceous plant native in Europe and in the Mediterranean. Marjoram belongs to family *Lamiaceae*, formerly *Labiatae*. It has often been mistaken for oregano in botanical description, for many years both marjoram and oregano were known as *Origanum majorana* L., now marjoram is identified as *Majorana hortensis* as a member of the mint family. Distinguishing marjoram from oregano is possible during flowering mainly due to the characteristically different inflorescences (Senderski 2004).

Marjoram is widely known in the form of crushed, gray-green herbal spice *Herb Majoranae* with a characteristic smell. Commercially is most common marjoram rubbed, very rarely whole dried sprigs. Marjoram is characterized by light gray-green color, has a strong, aromatic smell and a bitter, spicy taste. Marjoram owes its popularity to this characteristic aroma, widely used in cookery (Newerli-Guz 2012).

Material and method

Chemicals

2,2-diphenyl-1-picrylhydrazyl (DPPH•), Folin-Ciocalteu reagent, Gallic acid were purchased from Sigma Aldrich GmbH (Steinheim, Germany), HPLC grade methanol was obtained from POCH S.A. (Gliwice, Poland).

Plant material and sample preparation

Dried spices - 48 samples (15 black pepper, 9 cinnamon, 8 ginger, 16 marjoram) were obtained from the Tricity market (18 different producers) and investigated. Five packaging of all investigated spices were taken from market according to PN-ISO 948 (PN-ISO 948 Spices sampling), then milled in case of black pepper and cinnamon according to PN-ISO 2825 (PN-ISO 2825 Herbs and spices, preparation of the ground sample for analysis) and then were prepared 1% aqueous extracts.

Total phenolic content

The total phenolic content in water crude extracts was determined by Folin – Ciocalteu method. Antioxidant activity was determined using the DPPH reagent and showed as DPPH radical scavenging percent.

The total phenolic contents (TP) were measured using the Folin-Ciocalteu (FC) method with modifications (Amin at all.2006)

2.5 mL of 0.2 N FC reagent were added to tested solutions and mixed. After 5 min, 2 mL 75g / L Na₂CO₃ solution were added. After 120 minutes incubation, the absorbance relative to that of a prepared blank was read at a 760 nm using a spectrophotometer (Unicam UV2, Varian). The total phenolic contents are expressed in mg of Gallic acid equivalents (mg GAE/g of product).

DPPH• free radical scavenging

Free radical scavenging effect was determined using the free radical DPPH•(2,2-diphenyl-1-picrylhydrazyl)reagent. 1 mL of the extract was added to 2mL DPPH•. The samples were gently mixed and left to stand in the darkness for 60 min. Absorbance was read at 517 nm using spectrophotometer. A control sample was prepared by mixing DPPH• with distilled water. The ability of extracts to scavenge DPPH free radicals was calculated according to the following equation:

$$\text{Radical scavenging activity}[\%] = (1 - (\text{Abs. of sample} / \text{Abs. DPPH}\bullet)) \times 100 \%$$

The values are presented as the means of triplicate analyses.

Statistical analysis of the results consistent of the calculation of basic measures such as: the average and standard deviation. One way analysis of variance (ANOVA) and the Kruskal –Wallis test were used to determine the difference among the means P values < 0.05 were regarded to be significant.

Results and discussion

Table 2 shows the results of total polyphenols content (TP) and antioxidant activity (AA) of the investigated extracts.

Table 2. The average total polyphenols content (TP) and antioxidant activity (AA) of investigated types of spices

Sample	n	TP [mgGAE /g]	AADPPH [%]
Black pepper(<i>Piper nigrum L.</i>)	15	13.45±5.20	82.96±2.13
Cinnamon (<i>Cinnamomumverum</i>)	9	77.98±13.17	81.14±15.52
Ginger (<i>Zingiberofficinalis</i>)	8	12.81±4.54	52.69±15.77
Marjoram (<i>OriganumMajorana L.</i>)	16	55.88±22.21	89.14±0.74

Source: own research

Total phenolic content

The results of the phenolic analyses show that in examined spice samples total phenolic content vary. Black pepper and ginger have the contents of a similar level of about 13 mg GAE/g, marjoram contained about 50 mg GAE/g, while the tested cinnamon samples contained the nearly fourteen times more than marjoram samples.

The determined total phenolic content of the black pepper extract amounted from 10.73mg GAE/g to 32.11mg GAE /g, with average total phenolic content for all tested samples 13.45 mg GAE/g.

Investigated by Gülçin (Gülçin 2005) water and ethanol extracts of black pepper exhibited strong total antioxidant activity. Total phenolic content were determined as Gallic acid equivalents on the level 54.3mg GAE/g in water extracts.

Confirmation of the antioxidant properties of black pepper is research in which was found about 200 times greater than BHT and over 300 than BHA black pepper's ability to inhibit lipid oxidation (Shobana, Naidu Akhilender 2000).

Black pepper compared with spices from other parts of the plant has phenolic content at a lower level than the cinnamon, ginger, nutmeg, star anise, bay leaf and basil (Su et al. 2007, Hinneburg et al. 2005).

The content of phenolic compounds in the studied cinnamon samples was in the range of 53.16 to 94.26 mg GAE / g, average content 77.98mg GAE / g of product.

Average total phenolic content in ginger samples was 12.81mg GAE/g, and the maximum was reached 17.92 and minimum 11.97 mg GAE/g. The lowest content of total phenolic was determined in raw ginger - 2.12 mg GAE/g.

The content of total phenolic in ginger have been previously evaluated by other authors, and they were diverse.

Hinneburg (Hinneburg at al.2006) determined 23.5 mg GAE/g in investigated ginger samples. Other analyzed spices from *Apiaceae* family like cumin, fennel, parsley have bigger amount of total phenolic.

Isabelle (Isabele, Bee Lan Lee et al. 2010) determined fresh ginger phenolic content at the level of 1.45 mg GAE / g.

Ginger has excellent antioxidant properties (Kikuzaki, Nakatani 1993), studies include the role of components such as gingerol in inhibiting linoleic acid autoxidation.

The total phenolic content range from 26.66 mg GAE/g to 98.43 mg GAE/g in analyzed marjoram samples, with average value 55.88 mg GAE/g.

Tsimidou (Tsimidou, Boskou1994) concluded that among the spices extensively studied, the plants obtained from the *Lamiaceae* (formerly *Labiatae* family) possess a significant antioxidant activity. Observed by other authors total polyphenol content in marjoram ranged from 21.18 - 84.26, average 47.92 mgGAE/g¹(Feck, Turk 2008).

Polyphenol content determined by Hinneburg (Hinneburg et al. 2006) for basil, also family *Lamiaceae*, by the Folin – Ciocalteu method has amount 147 mg GAE/g.

DPPH• scavenging activity of plant extracts

Determined in this study the antioxidant activity of water extracts from 4 spices vary. The extracts from marjoram have the highest antioxidant activity as DPPH• scavengers, although black pepper showed high antioxidant activity. The lowest antioxidant activity from determined samples has ginger, but also more than 50%.

The water extracts of black pepper have strong scavenging activity (about 80%), the highest ability to free radicals scavenge was 85,4%, the lowest 78%.

Other studies have shown the impact of γ -irradiation (the power of the radiation and the operating time) on the ability to scavenge free radicals. Suchajin his research showed a statistically significant effect of black pepper radiation on the growth of antioxidant activity using DPPH• after 2 months of storage. After 4 months of storage, these changes reached levels 4 to 9%(Suchaj et al. 2006)..

The ability of the extracts to scavenge DPPH• free radicals differs in investigated samples of cinnamon. It was on the level from 36,33% to 90,81%, on average level of 81,14%.

Prasad (Prasad et al.2009) found the difference in DPPH• scavenging ability among different cinnamonspecies, they decrease as *C. zeylanica*>*C. cassia*> BHT>*C. pasiflorum*>*C. burmannii*>*C. tamala*.

All tested ginger samples were characterized by high DPPH • radicals scavenging, on average 52.69%, in the range of 34.8% to 79.2%. In comparison, fresh ginger has 40.88% antioxidant capacity.

This result is very similar to the result of other authors (Shobana,Naidu Akhilender 2000) who determined the antioxidant capacity of fresh ginger at 41%. But compared with the literature concerning other spices, this is not too high level (Drużyńska,Wojda 2007).

For ginger it seems advisable to use extraction media which are able to extract the lipophilic antioxidant compounds (Hinneburg et al. 2006]. Other studies (Murcia et al. 2004) seem to contradict this findings.

The marjoram extract showed a significant effect in inhibiting DPPH• radicals, reaching up to 90,7%, average value is 89,14%. Antioxidant activity of marjoram shown by Jin Jun (Jin Jun et al. 2001) was at a similar level and amounted to 85.5%.

Table 3 presented results of statistics ANOVA, ANOVA Kruskal-Wallis for studies samples. All results showed a statistically significant effect of the origin/producers on spices antioxidant activity.

Table 3. Impact of origin/producer on investigated parameters.

Spice	Parameter	Value of statistics (ANOVA, ANOVA Kruskal-Walis)	
Black pepper	TP	F(14,45)=77.57	p= 0.000
	DPPH	F(14,45)=620.98	p= 0.000
Cinnamon	TP	KW(8,27)= 11.82	p= 0.159
	DPPH	F(8,27)= 650.43	p= 0.000
Ginger	TP	F(7,24)= 116,69	p= 0.000
	DPPH	F(7,24)= 2118,14	p= 0.000
Marjoram	TP	KW(15,48)= 46,55	p= 0.000
	DPPH	KW(15,48)= 43,96	p= 0.001

TP –total phenolic content

DPPH- DPPH• scavenging activity

Source: own research

Conclusion

The results of the study imply that different spices are a potential promising source of antioxidant. Their origin/producer have impact on the total phenolic content and the ability to neutralize free DPPH• radicals.

The differences in the phenolic content and antioxidant activity result primarily from characteristics of the raw material. The influence of these parameters can also have processing and storage (especially considering a 24 month shelf-life of spices). Other authors confirm that factors determining the antioxidant activity of spices beyond genetic one are mainly environmental technique and growing conditions (Sgherri et al.2010). Cousins (Cousins et al. 2007) mentions that many inherent variation in commercial processes across the production region including variation in drying techniques, time in the field, harvesting methods, and clone can lead to quality differences in the final products.

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CONCEPTUAL PRINCIPLES OF PRODUCT DEVELOPMENT FOR ENTERAL NUTRITION

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Introduction

Patients' nutrition is one of the urgent and important problems of modern society. Analysis of current global and domestic scientific data [1, 2, 3, 4] shows that the problem of nutritional needs of people with hyper metabolism with necessary nutrients can't be solved only by a simple substitution of protein, lipid, carbohydrate and vitamin and mineral costs. Numerous studies in this area [3,10] make it possible to reveal the specifics of the physiological processes of the organism in this state and became the basis for developing the basic principles of human nutrition with hyper metabolism . These provisions are based on advanced data features of the metabolic response of the organism to the action of the etiological factors and caused by it stress and their physiological effects, biological constitution (genotype) of the victim, energy and plastic nutrients value, due to which there is provision of food needs. Scientists like E. Fistal, H. Kozinets, A. Belyaev, M. Marshak, I. Khoroshilov, O. Pochepen, A. Botkin, Wilkinson A.W., Selye H., Wilmore D., Mayers I., Barendgret K., Moore F.D., S. Ortega, M.H. DeLegge, D. Royall, K. FarverA have done significant contribution to the development of scientific principles of human nutrition with specific diseases. Their numerous long-term studies have allowed to analyze the specific metabolic processes in the human body with hyper metabolism and became the basis for the development of the concept of food in this state.

The aim is to conduct a critical analysis of existing approaches to patients' diet and the development of conceptual foundations to develop products for enteral nutrition taking into account the specific needs of the body of people during the period of treatment and rehabilitation.

Results and discussion

For the metabolic response of the organism to certain factors (such as after surgery, trauma, burns, disorders of the gastro- intestinal and nervous systems, septic conditions, etc.) characteristic is the development of non-specific reactions hyper metabolism – hyper catabolism , resulting in a complex disordered metabolism of proteins , carbohydrates, lipids, enhanced cost of carbohydrate lipid reserves and the collapse of tissue proteins , loss of body weight. Nutrient needs of patients who are in critical condition are especially high. To replenish essential nutrients of the patient two types of food are used on practice - enteral and parenteral [6, 7].

Selecting patients for enteral nutrition is based on a study of anthropometric data to determine nutritional status, anticipated duration of illness and severity of damage. Patients with weight loss of 10% or more are defined as depleted patients requiring enteral nutrition [9].

Choice of the type of food - enteral or parenteral is a contentious issue at present. Muggia-Sullam and colleagues in their studies have shown that patients with different surgical pathology both parenteral and enteral nutrition are maintaining an equivalent positive nitrogen balance, maintaining body weight, providing protein synthesis [2]. Some other studies have shown that parenteral nutrition is a significant advantage in securing calorage , positive nitrogen balance, maintaining weight. However, even modern total parenteral nutrition is not replacing many very important functions of the intestine, and in its conduct mechanical, septic, metabolic complications occur [10]. There are indications that parenteral nutrition can inhibit its own immune system. Intestine atrophies during total parenteral nutrition. In addition, the parenteral nutrition requires strict adherence to sterility and speed the of ingredients introduction due to some technical difficulties.

Enteral nutrition, like any other method of intensive therapy, should be used based on a number of indications and contraindications [7]. The main indications for enteral nutrition include: state, accompanied by hyper metabolism (trauma, burns, sepsis, major surgery, preoperative depletion), central nervous system disorders that result in disturbances of nutritional status, prolonged dysfunction of individual organs (cachexia, closed head injury, surgery, kidney failure, liver failure). Intestinal obstruction, acute pancreatitis and severe malabsorption are contraindications to the use of enteral nutrition.

The works by Fry D.E. have shown that enteral nutrition can prevent immune system abnormalities, reduce the incidence of septic complications by restoring the integrity of the affected intestine wall [3, 7]. Moreover, many clinical studies have shown that enteral nutrition is more physiological, effective and safe. In addition, this type of food does not require sterility.

Enteral nutrition helps maintain digestive tract in normal physiological condition and prevent many complications that occur with patients in critical condition. Thus, whenever possible, the choice of method of nutritional support should in favor of enteral nutrition.

Cost of food components is about the same in group of enteral and parenteral nutrition, but the cost of auxiliary components is significantly higher in medication for parenteral nutrition (cost of non-food components of parenteral nutrition is 43 % of the total cost of the medication, while for preparations for enteral nutrition value nonfood additives is only 13 %) [10]. Additionally, the frequency of complications occurring during enteral nutrition is significantly lower than during parenteral, so when you consider the cost of treating complications that arise when a particular type of food is consumed, it can be concluded that the value of enteral nutrition is lower than parenteral [7]. These studies show that during enteral nutrition it is not always possible to achieve calorage and nitrogen balance as during parenteral, however, immunity and nourishing effect of enteral nutrition is significantly higher than parenteral [3, 4].

It should be noted that the treatment of complications arising during early enteral nutrition requires much lower economic costs than treatment of complications arising during parenteral nutrition. Thus, enteral nutrition allows on the one hand to effectively ensure a positive nitrogen balance, restore lost cell mass of patients, restore cellular immunity, improve wound healing by providing the patient's body with the necessary plastic substrates. On the other hand, the cost for enteral nutrition is considerably lower than the equivalent parenteral energy supply.

Nutritional support products development is based on current scientific research on nutrition composition balance and its conformity to the specifics of metabolic needs of people with hyper metabolism. Enteral nutrition can be used both as a food additive and used as a source of nutrients and energy for a long time as part of such compounds include proteins, fats, carbohydrates, vitamins and minerals in balanced proportions. Modern mixes for enteral nutrition are mainly of two types: balanced (polymer), and modified. Most mixtures are isotonic and contain no lactose.

Polymeric formulas contain whole protein isolate (milk, egg whites, meat, soy protein, etc.), partially hydrolyzed starch and triglycerides, as well as the necessary set of vitamins, micro and macro elements. Before absorption, these mixtures are subjected to enzymatic processing in the gut. Often mixture is produced in powder form and prepared immediately before use, but there is ready suspension. Energy density of polymer compositions is usually 1 to 2 kcal \ ml and osmolarity is not usually high - 300-450 mosm\kg. Balanced mixtures contain enough calories and are relatively cheap. In the practice of intensive care modified mixtures have proven being significantly better which

contain crystalline amino acids or oligopeptides and medium link triglycerides [2]. Simple molecules do not require digestion, which is especially important for patients in critical condition because their fermentation processes are violated [10]. Under conditions of stress occurs mobilization of branched amino acids from skeletal muscle [6, 7, 10]. In modern balanced formula branched amino acids content is not less than 30% of the total protein content, and nutrient mixtures used for the critically injured branched amino acids content is not less than 45 %.

With the introduction of modified mixtures containing branched amino acids the severity of catabolic reactions is reduced, nitrogen balance is reconstructed and mortality among survivors is reduced. The presence of these compounds can not only ensure the patient's body with nutrients and energy, but also for the enrichment of special pharma nutrients and compliance with certain proportions of nutrients exercise aimed at correcting disorders caused by certain diseases. Protein availability in mixture is usually defined as the ratio of 1 gram of nitrogen per 150 non-protein calories [2]. Due to the high demand of the critically injured in nitrogen, some modified mixtures ratio of nitrogen to non-protein calories is 1:80, 1:100. The identified deficiencies of modified mixtures include hyper osmolarity and relatively high cost.

When developing products to provide the necessary needs of the patient, it is important to use the available raw materials taking into account specific stages of treatment and recovery of the body. Thus, due to the need for additional energy to break down protein more convenient to the patient are proteins and animal fats in concentrated and isolated forms easily accessible to digest peptides and amino acids. Increasing the amount of protein in a balanced mixture can be achieved by introducing protein supplements into the mixture. Thus, the present mixtures for enteral nutrition contain glutamine [2]. This amino acid is a major component of food of intestinal villi, is involved in the regulation of reabsorption of water and sodium, prevents the development of stress ulcers, normalizes the secretion of immunoglobulin. Glutamine is an essential amino acid during serious illness because its use by tissues exceeds several times the body's ability to endogenous biosynthesis of glutamine. Glutamine dosage recommended is 0.57 mg / kg / day or 2.6 g per day [8]. Amino acid arginine has equally important role in the diet of patients, which increases the production of nitric oxide synthesis of peptides, proteins, creatine phosphate, and also participates in the synthesis of nucleotides [1]. Arginine also increases collagen synthesis, thereby accelerating wound healing, helps to normalize the metabolism of nitrogen.

Fat is a major source of non-protein calories and allow you to create a reserve of approximately 140,000 calories per adult. The structure of the fat must include essential fatty acids such as linolenic in an amount of 2-4% of total calories as fatty acid deficiency this causes a decrease in platelet

aggregation [7]. In general, the amount of fat in the mixture for enteral nutrition must not exceed 50%. One of the highlights is the ratio in mixture for enteral nutrition of long-chain and middle-chain fatty acids. Equally important is the presence in mixtures for enteral nutrition components such as omega -3 unsaturated fatty acids, the main action is directed at the inhibition of excessive activation loop of arachidonic acid, resulting in inhibited formation of prostaglandin E2 immunodepressant. The use of omega -3 fatty acids also leads to accumulation of antioxidants, particularly vitamin E [7].

To provide support for the body's carbohydrate is important to use sugars in the diet of patients having no adverse effect on the pancreas and not injure the gastrointestinal tract. 30 to 70% of the energy needs of the organism are provided by carbohydrates from 2 to 5 grams per kilogram per day [4]. Often, carbohydrates are glucose, fructose and lactose sometimes. Compositions for enteral nutrition intended for use for patients with diabetes carbohydrates contain fructose and starch [5,6]. You must also take into account the fact that the respiratory rate of the oxidation of carbohydrates is the highest, which is important with patients with respiratory failure.

Ascorbic acid plays particularly important role in patient's body, its metabolism is strongly correlated with the protein, and vitamins B₁, B₂, B₆, B₉, E, trace elements - iron, zinc, selenium, dietary fiber and so on. In view of the fact that the metabolism of proteins, carbohydrates, fats necessarily requires vitamins catalysts of biochemical reactions and trace elements-cofactors of fixed enzymes. All modern mixes for enteral nutrition contain the above mentioned substances in sufficient quantities [3]. It is well known fact that the lack of one component leads to a lack of effectiveness of the enteral nutrition.

Achievement over the last two decades is development of a formula for enteral nutrition that takes into account the metabolic characteristics of the organism, resulting in lack of function of specific organ systems. In particular in case of hyper metabolism mixes for enteral nutrition are used ensuring 1-1.5 g / kg per day of protein and calorie content equal to 25-30 kcal / kg / day with a low ratio of nitrogen to non-protein calories 1:100 [2]. Such mixtures for enteral nutrition contain a large number (40%) branched amino acids.

However, even using the most advanced balanced and modified mixtures for enteral nutrition, especially in the early stages, occur some difficulties. Therefore, we carry out a systematic way to develop a series of foods that would take into account the specific needs of the organism of patients throughout the period of treatment and rehabilitation, namely the early stage of treatment (immediately before and after surgery), intensive rehabilitation period, the main period of rehabilitation of people recovering, remote recovery phase outside medical institutions (for patients with injuries requiring nutritional support for a long time).

Conclusion

So, one of the conditions for successful treatment and rehabilitation of people with hyper metabolism is nutrition developed on the basis of scientific data on the specific occurrence of physiological processes. This makes it necessary to find new ways to provide the patient with full and comprehensive nutrients needed, which in turn requires research and scientific reasoning for the development of products specific to the flow of metabolic processes at different stages of treatment and rehabilitation that would help restore the body's efficiency of people as a result of diseases and injuries of different origin.

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EXTENSION OF STORAGE DURATION OF SOME KINDS OF BREADS THROUGH RADURATION

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Introduction

One of the major indicators of bakery foods competitiveness in conditions of market economy is their shelf life without deterioration of the basic parameters of quality.

To improve the quality of baked bread, various additives – improvers of flour quality, have been used recently everywhere, however the question of justified application of improvers of bread quality remains open (Nechaev 2006), especially that their application frequently does not solve the problem of storage length. The great success is achieved when applying new technologies for dough preparation (Erkinbaeva, Kozyukina & Sherbakova 2002; Nilova & Naumenko 2007). To increase the shelf life, various methods of physical exposure, such as both heat treatment and ionizing rays (infra-red, ultra-violet, etc.) treatment are mainly used (Snapyan, Sahradyan et al. 1996; Polandova & Kventniy 2002).

We previously showed that bread irradiation with gamma rays led to protection of bread from mold thus increasing duration of its storages (Snapyan, Sahradyan et al. 1996; Sahradyan & Voskanyan 2011).

The objective of the present work is to develop a method for treatment of packaged bread - a product that is most subjected to microbiological spoilage (moldiness) due to the high moisture content. We aimed to develop the method, which will provide chemical and microbiological safety, guaranteed storage for long terms.

Material and methods

The study involved samples of formed bread of extra, first and second grade wheat flour. For bread packing plastic film was chosen owing to its high

physicochemical properties and resistance to radiation doses used by us ($D\gamma = 0-10.0$ kGy) that are within the limits of recommended international norms Codex Stan 1883 REV - 2005 (Codex General standard... 2005). Treatment of the investigated product samples was performed in the Institute of Physical Researches NAS RA on ^{60}CO isotope installation K-120 000 with energy 1.25 MeV and power 1.0 Gy/sec. The energy of the Institute was also within the recommended limits of the international norms of CAC RCP 19-1979. The researches were performed by standard organoleptic, physicochemical and microbiological methods of analysis. First signs of molding and potato disease were defined visually and by seeding technique (GOST 26972-86... 1986).

Experimental section

Samples of formed bread of extra, first and second grades wheat flour packed in plastic film of 15 μc thickness were treated on ^{60}CO isotope installation with gamma rays at $D\gamma = 0.5-5.0$ kGy, control samples of bread were not irradiated ($D\gamma = 0$ kGy). After treatment, the whole of bread (control and treated samples) was stored under room uncontrolled temperature and humidity conditions (20-25°C and relative humidity 60-75%) for identification of its shelf life. Frequency and the research program were conducted according to MU 4.2.727-2002 (MU 4.2.727-99 ... 1999).

Before storage and at the end of exposition, in addition to organoleptic, physicochemical and microbiological researches of bread, amino-acid composition was also determined.

Results and discussion

Study of physicochemical parameters of investigated samples of bread before treatment with gamma rays (control samples) and immediately after treatment showed their compliance with the established standard (AST31-94-32, 1994). Results of the research are given in Table 1.

As follows from Tab. 1, treatment with gamma rays did not affect physicochemical parameters of bread. Change of organoleptic parameters of bread occurred during its storage in parallel with staleness. Upon storage the treated samples of bread lost their freshness at different rates.

Freshness of bread was assessed by the accepted 5-point system. Data of Table 2 prove that parameters of bread freshness were higher than those of control samples. Control samples ($D\gamma = 0.0$ kGy) lost freshness in 48 hrs while samples treated with gamma rays ($D\gamma = 3.0-5.0$ kGy) lost freshness in 96-120 hrs. . It is necessary to note that high doses of irradiation, e.g. $D\gamma = 10$ and $D\gamma = 20$ kGy led to the appearance of extraneous smell and taste: sour at $D\gamma = 10$ kGy and fishy at $D\gamma = 20$ kGy that sharply worsened organoleptic parameters of bread.

Table 1. Physicochemical parameters of formed bread of different quality wheat flour

Quality	Dose of irradiation kGy	Porosity, not less than %		Humidity, %		Acidity, %	
		Norm AST31-94	Result	Norm AST31-94	Result	Norm AST31-94	Result
Second	Bread	63.0		40.0-48.0		3.0-5.0	
	0.0 (control)		63.0		44.2		2.8
	1.0		63.0		44.2		2.8
	2.0		65.0		44.2		2.8
	3.0		63.0		44.2		2.8
	6.0		63.0		44.2		2.8
	10.0		63.0		44.0		2.8
First	Bread	65.0		40.0-47.0		3.0-4.0	
	0.0 / control/		65.0		42.3		2.3
	1.0		65.0		42.3		2.3
	2.0		65.0		42.3		2.3
	3.0		65.0		42.3		2.3
	6.0		65.0		42.3		2.3
	10.0		65.0		42.5		2.9
Extra	Bread	68.0		39.0-46.0		3.0-4.0	
	0.0 / control/		68.0		43.8		2.8
	1.0		68.0		43.8		2.8
	2.0		68.0		43.8		2.8
	3.0		68.0		43.8		2.8
	6.0		68.0		43.8		2.8
	10.0		68.0		44.3		2.9

Source: own research

Table 2. Change of freshness of bread of extra quality wheat flour at storage

Dose of irradiation kGy / Storage time, hrs	Freshness score							
	3	24	48	72	96	120	144	168
0	5.0	4.0	3.0	2.5	2.0	1.0	1.0/0/	1.0/0
1.0	5.0	4.5	4.5	4.2	4.0	1.0	2.0	1.0
3.0	5.0	4.5	4.5	4.5	4.2	4.0	2.5	1.0
5.0	5.0	4.0	4.5	4.2	4.0	4.0	2.5	1.0
10.0	3.0	3.0	3.0	3.0	1.0	3.0	2.0	1.0
20.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	1.0

Source: own research

Determination of physicochemical parameters of the control and treated samples of bread at storage revealed that porosity was the main parameter, subject to a change. Table 3 presents data on the dynamics of porosity change in formed bread of extra quality wheat flour.

Table 3. The effect of storage time of the bread of extra quality wheat flour treated with gamma rays on its porosity

Dose of irradiation kGy	Point, %							
	3	24	48	72	96	120	144	168
Storage Time, hrs								
0	68.0	64.0	62.0	61.0	56.0	55.0	53.0	52.0
1.5	68.0	65.0	62.0	64.0	64.0	62.0	61.5	61.5
5.0	68.0	66.0	63.5	64.0	63.0	62.0	62.5	62.5
10.0	68.0	66.0	65.0	65.5	62.0	61.5	61.0	60.5
15.0	67.0	65.0	65.0	65.0	61.0	60.0	60.0	59.5

Source: own research

In the course of the research changes in microbiological parameters of bread quality were studied in dynamics according to [4]. Samples of bread were visually checked for the appearance of the first signs of molding and potato disease. Visible growth of mold fungi colonies was noted in control samples as early as on the 4th day, while some treated samples remained outwardly clean even after 4 months.

In the treated samples the rate of moldiness appearance depended sharply on a dose of irradiation. The dependence of molding rate on the dose of irradiation was ascertained; thus, at $D\gamma = 5.0$ kGy mold fungi visually appeared on the 9th day after treatment; at $D\gamma = 1.0$ kGy on – on the 15th day; at $D\gamma = 2.0$ kGy – on the 25th day; at $D\gamma = 3.0$ kGy on the 30th; at $D\gamma = 5.0$ kGy on the 35th; at $D\gamma = 10.0$ kGy on the 100th; at $D\gamma = 20$ kGy on the 120th day after treatment. In the course of storage no potato disease was observed in both treated and untreated sampled.

Besides visual observation seeding technique was used to determine the total amount of yeasts and mold fungi in bread samples according to GOST 26972-86 [4]. The results of analysis showed that the samples treated with $D\gamma = 1.0$ and $D\gamma = 2.0$ kGy about 30 times slowed down mold growth as compared to the control, increasing shelf life of the packed bread up to 90-100 days, correspondingly.

To establish the availability or absence of radical changes in gamma rays treated samples of bread, amino-acid composition and the amount of nitrogenous compounds in the studied samples of bread before radurization ($D\gamma = 0.0$ kGy), after it ($D\gamma = 1.0 - 5.0$ kGy) and in the course of storage (on the 1st, 8th and 30th days) were determined. The total nitrogen was determined by the Kjeldahl arbitration method (for protein conversion $K = 5.68$).

It was found that protein substances of bread when treated with $D\gamma = 1.0 - 3.0$ kGy practically did not undergo changes, and when treated with $D\gamma = 10.0$ kGy the loss of proteinaceous substances was significant and about 1.6 % in relation to the first day.

Study of amino-acid composition (analysis was carried out on aminoacid analyzer "T-339") showed that when irradiated with $D\gamma = 0.0 - 3.0$ kGy nor quantitative, nor qualitative amino-acid composition of the bread samples of the extra quality wheat flour changed. Fig. 1 shows amino-acid composition of the investigated bread samples.

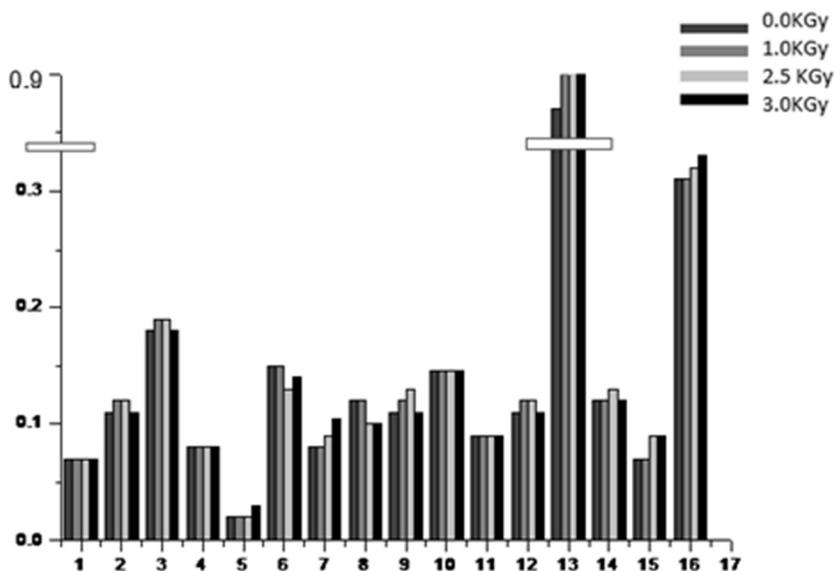


Figure 1. The effect of gamma irradiation ($D\gamma = 0-3.0$ kGy) on amino-acid composition of bread samples of extra quality wheat flour
 1 - 11- threonine, 2 - valine, 3 - leucine, 4 - lysine, 5 - methionine, 6 - phenylalanine, 7 - isoleucine, 8 - alanine, 9 - arginine, 10 - aspartic acid, 11 - histidine, 12 - glycine, 13 - glutamic acid, 14 - serine, 15 - tyrosine, 16 – proline

Source: own research

As follows from Fig.1, samples of bread of extra quality wheat flour contained nonessential and essential amino acids: alanine, leucine, lysine, phenyl alanine, isoleucine, arginine, aspartic acid, glycine, glutamic acid, serine, tyrosine, proline. Tryptophane out of essential and cysteine out of nonessential amino acids were missing in the composition of these samples. As seen from Fig. 1, radurization of bread did not lead to significant changes in the amino-acid composition of bread. Exception was glutamic acid the content of which in comparison with the control samples at $D\gamma = 10.0$ kGy decreased.

Conclusions

The given experimental data prove that gamma rays irradiation at the dose of $D\gamma = 3.0 - 5.0$ kGy is optimum for treatment of bread of different grades wheat flour and 9-10 times increases the storage time as compared to untreated samples not worsening organoleptic and physicochemical parameters, nor leading to the change of nutritious properties.

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THE EFFECT OF INFORMATION ABOUT LOWERING SALT CONTENT ON FLAVOR ACCEPTANCE OF SELECTED MEAT PRODUCTS

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Introduction

Sodium is a mineral, essential in small amounts for the proper functioning of the body. It determines the maintenance of homeostasis of the body, and particularly the proper water-electrolyte balance. Salt is the main source of sodium in the diet. It plays an important role in nutrition, by giving products a proper flavor, often decisive of their attractiveness and consumer acceptability (Jarosz *et al.* 2011, Słowik 2009, Tsugane 2005).

There is scientific evidence that increased salt intake contributes to elevated blood pressure. This in turn can lead to increased risk of cardiovascular diseases. According to the World Health Organization (WHO), the salt content in the diet should not exceed 5 g per day, which is equivalent to 2 g of sodium per day. However, the data on global consumption of salt indicate its excessive consumption, which may cause adverse health effects to consumers. Therefore, the WHO recommended that countries, in which salt intake exceeds the recommended daily dose, should develop and implement programs persuading the society to reduce the salt intake in the diet. The WHO also recommended introduction of separate, original programs by individual countries due to cultural differences and variations in patterns of consumption. Social campaigns conducted in countries such as Ireland, Finland and France, and encouraging manufacturers to reduce the salt content in products manufactured by them, contributed to the significant achievements, as measured by the declarations of consumers, changing habits associated with the consumption of salt and the actual decrease in the incidence of coronary heart disease and stroke (WHO 2011, Jarosz *et al.* 2011, Respondek, Grodowska 2011, Staniewska, Staniewski 2010, Traczyk, Jarosz 2011).

In Poland, the excessive intake of salt is an enormous problem, and the average daily dose, similarly as in many other European countries, exceeds 12

g per day. The reasons for this situation include the excessive use of salt in the industrial production of food products, such as canned goods, meats, cheeses, pickled food, smoked bacon, soups and sauces, and some spices. As part of a comprehensive approach to the problem, it is necessary to take actions aimed at reducing salt consumption in the households, food serving establishments and catering companies. Manufacturers are aware of the problem and modify the composition of their products by lowering the salt content and search for simple and understandable ways of passing this message to consumers. Labeling of packaging with precise nutritional information may play an important role. However, the above-mentioned actions of producers must be accompanied by measures raising nutritional awareness of consumer. Only conscious consumer, who has an appropriate knowledge and knows how to use it, can become a demanding client capable of making mature and rational purchasing decisions, bearing no threat to his health and life (Goryńska-Goldmann, Ratajczak 2010, UE 2012).

Numerous scientific studies showed that consumers, when buying food products, are increasingly driven by information on the content of essential nutrients, such as fat or sugar (Grimes *et al.* 2009, Kristal, Patterson 2001, Neuhouser, Kristal, Patterson 1999, Weaver, Finke 2003). Hence, it seems interesting to answer the question whether a reduction in the salt content and the placement of such information on the product's packaging will significantly affect consumer acceptance, expressed by perceiving the salty taste and overall flavor desirability. This is particularly important in view of intensified work on the shaping of appropriate food choices.

Therefore, the aim of this study was to assess the impact of information of a reduced salt content on the acceptance of the flavor in the selected assortment of meat products.

Materials and Methods

Materials and experimental design

The material consisted of sausages (frankfurters) of varying salt content from a single manufacturer, available in retail on domestic market. The study involved 100 students of the University of Warmia and Mazury in Olsztyn, 79% of whom were women and 21% men. These respondents declared that they are consumers of meat products, including sausages.

The study of consumer acceptance proceeded in two stages, in the same conditions, in the sensory laboratory at the Department of Commodity Science and Food Analysis, UWM in Olsztyn. First, consumers evaluated coded samples of sausages of standard and reduced salt content, and then the same group of consumers assessed decoded samples, with information label.

Methods

The even method. The even method was used to examine differences in the preferences with respect to sausages of standard and reduced salt content. Evaluators were asked to indicate the preferred sample. Evaluation was performed using coded samples, and then the same group of consumers assessed decoded samples, with information label.

Scaling method. To assess the desirability of salty taste of sausages with standard and reduced salt content, a classic nine-point hedonic scale was applied with the following marginal designations: 1 – "the product is not salty enough", 5 – "the product is sufficiently salty", 9 - "the product is too salty "

The evaluation of expected product palatability, based on a visual and information layer of the packaging, was performed using a 9-point scale with designations: 1 - "the product does not taste good at all", 5 - "the product taste is just fine", "9 - the product taste is above average."

Indirect questionnaire. The consumer survey analyzed respondents' opinions as to the choice of the product on the basis of a visual and information layer of its packaging, with particular regard to the information about the salt content and its role in the selection of sausages.

Preparation and presentation of test samples. Directly prior to the evaluation, sausages were taken out of commercial packaging and placed in the amount of about 25 g to the odorless, disposable containers. Samples for evaluation were coded with the three-digit codes and presented to evaluators in a random order. Non-carbonated water was served between samples to neutralize the taste.

Statistical analysis

The data collected were processed using a Statistica 9.0 software package. Basic statistics were used in the initial analysis of the data, i.e., means and standard deviations. One-way ANOVA was applied to assess the differences in the mean values. Normality of distribution was tested by Shapiro-Wilk and Levene's tests, and the homogeneity of the variable by Levene's test. The significance of differences was tested at the level of 0.05.

Results and Discussion

Consumer preferences relative to the sausages of a standard and reduced salt content

The results of the test of consumer preferences with respect to the sausages evaluated with the use of coded samples showed that most

consumers preferred the product with a standard salt content (68 selections), while the product with a reduced salt content received more than half of the votes less (32) (Fig. 1).

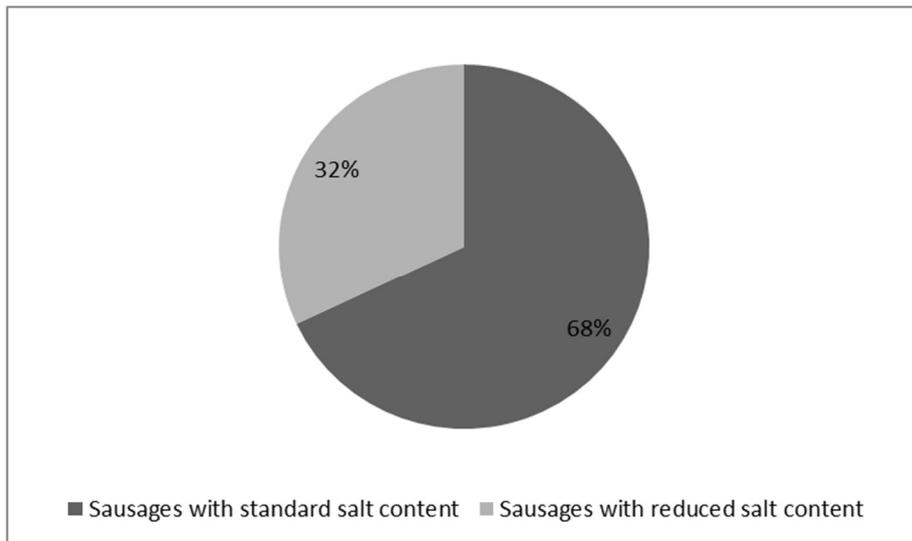


Figure 1. Consumer preferences for sausages with standard and reduced salt content -encoded samples (% of respondents)

Source: own research.

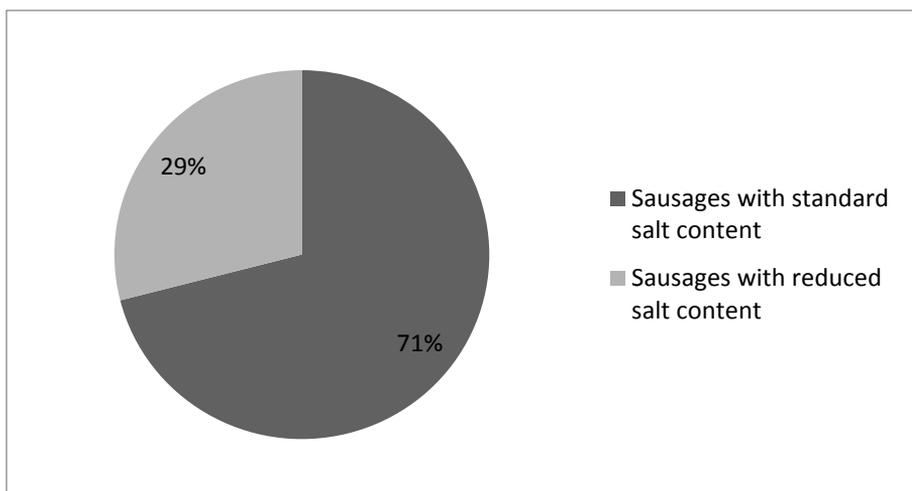


Figure 2. Consumer preferences for sausages with standard and reduced salt content- decoded samples (% of responses)

Source: own research.

The test using the products with the information about standard or reduced salt content demonstrated similar relationships. Most consumers preferred the product with a standard salt content (71 selections), while the product with a reduced salt content was selected by 1 out of 3 respondents (Fig. 2). This result demonstrated that the appearance of information about the lowering of the salt content in the tested products had a negligible effect on the change in consumer preferences towards the products evaluated.



Figure 3. Preference for standard and reduced salt content sausages, by gender of the respondents - encoded samples (% of responses)

Source: own research.

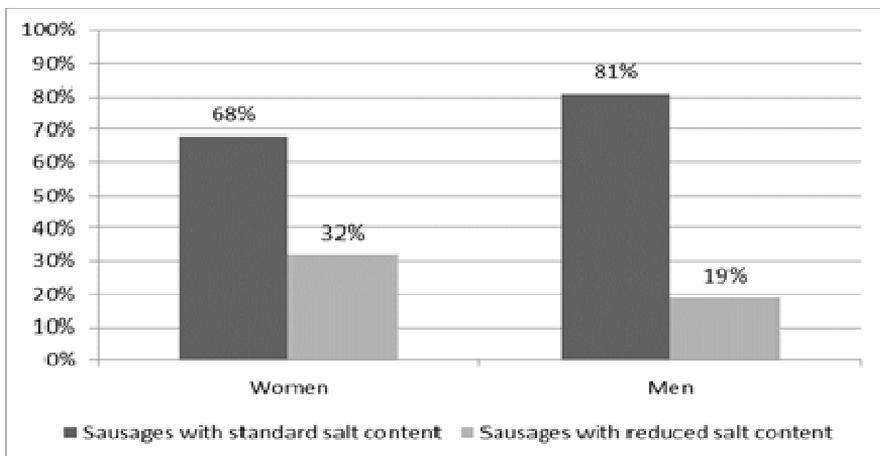


Figure 4. Preference for standard and reduced salt content sausages, by gender of the respondents - decoded samples (% of responses)

Source: own research.

It should be noted that in the experiment with coded samples, men (38%) were a group that more often than women (30%), preferred sausages with reduced salt content. In contrast, the occurrence of information about lower salt content caused a change in preferences of men who were inclined toward the product with standard salt content (81% of responses in the group) (Fig. 3, 4).

The results might indicate a negative consumer perception of the process of decreasing the salt content in the analyzed meat products. The addition of sodium chloride in the production of meat is not only technologically justified, but provides general consumer acceptance. Technological barriers and limitations related to the safety of manufactured products as well as the need to meet individual consumer preferences, will not in this case serve as factors that would facilitate the process (which is slow anyway) of reducing salt intake in the population of Europeans, including Polish society (Czerwińska 2011).

In addition, the study found that women were less willing to change their preferences when information about reducing the salt content in the product was added. Similar results were obtained in the study conducted by Liem et al. (2011). The study was based on determining the expected and actual saltiness of "Chicken Noodle" soups and their palatability. These authors found that information about the lower salt content in the product negatively affected its perception. Consumers were adding salt to the soup that was provided with information about the lower salt content. A different situation occurred in the case of soups with a logo demonstrating the health benefits of the product, but without information on the reduction of salt in the product. Therefore, the need is emphasized, to consider a more evocative and visually exposed forms of communication of important nutritional information.

Information about the standard and lower salt content in sausages and consumer desirability of salty taste

Analyzing consumer impressions in relation to salty taste, it was noted that the study participants evaluated sausages with a standard salt content as "salty just right" both in the test with coded (5.42 ± 1.46) and decoded samples (5.69 ± 1.08) (Tab. 1). The presence of the information about the salt content significantly differentiated the desirability of salt taste in the test group, in the case of sausages with a reduced salt content. The coded samples were rated significantly higher than the non-coded samples (Tab. 1).

In the group of men the desirability of salty taste changed significantly, to the detriment of the product with the information about lower salt content (Tab. 2).

Table 1. The degree of salty taste desirability of tasted sausages

	Standard salt content sausages	Reduced salt content sausages
Encoded samples	5,42±1,46	4,61±1,62 ^a
Decoded samples	5,69±1,08	4,04±1,71 ^b

Evaluation was performed with using a 9-point scale, where 1 meant that the product is not salty enough, 5 - the product is salty enough, 9 - product too salty
a, b - statistically significant differences within a column for $p \leq 0.05$
Source: own research.

Table 2. The degree of salty taste desirability of tasted sausages, by gender of respondents

	Female		Male	
	Standard salt content sausages	Reduced salt content sausages	Standard salt content sausages	Reduced salt content sausages
Encoded samples	5,44±1,41	4,52±1,69	5,33±1,68	4,95±1,28 ^a
Decoded samples	5,64±1,05	4,16±1,74	5,85±1,19	3,57±1,53 ^b

Evaluation was performed with using a 9-point scale, where 1 meant that the product is not salty enough, 5 - the product is salty enough, 9 - product too salty
a, b - statistically significant differences within a column for $p \leq 0.05$
Source: own research.

In the group of men the desirability of salty taste changed significantly, to the detriment of the product with the information about lower salt content (Tab. 2).

Determination of the expected palatability and selection factors of sausages with standard and reduced salt content

Another element of the study was to determine the expected palatability of the product evaluated on the basis of visual and information layer of the packaging. Subjects evaluated higher the palatability of the product with standard salt content (Tab. 3). This relationship was also confirmed by the analysis taking into account gender of the consumers (Tab. 4).

Table 3. The expected tastiness of tested sausages determined on the basis of visual - information layers of package

	Average	Standard Deviation
Standard salt content sausages	5,48	1,12
Reduced salt content sausages	4,27	1,61

Evaluation was performed using a 9-point hedonic scale, where 1 meant that the product does not taste at all, 5 - is very tasty enough, 9 - product tastes good above average
Source: own research.

Table 4. The expected tastiness of tested sausages determined on the basis of visual - information layers of package, including the gender of respondents

	Female	Men
	Average / Standard Deviation	
Standard salt content sausages	5,43±1,07	5,62±1,36
Reduced salt content sausages	4,18±1,38	4,38±2,27

Evaluation was performed using a 9-point hedonic scale, where 1 meant that the product does not taste at all, 5 - is very tasty enough, 9 - product tastes good above average

Source: own research.

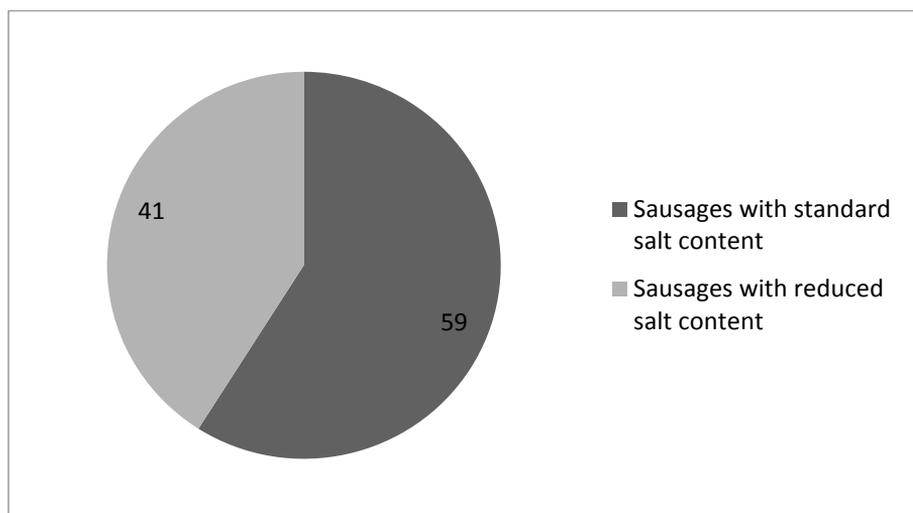


Figure 5. Choice declarations of the product on the basis of visual - information layer of package (% of responses)

Source: own research.

Analysis of consumers opinion concerning the choice of the product on the basis of visual and information layer of its packaging allowed to conclude that about 60% of the respondents would choose sausages with standard salt content (Fig. 5).

The results pertaining to the declaration of product selection based on the assessment of visual and information layers of the packaging showed that men (67%) more frequently than women (57%) declared the choice of sausages with standard salt content (Fig. 6).



Figure 6. Choice declarations of product on the basis of visual - informational layer of package, including the gender of respondents (% of responses)

Source: own research.

The evaluators were asked in the next stage of the study to list the factors, which motivated them to select a particular version of the flavor of the product. The appearance of packaging, product design, product knowledge and familiarity with the taste of the product was most frequently mentioned regarding the sausages with standard salt content. Paying attention to these factors can be simplified to the familiarity with the product, its characteristic taste, appearance and visual layer of packaging. In turn, those who declared the selection of sausages with reduced salt content, most frequently specified the information about the reduced content of this component, next to the information on the lower fat content. Other, less frequently occurring answers in this group of consumers were the appearance of the packaging and information that this is a new product on the market.

As shown by numerous studies, the selection of the food is influenced by many factors. Krasnowska and Salejda (2011) emphasized that Polish food consumers during shopping were primarily driven by price. The study of Radzyńska and Jakubowska (2010) observed that the most important factors in the selection of meat products were associated with the product itself, with the organoleptic attributes prevailing. However, the study conducted by Mieczkowska and Panfil-Kuncewicz (2011) found that the choice of food was mainly affected by the information about the expiry date.

Our study concerning the effect of information on the acceptance of the taste in a selected group of meat products has shown that the information, in

broad terms communicated with the package, did not play such a significant role in the selection of sausages with standard salt content that are available on the market for many years. However, nearly 89% of respondents declared that their attention was drawn to the information placed on the package about the reduction of the salt content in sausages. This percentage was higher among those declaring selection of products with reduced salt content (over 92%) compared with the declarations of a group of people tending to choose products with standard content of this component (86%).

It can be concluded that the average consumer, despite an increasingly higher level of awareness and nutritional education, still does not show much interest in products with reduced salt content. However, any action affecting consumer choices that will be based on a dependable knowledge of nutrition will be a socially desirable phenomenon.

Conclusions

1. The appearance of the information on the salt content of evaluated flavor versions of products, affected minimally the changes in women preferences, whereas among men it resulted in leaning towards the product with standard salt content.
2. The feeling of salt taste desirability was differentiated by the information of the reduced salt content of the product - coded samples were rated significantly higher than the decoded ones. The desirability of the salt taste in the group of men changed considerably to the detriment of the product provided with the information of the lower salt content.
3. Nearly 90% of respondents declared that their attention was drawn to the information placed on the package about the reduction of the salt content in sausages. In the group of respondents declaring selection of products with reduced salt content, information about the lower content of this component in addition to the information about the reduced fat content were the most frequently mentioned factors in the hierarchy of their choices regarding the evaluated group of meat products.

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FACTORS DETERMINING THE QUALITY LOSS OF APPLES IN INTERNATIONAL TRANSPORT

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Introduction

Nowadays, we meet more and more often with infections related to food product quality. According to the control report of the Quality Inspection of Agricultural and Food Products in 2011, in the first quarter of that year the quality of fresh fruits and vegetables was controlled in 1939 batches (total weight of 203,7 tons) including 1219 batches of fresh fruits (total weight of 152, 2 tons) and 720 batches of fresh vegetables (total weight of 51,5 tons) (IJHARS Report 2011). The quality requirements in terms of their trade value were not met by 18% of all controlled products, with 216 batches of fruits and 126 batches of vegetables. Some of the irregularities were the result of improper labelling; however, as many as 7 tons of food products (ca. 3% of controlled batches) did not meet the quality requirements. Some of the products were not fresh and had visible signs of rot and decay. The results of that inspection show the importance of proper storage and transport of food products, especially fruits and vegetables. The quality inspection was conducted in large supermarkets only, and that was the first inspection of that kind in 2011. The report results were, however, much more positive compared with the previous ones. It is therefore clear that the consumers' and tradesmen's awareness regarding compliance with the conditions and standards applicable in the sales of food products is going up.

The quality in highly competitive reality of our times is oftentimes the key factor influencing consumer's purchase decision just like the level of transport services and sales is an important aspect in terms of quality. The majority of product damages occurs on the level of food transport and distribution, and that affects the final sensory impressions of the product on offer.

Since Poland joined the European Union, all professional carriers have had to comply with strict regulations regarding food product transport and,

consequently, had to heavily invest in proper equipment and infrastructure. As the result, the freight prices went up. Moreover, the prices of fuel rose dramatically. In the analysed offer the changes in fuel prices showed a rapid increase – which was presented in the graph below. The oil prices as of November 30th 2011 were on the level of 4.41 PLN. At the beginning of 2009 they were on the level of 3.53 PLN, which means that within less than two years, the prices went up by 0.88 PLN.

The aim of this study is to analyse the real data and find solutions that might facilitate the decisions in terms of logistics. The study is mostly explorative and focuses on evaluating the effectiveness of currently used food product transport methods. The main goal of this research is to analyse the level of unsold products and their cost due to wrong decisions in terms of the use of transport methods. Such mistakes do not only affect the company finance (the cost of returned or re-called products) but also the company's image.

The study was conducted in October-November 2011. The documents of the forwarding and transport company “Trans” were monitored and analysed with the focus on data – the company documentation was not allowed to be presented.

Elements of cost in food products road transport

The overall costs are the monetary value of raw materials cost, labour cost, external cost and manufacturing cost. The cost of transport means something different to the entity commissioning the transport services and something completely different to the forwarding company, rail or airline. The standard sales of transport services, excluding logistics companies, means that the real cost is incurred during the actual transport from one place to another and loading, reloading and unloading the goods. Such costs are characterized by their variability. For the company commissioning the transport services other costs might be of more importance, such as: the costs of storage, reloading, sorting, packaging, labelling and price tagging, product return or recall, product damage in transport and administration. It is therefore crucial to group different costs in order to be able to analyse them thoroughly (Bolton 2000, Button, Vega 2005, Dyszy 2008, DG TREN 2008, Follett 2007, Glaeser, Kohlhase 2003, Handbook on estimation of external costs in the transport sector 2008, Harris 2005, Philipp 2007). According to Jerzy Tarkowski, the following tripartite system should be used (Tarkowski 1995):

- Cost type – mostly costs depending on time such as: amortization, interest rate, taxes, insurance policies, labour cost; but also costs not directly depending on time: mileage, the use of tires, repair costs and services.
- Place of incurring the cost – understood as the type of vehicles used.

- Cost carrier – determining transport tasks, single transporting services or long-term, regular deals etc.

The calculation of the vehicle cost is the basis of the future cost relations. Fixed costs should be analysed here such as taxes and insurance, as well as variable costs such as mileage, fuel consumption and driver's effectiveness. The aim of such calculation is to evaluate the level of possible costs in the future. Total costs are divided into several elements which facilitates the assessment of the level, dynamics and diversity of costs. Additionally, a close monitoring of cost parameters enables the person responsible for planning transport and logistics a quick identification of potential threats and reaction or introduction of preventive measures.

The elements which have a direct impact on the level of transport costs are: technology, weight and volume of goods, transport distance, dangers connected with the transport and the value of products themselves. The total costs can be presented in the form of the following equation (Twaróg 2003):

$$K_t = K_{as} + K_{pts} + K_{mts} + K_b + K_u + K_{rs} + K_d + K_{ints} + K_{ut}$$

Where: K_{as} – amortization costs of cars and car base buildings
 K_{pts} – labour costs of drivers and operating personnel
 K_{mts} – materials, fuel and energy costs related to car exploitation
 K_b – office costs
 K_u – car insurance costs
 K_{rs} – car repair and maintenance costs
 K_d – lease costs
 K_{ints} – other car base costs
 K_{ut} – external costs of transport services

Currently, the most common breakdown of costs is a system based on the type of cost, which enables to group different costs in terms of their economic elements, expenditures and the means of production. In other words different costs of the company can be put together in a generic system of costs (Handbook on estimation of external costs in the transport sector 2008). The most standard elements are: amortization, use of materials, taxes, external services, labour costs and other costs. The exploitation costs of cars used in transport are usually connected with amortization, use of fuel, oil and lubrication and drivers' remuneration.

The prices of transport services can vary depending on the type of transport and the type of services as well as any additional services on offer required by the client. That is why the prices are usually different in terms of the means of transport used. The price itself is of course a very powerful tool in generating the demand for transport services. The following factors are considered while determining the prices of different transport services (Taryfy transportowe, ceny w transporcie 2011):

- the cost formula, eliminating the impact of the market on the price
- the size of single batch
- the scope of additional services
- the time of transport
- the space of transport

In international transport the following factors have to be considered (Ruciński 2005):

- the number of other companies offering similar services
- the level of internal costs
- the changes in energy costs: oil, fuel, electricity
- the rarity of services
- the level and diversity of demand for transport services
- the time of actual transport
- the level of taxes

A very common practice on the market of transport services is the use of tariff prices, which are usually determined by relevant public authorities. In such cases the prices of services cease to be decision-based.

Determining the total costs of transport services in tarpaulin-covered dumper trucks based on the data obtained from a transport company

The abovementioned transport company, depending on the time of the year, imports up to hundreds of tons of fruits and vegetables from Germany to Poland. For the purpose of this case study, the transport of apples called LOBO will be analysed. The apples are transported from Brandenburg, Germany to the distribution centre in Wrocław, directly to Cash and Carry shops – the route is 487 km long.

The driver is able to cover such distance as many as three times a week, considering the time of labour and all applicable road and traffic regulations. It is economically groundless to send the vehicle to Germany without any load, because an empty lorry generates losses. That is why the specialist in logistics working for the “Trans” company had to find some clients, who export their goods in the same direction. Usually, the products of other key accounts are then transported to Germany, to areas near Berlin and Nuremberg which ensures the continuity of providing transport services and of profit. When there are no clients interested in the company services in a given period, the forwarder looks for potential clients or transport goods on the commodity exchange market. Only if there is an exporter involved in the transport, the

journey is profitable. This study, however, focuses on the import of apples only. The total weight including the load must not exceed 40.8 tons. The vehicle itself weighs about 15 tons, consequently the total weight of load must not exceed 25.8 tons. Generally, the load is estimated at 24 tons.

Both loading and unloading of apples is performed at the presence of the driver and usually takes around thirty minutes. During that time fruit and vegetables are put onto the vehicle, and during the unloading phase the apples or potatoes are then segregated and packed in larger packaging and only then are ready for further distribution. The costs of transport en route Poland-Germany have gradually increased in the past few years as a consequence of the global rise in fuel prices. At the beginning of 2009, 1 km usually cost about 0.85 €, at the end of the analysed period the prices were around 1 €, and in areas near Berlin - nearly 1.20 €. The table below shows usual rates per kilometre.

Table 1. Rates per kilometre from Poland to Germany, based on the company data (“Trans”)

Rates per kilometre according to „Trans” company in EURO		
Months	Year 2009	Year 2010
<i>January</i>	0,85	0,96
<i>February</i>	0,84	0,92
<i>March</i>	0,85	0,90
<i>April</i>	0,87	0,95
<i>May</i>	0,87	0,95
<i>June</i>	0,90	0,98
<i>July</i>	0,93	1,00
<i>August</i>	0,90	1,00
<i>September</i>	0,90	0,98
<i>October</i>	0,90	0,98
<i>November</i>	0,95	1,00
<i>December</i>	0,95	1,10

Source: Own work based on the company’s internal documentation.

The research was conducted in November 2011 and refers to the period: 2009-2010. The transport of apples was analysed. All data received from the “Trans” company are connected with the costs of transport and the amount of transported goods. Data regarding product losses come from the distribution centre in Wrocław and had been obtained courtesy to the Director of Logistics and then developed and analysed for the purpose of this project. The apples transported from Germany are all local crops and are intended for direct consumption. Data presenting the amount of transported products and the level of losses are shown in table 2 and 3 together with transport costs. In order

to determine the overall costs of transport the route both ways was calculated, without considering any other services provided to other clients on the same route.

Table 2. The costs of transport en route: Brandenburg - Wrocław including losses and total tonnage

Transport of apples Brandenburg – Wrocław 487 km								
Year	2009				2010			
Months	Total amount [t]	Losses [t]	Transport costs [EUR]	Share of losses [%]	Total amount [t]	Losses [t]	Transport costs [EUR]	Share of losses [%]
<i>January</i>	288	14,4	9 934,80	5,00	268	11,9	6 545,28	4,44
<i>February</i>	240	11,7	8 181,60	4,88	240	12,1	8 960,80	5,04
<i>March</i>	240	9,8	8 279,00	4,08	192	9,7	8 766,00	5,05
<i>April</i>	192	6,5	6 779,04	3,39	168	6,6	6 477,10	3,93
<i>May</i>	192	3,8	6 779,04	1,98	144	2,8	5 551,80	1,95
<i>June</i>	168	2,9	6 136,20	1,73	144	2,2	5 727,12	1,53
<i>July</i>	144	3,7	5 434,92	2,57	120	1,6	4 870,00	1,33
<i>August</i>	168	4,3	7 012,80	2,56	144	1,9	5 844,00	1,32
<i>September</i>	288	4,4	10 519,20	1,53	168	2,4	6 681,64	1,43
<i>October</i>	312	3,6	11 295,80	1,15	264	2,3	10 499,72	0,88
<i>November</i>	360	3,4	13 879,50	0,94	288	3,1	11 688,00	1,08
<i>December</i>	312	4,2	12 028,90	1,35	264	3,7	11 785,40	1,40
Total	2 904	72,7	106 260,8	2,51	2 400	60,3	93 396,86	2,51

Source: Own work based on the company's internal documentation.

The greatest level of losses was sustained towards the end of January and the beginning of February 2009. As much as 5% of the total amount of transported goods was lost, which was most probably the result of particularly low temperatures in that period. Some apples were simply exposed to low, freezing temperatures for such a long period that the changes in their quality were irreversible. This situation repeated itself in 2010, this time at the end of February and at the beginning of March. The best period for transporting apples in tarpaulin-covered lorries/trolleys is the summertime; in 2009 it was May and June and in 2010 May-August. A particularly low level of product losses was the result of the temperatures outside similar to the ones in which apples ripen. The only sustained losses in those periods were the ones connected with the ending of the acceptable storage period. A considerable level of losses was also observed shortly after the harvest. The fruit were very fresh then and susceptible to damages, but the total losses were very low.

It can be observed that losses incurred only in transport are already quite significant. This most certainly is caused by the type of vehicle used. If the transport was performed in vehicles with temperature-controlled conditions and the loading and unloading was conducted in a different way, the level of losses would be much lower.

In conclusion, the biggest losses are sustained during the transport at the beginning of the year (in winter) with the total level of losses around 5%. The losses are the least significant in autumn (1.25%), the average annual product loss in transport is 2.51%

Determining the cost of product losses in transport

The costs arising from transportation of food products are not only connected with the transport costs, but also the cost of side effects such as commodity losses. As it was presented in table 2, the total losses of that kind are on the level of 2.5% which is quite a significant figure in terms of the total tonnage of transported goods. These costs and losses can be minimized though if proper logistic decisions are made. Considering the losses incurred in the transport of apples – 2.51% on average between 2009 and 2010 it gives the monetary loss of circa 5 011.41 € in just two years.

The losses in goods during transport processes

The losses in food products relate to the decrease in the weight of products ready for consumption within the supply chain, which of course leads to rejection of products that do not meet strict requirements and thus are not ready for direct sale and consumption. The starch test was used to assess the level of apple ripeness. While considering the safety of transported goods, the planned action is aimed at minimizing the level of product losses during transportation. Although the majority of forwarders assesses their own safety in terms of potential risks of theft, the safety in transport cannot be limited to the actual theft or robbery. During the process of transportation an important role is played by drivers. It is essential for them to understand and possess knowledge of the goods that are transported thus being fully responsible for the products in transportation. It is the driver's role to supervise the proper handling of goods during loading and reloading, proper documentation and handing over the products to authorized persons. It is also his role to make sure the products are transported properly and the load will not be critically damaged. The safety of the transported goods is the responsibility of the driver; however, the total liability regarding the load is on the carrier. In order to raise awareness among the drivers it is important to train them regarding the entire logistics of transport, electronic devices and systems that should be used, and of course the general safety of transported products and how to handle them properly. It is clear that one of the most crucial factors of proper handling are the conditions of transport, because they affect the food products directly leading to an increase in damages and losses. The reasons for incurring losses and the events leading to them shall now be analysed.

The reasons for incurring losses

There are several reasons for incurring food losses. According to international research results almost one third of all food products is wasted, which brings around 1.3 billion of tons every year (Global Food Losses and Food Waste, FAO 2011).

The losses can be incurred at different levels of supply chain and depend on many factors, from improper harvesting techniques through storage in bad conditions to transport and, of course to a large degree, the wrong choice of the means of transport. In considerably well-developed and developed countries a lot of food is wasted on the level of consumption, which means that a lot of products are thrown out despite being fit for consumption. In developing countries a lot of losses are incurred at the beginning of the supply chain due to lack of means of ensuring proper storage and cooling of food products. Different foods can also be rejected or wasted due to quality standards prevailing in a given country. If then the product is defective on the level of aesthetics, it might not be considered on offer. After harvesting, the apples are stored in temperature-controlled cold rooms. If then the products are removed from such storage conditions and placed inside the tarpaulin-covered vehicles, the storage cycle is broken and it can lead to irreversible changes in the quality of apples. In the summer periods, when the temperature outside is similar to the ones at which apples ripe, the transport in such conditions may lead to apple withering. It is also relatively common that the apples are loaded inappropriately by being dropped from above onto the vehicle thus losing their aesthetic value and in many cases being already at this stage unfit for sale. Unfortunately, it is impossible to evaluate the amount of products discarded, after they were taken by the shops, due to their unattractive appearance and consumer rejection. Summing up, it can be stated that the quality of fresh food products is strongly affected by the conditions in which they are stored and transported. Proper conditions of such product storage were examined and described in great detail by the National Institute of Fresh Produce. Apples should be transported in temperatures ranging from **-1°C to +3°C, which of course means that the use of tarpaulin-covered lorries/trolleys in the summer period diminishes the value of these food products.**

Proposals to minimize the amount of losses arising during transportation

The reasons for the losses arising during transportation, as proved in the research, depend to a large extent on the means of transport used and the conditions of storage. If the requirements behind transport conditions of

apples are fully understood, the choice of means of transport should be reconsidered. The products stored in temperature-controlled cold rooms should be also transported in similar conditions, especially if the temperature outside is significantly different from the conditions in which they were stored e.g. hot summer or particularly cold winter. In order to ensure the high quality of fresh fruits and vegetables it has to be remembered that the conditions of storage have a direct effect on the quality of these products and any kind of unnecessary reloading and re-packaging may lead to the decrease in the final level of quality of the products. This could be avoided if the apples were put into the containers which could be then returned when the driver is back in Germany. All unnecessary re-packaging is then minimized and the products retain their high aesthetic value.

Another important issue is the temperature and humidity during transportation. A certain level of temperature and humidity should remain stable, all sudden changes affect the quality of products directly. It is therefore much better to store the food products for a couple of hours in temperatures relatively higher than they should be, provided that there are no drastic changes in these temperatures. Sudden and brisk changes in the temperature of storage adversely affect the quality of fresh food. Both means of transport: tarpaulin-covered vehicles and lorries with cold rooms were then analysed and compared in order to determine why the vehicles with cold rooms are more suitable for the transport of fresh fruit.

The comparison of the incurred losses and costs between the transport in tarpaulin-covered dumper truck and refrigerator truck

The table below presents the losses arising from the transport in vehicles with cold rooms, exemplified by the transport of apples. The losses were estimated by the Director of Logistics at the Logistics Centre in Wrocław and then analysed for the purpose of this study.

Comparing the tables 2 and 3, it can be observed that the costs of transport in a vehicle with a cold room are perhaps higher (up to 4 thousand euro) but the losses generated in both cases make the second solution much more effective economically. Apples transported in vehicles with a cold room are also put into special boxes which prevents the fruit from sustaining damages. Figure 1 presents the full comparison between these two means of transport and losses incurred in both cases.

**Table 3. The costs of transport in the refrigerator truck en route:
Brandenburg - Wrocław including losses and total tonnage**

The costs of transport in a vehicle with cold room en route: Brandenburg – Wrocław 487 km								
Year	2009			2010				
Months	Total amount [t]	Losses [t]	Transport costs [EUR]	Share of losses [%]	Total amount [t]	Losses [t]	Transport costs [EUR]	Share of losses [%]
January	154	0.03	10 934.80	0.02	264	0.03	7 545.28	0.01
February	242	0.04	9 181.60	0.02	242	0.04	9 960.80	0.02
March	242	0.07	8 279.00	0.03	198	0.07	8 766.00	0.04
April	198	0.05	6 779.04	0.03	176	0.06	6 977.10	0.03
May	198	0.05	6 779.04	0.03	154	0.04	6 551.80	0.03
June	176	0.06	7 136.20	0.04	154	0.03	5 727.12	0.02
July	154	0.04	5 934.92	0.03	110	0.03	4 870.00	0.03
August	176	0.04	7 312.80	0.02	154	0.04	5 944.00	0.03
September	286	0.07	10 519.20	0.02	176	0.08	6 881.64	0.05
October	318	0.08	11 295.80	0.03	264	0.07	10 999.72	0.03
November	362	0.06	13 879.50	0.01	286	0.06	11 688.00	0.02
December	318	0.04	12 228.90	0.01	264	0.04	11 985.40	0.00
Total	2 824	0.63	110 260.80	0.02	2 442	0.59	97 396.86	0.02

Source: Own work based on the company's internal documentation

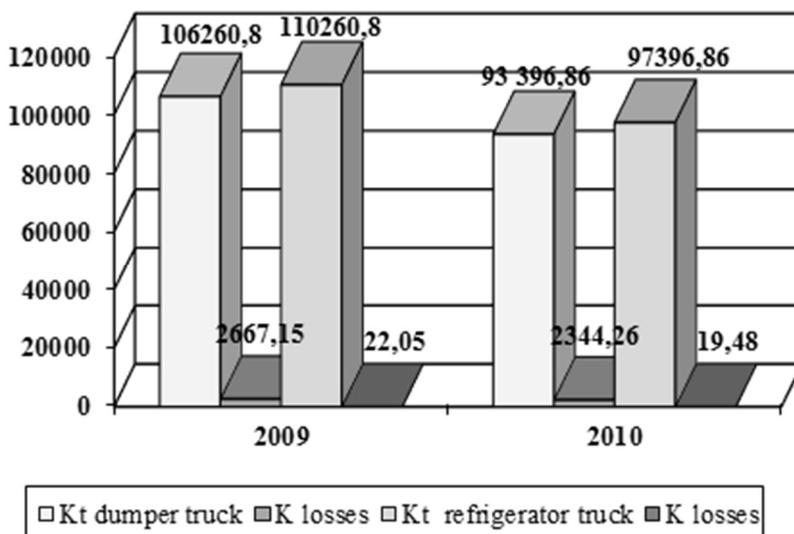


Figure 1. The list of costs and losses in the transport of apples (tarpaulin-covered dumper truck and refrigerator truck) in 2009-2010

Source: Own work based on the company's internal documentation

It can be observed that the costs of losses in these two cases are significantly different. It is altogether cheaper to transport apples in vehicles with a cold room, despite the fact that the cost of freight is higher than in case of the tarpaulin-covered vehicle. By not generating significant product losses the forwarding company can also build up the relationships with key accounts, gaining more respectability, trust and reliability. Key clients nowadays expect their forwarders to possess thorough knowledge about the transport conditions of food products whose temperature must be controlled. The most common reason for losses in transport (vehicles with a cold room) are again the human errors. If the temperature-control system breaks down, there is usually little that a driver can do about it, however when the driver makes a mistake – it is a completely different case. The most common human error here is setting the temperature of isotherm to automatic cooling, not to the continuous one. They also rarely check whether the temperature inside the cold room complies with the requirements, that is why the driver does not give any feedback en route about possible breakdowns of the system. Moreover, sometimes the drivers want to save fuel and energy by setting the temperature on a different level than stipulated in the transport documentation. Such behaviour may lead to incurring big losses and should be prevented in any possible way. It can also result in the waiver of the insurer to pay compensation. This can be, of course, prevented by the use of telematics devices allowing to monitor and report the conditions inside the cold room along the entire route (TX-CONNECT BACK OFFICE SOFTWARE 2012).

Conclusions

The conducted research proved that the quality offered by the apple grower is rarely retained in storage and transportation, although it represents approximately 50-60% of the total expenditures incurred in the entire production cycle. The final level of quality of fruits and vegetables depends mostly on the proper choice of the means of transport and may help minimize the product losses, increasing the reliability of the forwarder and its good opinion among clients. The quality also depends on the distance that has to be covered in transport, and the longer the journey the higher product damages and losses. According to the analysis, transport in a vehicle fitted with a cold room is a better choice in case of fruits and apples in particular. It enables to keep the products in stable and controlled conditions throughout the time of transport, thus ensuring the conditions similar to those of proper storage and minimizing the damages. The best transport packaging in case of apples is a special pallet case (volume: 300 kg). The use of such pallet cases reduces the product losses. Efficient and cost-effective transportation of food products has to meet specific requirements regarding fruits and vegetables. In case of apples, the best way of transporting means that not only certain levels of

temperature and general conditions are kept, but re-loading and re-packaging is relatively rare as well. In the analysed transports re-loading and re-packaging was a common practice, which of course directly affected the final quality of apples. The effectiveness of the vehicle depends on the distance it has to cover, tonnage, time of loading and unloading, and the average speed of driving. These figures are interdependent; the effectiveness of transport is proportional to the tonnage and inversely proportional to the total time of transport. That is why a large tonnage vehicle will not be as effective as a smaller one when the distance is not very long.

The company that served for the purpose of this case study transported 5266 tons of apples in two years. The average level of product losses was 2.51%. The transport of apples was analysed from the perspective of tarpaulin-covered vehicles and vehicles with cold rooms. It has to be stated, however, that the results of transport in vehicles with a cold room were only an estimation because the company did not use such vehicles to transport food products.

The vehicle with a cold room is a better solution in food product transportation. It minimizes the product losses. An increase in freight costs by four thousand euro resulted in the level of losses decreasing by 60%. Therefore, it is altogether cheaper to transport apples in vehicles with a cold room, despite the fact that the cost of freight is higher than in case of the tarpaulin-covered vehicle.

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THE STUDY ON THE GLOBALIZATION STRATEGY OF JAPANESE SAKE - FOCUSED ON THE VISEGRÁD GROUP (V4) MARKET

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Introduction

Recently, the circumstance surrounding sake, the representative liquor of Japan, is becoming tough. The reason is that consumption of young generations, 20s and 30s, of Japanese traditional liquor (sake) is diminishing while demand for wine or other kinds of liquor is growing. Accordingly, production of Japanese liquor and the manufacturing companies which manage 'kuramoto' (place where Japanese sake is produced and stored) are both decreasing. The '도우지' (technician who makes sake) system which has long been maintained from the old time, and methods or know-how are being lost. According to statistics of Japanese Revenue Service, domestic consumption of Japanese liquor was below consumption of soju in 2003, and in 2008 the consumption dropped to almost 50% of the 1989 consumption.

On the other hand, Japanese food is in boom outside Japan and demand for Japanese foods such as sushi and Japanese liquor is growing all over the world. Japanese government is promoting globalization of Japanese liquor and foods, as a part of Japan Regeneration Strategy, targeting export expansion of agricultural and marine products and foods. The purpose of this paper is to review globalization strategy of Japanese liquor industry, to summarize the actions in the Eastern European market which is not large yet, particularly in the V4 market, and to study the future prospect.

Overseas expansion of Japanese liquor

Comparing export and import of overall liquor in Japan, import is dominating in the ratio of 1:10 in both volume and amount. This trend is expected to continue consistently in the future. In classification by item, beer

export shows substantial growth rate in both volume and amount. Sake, the Japanese traditional liquor, is still in growing trend of export although growth is slowed down recently. On the other hand, in import, import of sparkling wine is noticeably expanding, and import of wine is also in the recovering trend.

Export volume of sake was about 6700 kl in 1989, and after that kept growing to 12000 kl in 2008, which was almost twice of the 1989 volume, and to about 14000 kl in 2012. Export amount is also growing, and recorded 8.95 billion Yen in 2008.

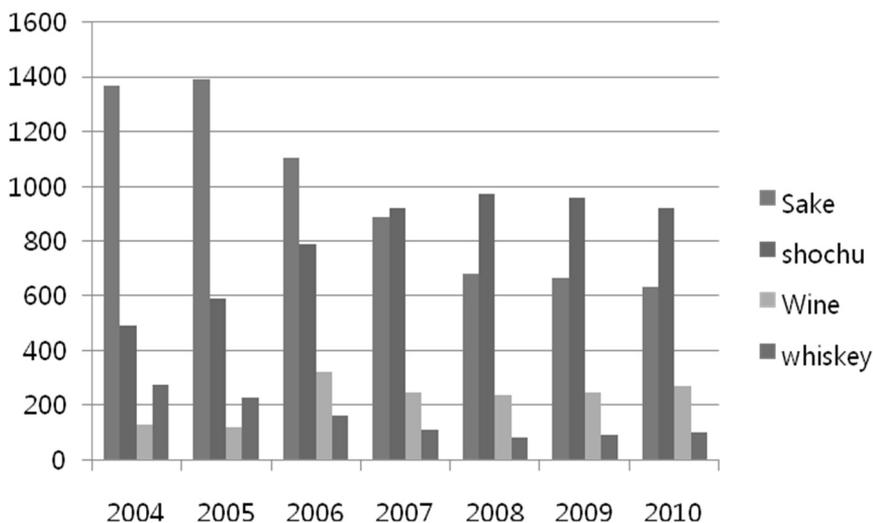


Figure 1. Alcohol Export value according to Country

Source: Agriculture, Forestry and Fisheries Department

In classification by importing country of Japanese liquor, in 2013 USA imported the most Japanese liquor, about 3,900 kl (28% of the whole export volume from Japan), Korea was 2nd with about 2,900 kl which was 20.6% of the whole export volume from Japan, Taiwan was 3rd (about 1,600 kl, 11.4%), and Hong Kong was 4th with about 1,500 kl which was 10.6% of the whole export volume from Japan. On the other hand, in amount, USA was still the highest with 3.25 billion Yen (36.3% of the whole export amount from Japan), Hong Kong was 2nd (1.50 billion Yen, 16.7%), and the rest of order was Korea (1.2 billion Yen, 13.5%), and Taiwan (0.51 billion Yen, 5.7%).

Table 1. SAKE Export value according to Country

	Country	Export (million yen)	Proportion of export(%)	Quantity	Proportion of export(%)
1	US	3245	36.3	3952	28.0
2	Hong Kong	1495	16.7	1492	10.6
3	South Korea	1204	13.5	2904	20.6
4	Taiwan	513	5.7	1603	11.3
5	China	412	4.6	666	4.7
6	Singapore	375	4.2	399	2.8
7	Canada	245	2.7	489	3.5
8	Australia	214	2.4	281	2.0
9	England	182	2.0	238	1.7
10	Thailand	168	1.9	446	3.2

Source: National Tax Agency

Export of Japanese liquor to Eastern European Region

“I heard that there were only 30 Japanese restaurants in this country in 2006 but the number has rapidly increased to about 270 now, 9 times in 7 years. If this trend continues, I think Japanese restaurants may be expanded to 2,500 places until 2020“ - this is a part of speech given by Japanese Prime Minister Abe when he visited Warsaw, Poland in June 2013 and did top sales of Japanese food. As can be noted from this statement, there is certainly a boom in Japanese food. Also, according to survey of JETRO (2012), in a few countries such as Turkey or Russia where Japanese food market is large, import of Japanese foods and agricultural & marine products is certainly decreasing, whereas in countries such as France, Greece, Ireland, Poland, Czech Republic, Latvia, Lithuania, Slovak, Hungary, Bulgaria, Estonia, and particularly Eastern European countries, demand for Japanese foods and liquor is reported to grow rapidly. Economically, the Eastern European occupation in the EU economy is largely expanded compared to the past, as the Eastern European GDP occupied about 5.4% of the EU GDP in 2012. Considering that the occupancy had been about 2% until 1995, the recent consistent growth is understandable.

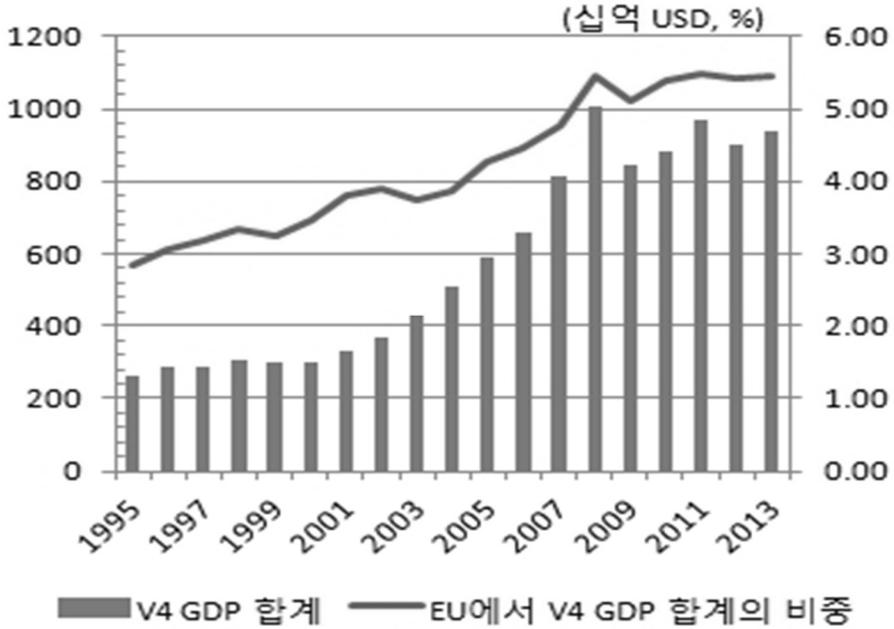


Figure 2. Visegrad V4 GDP & Scale of Trade

Source: IMF, KOTRA

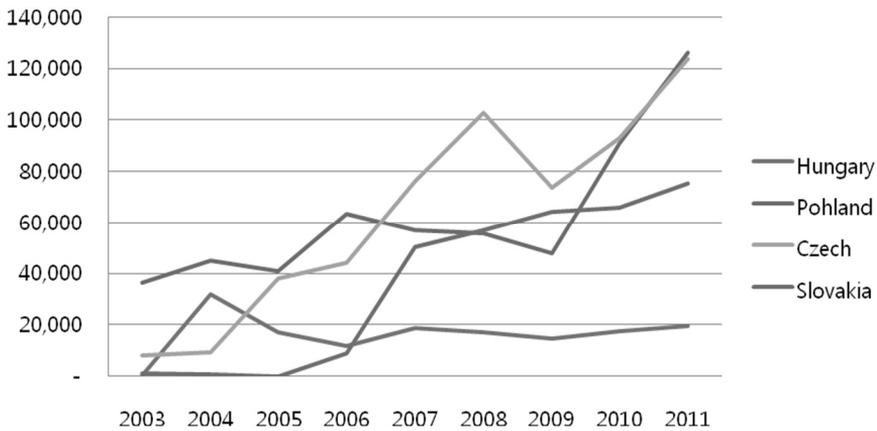


Figure 3. SAKE Export value according to V4

Source: Agriculture, Forestry and Fisheries Department

The rapid increase of demand for Japanese foods in recent Eastern Europe is considered to be caused by entering of multinational companies into Eastern

Europe due to economic vitalization there and the resulting expansion of resident employees, rise of standard of living in Eastern Europe, and growing residents of companies from advanced countries. Because rapid rise of demand for Japanese foods is directly connected to demand for Japanese liquor, studying future prospect of demand for Japanese foods is important.

Spread strategy of Japanese liquor in Eastern Europe

When spread of Japanese liquor in Eastern Europe is considered, it should be understood that demand for Japanese liquor in Eastern Europe is not so high right now. Certainly, it is in the growing trend compared to the past because of the increasing number of Japanese residents who live there, but demand from Japanese businessmen is certainly limited. To succeed in globalization of Japanese liquor, making Eastern European people recognize and like Japanese liquor is important. Localization effort from Japanese liquor manufacturers is needed until Eastern European people will recognize that Japanese liquor is good. Also, needless to say, development of localized products to the level that local residents may feel Japanese sake is good is important.

Director Ishige Naomichi of Japan National Folk Museum commented on the Japanese liquor sold in Eastern Europe, "I once went to Eastern Europe for business. Eastern or Central Europe may be called the region farthest in cultural distance from Japanese liquor or Japanese culture. When we prepare Japanese foods to local residents for tasting, we usually present Japanese liquor and tea, but the Japanese liquor I drank in Eastern Europe was really bad". Storage management and quality management is important because exporting from Japan to Eastern Europe takes time, and this incident implies that Japanese sake exported to Europe has still many problems in those aspects.

Taste is important above all to enter overseas market. However high the liquor is estimated in Japan, it is meaningless if the liquor loses taste in the actual locality of Eastern Europe. And, what is important is to make good use of the image Eastern European people have for Japan and to develop market. Under the premise of development of good taste Japanese liquor and quality management for export, this paper emphasizes the following as the points to be considered at the time of advancing to Eastern Europe.

First, Japanese restaurants should be used. First of all, Japanese liquor needs to be supplied in priority to Japanese restaurants in Eastern Europe, and information on local restaurants should be collected using information of JETRO or local Japan Chamber of Commerce.

Second, collaborative selling with local competitive distribution companies is important. In the long term, to obtain wide demand from the

world, companies that are trying to export Japanese liquor should have strong will to make Japanese sake or soju a global product. To make an internationally standardized product, whether the Japanese liquor sold overseas suits the needs of Eastern European people or otherwise product modification is necessary should be reviewed.

After such a review, collaborating with local distributors, departments, or supermarkets, liquor manufacturer will be required to develop products that will satisfy market trend or preferences of local markets. Overall actions aiming at global market need to be considered, such as quality including taste or scent, vessel, label design, vintage, ways of providing familiar with consumers. Not actions for Japanese or Japanese liquor mania residing overseas but serious market-developing mechanism should be constructed.

Third, products should be developed targeting Eastern Europe. Japanese liquor including sake is distributed mainly to overseas Japanese restaurants, but if it is to penetrate widely into each country in the world which has completely different food culture, ways to drink in harmony with foods or food culture of the country should be developed. What is necessary is not mere development of products customized for the country but aggressive promotion for those residents who lack understanding of Japanese liquor. Raising awareness of Japanese liquor is important, for example through using private companies, by using a famous chef in Eastern Europe to develop foods suitable for Japanese liquor, or holding cuisine contests suitable for Japanese liquor or cocktail contests using Japanese liquor, in main markets.

Forth, differentiation strategy from wine is also important. This is not a simple issue. The history of wine's internationalization effort and localization effort cannot be easily benchmarked by Japanese liquor companies. According to Michael Potter, professor of Harvard Business School, source of differentiation includes brand image, independent technology, design or performance of products, customer service, and sales channel. From the viewpoint of Potter's theory, the current position of Japanese liquor is still far behind. It is reality that when compared with wine through each of Potter's elements for strengthening competitiveness, Japanese liquor is not more competitive in any element.

What competitive advantages does Japanese liquor have as a source of differentiation? Strategies of Japanese whisky, Yamazaki or Hibiki, to advance to world may be useful references. In "World Whisky Award 2012" which is the world best competition, Japanese whisky Yamazaki 25 years (Santori) and Taketsuru pure malt 17 years (Nika) won the highest award in the field of whisky. This paper tries to seek overseas expansion strategy for Japanese liquor by finding the reason.

When launching the premium whisky brands (Yamazaki and Hibiki), to the world, Santori carried out concentration strategy as can be seen in

promotion of winning in world class competitions, giving tasting seminars, promotion of competitive quality and obtaining of PR, and focused selling to overseas Japanese restaurants. And, while competing companies pursued local procurement, local production and low price strategy, Santori considered production in Japan very important, and made strict management of quality putting development of tasty whisky ahead of everything. Also, it emphasized the image of Made in Japan and strict quality system, and pursued the strategy of high price.

What is revealed through these strategies is that as in the progress of whisky, developing Japanese liquor that satisfies overseas consumers with the goal of winning at international competitions is important. But, what should be noted is that whisky basically existed in the western world and so products customized for Europeans could be developed in a short period, whereas in the case of Japanese liquor or soju, Japanese food culture or liquor culture does not exist in Eastern Europe. How to develop a market and spread Japanese culture under this circumstance is a subject that requires serious consideration.

Conclusions

On Sep. 4, 2012, Japanese government announced “Program to export Japanese liquor or other” to promote “Globalization of Japanese liquor”. This program includes many important policies to encourage export such as establishment of marketing strategy, establishment of brands, strengthening of industrial basis, and local vitalization by creating liquor tourism.

While it is important to expand Japanese liquor business in the world market including Eastern Europe with combined efforts of government and civil sector, what is most important is efforts of Japanese liquor manufacturers to develop markets. Success will be unlikely outside Japan in the mindset of pride in “This is popular in Japan”. Also, the level of understanding local areas and creation of native mania who understand Japanese liquor are points to strengthen competitiveness of Japanese liquor there.

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