

“Scientific researches in food production”, University of Debrecen, 7 September, 2018

University of Debrecen

Faculty of Agricultural and Food Sciences and Environmental Management



“Scientific researches in food production”



UNIVERSITY OF DEBRECEN
7th SEPTEMBER 2018

Proceeding of abstracts

7th September, 2018

Debrecen, Hungary

Young Researchers from University of Debrecen, University of Rzeszow, University in Zlin and Slovak University of Agriculture in Nitra

„Scientific researches in food production”

Proceeding of abstracts

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DAB Agrokémiai és Élelmiszertudományi Munkabizottság

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8.30-9.00 – Registration (in front of VIP room)

9.00-9.30 – Open ceremony (VIP room)

9.30-10.00 – Poster section and coffee break

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FUNGAL CHARACTERIZATION USING MATRIX-ASSISTED LASER DESORPTION TIME-OF-FLIGHT MASS SPECTROMETRY (MALDI-TOF MS)

10.00-12.00 – Oral presentations

10.00-10.15

E. Ivanišová, A. Kántor, H. Francáková, M. Kačániová

BIOACTIVE COMPOUNDS AND ANTIOXIDANT ACTIVITY OF SELECTED *VITIS VINIFERA* L. VARIETIES FROM THE SMALL CARPATHIANS WINE REGION

10.15-10.30

L. Godočíková, E. Ivanišová, M. Kačániová

MICROBIOLOGICAL STABILITY AND HYGIENIC QUALITY OF SLOVAK CHOCOLATES

10.30-10.45

A. Abid, J. Remenyik, E. Máthé

EFFECT OF SOUR CHERRY EXTRACT ON FLIES (*DROSOPHILA MELANOGASTER*)

10.45-11.00

P. Hanus, M. Kluz, M. Kačániová

IDENTIFICATION ON *LISTERIA MONOCYTOGENES* IN CHICKEN AND PORK MEAT AFTER HEAT TREATMENT WITH MALDI-TOF MASS SPECTROMETRY

11.00-11.15

L. Ismaiel

THE EFFECT OF REFRIDGERATION STORAGE CONDITION ON THE MOST IMPORTANT SENSORY, CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS OF PASTIRMA

11.15-11.30

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EXTRACTION OF FLAVONOIDS FROM MEDICINAL PLANTS WITH USE OF VARIOUS SOLVENTS SYSTEMS

11.30-11.45

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THE COMPARISON OF COUMARIN AND PHENOLIC COMPOUNDS CONTENT IN WILD AND CULTIVATED SWEET CLOVER (*MELILOTUS*)

11.45-12.00

A. R. Tóth, A. Tóthné Bogárdi, T. Rubóczki, M. Takácsné Hájos

RHEOLOGICAL EVALUATION OF INDUSTRIAL TOMATO IN RAW AND PROCESSED CONDITION

12.00-14.00 – Lunch

EFFECT OF SOUR CHERRY EXTRACT ON FLIES (*DROSOPHILA MELANOGASTER*)

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Sour cherry (*Prunus cerasus*) is consumed as a nutritional supplement claiming health effects. Ingesting sour cherries lowers uric acid levels in the blood, which helps prevent gout. In fact, cherry juice is an excellent natural remedy for gout pain. It is also known as a very good source of melatonin, which may help prevent breast cancer.

The aim of present study was to analyze the effect of our sour cherry extract on the viability of *Drosophila melanogaster*. The reason for choosing the fruit fly for this experiment was the straightforward monitoring of the immediate effects caused by food media supplemented with the given sour cherry extract.

In this experiment the w^{m4h} fruit flies were grown on four different food media such as zero-nutrient media, 0.15M normal-sugar media, 0.75M high-sugar media and 1M high-sugar media with or without of sour cherry extract. Results are indicating significant differences regarding the fruit fly of rate of development that varies in function of sugar, nutrient and sour cherry content.

The survival rate associated with the extract supplemented zero-nutrient media indicates that the sour cherry extract does contain substantial amount of nutrients that could facilitate the development of fruit flies. It is also interesting that the sour cherry extract did affect in a concentration dependent manner the viability of fruit flies in conjunction with the sugar content of the fruit fly food media. Our results are suggesting that the studied sour cherry extract represents a highly valuable nutrient source suitable for nutraceutical development.

MICROBIOLOGICAL STABILITY AND HYGIENIC QUALITY OF SLOVAK CHOCOLATES

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Microbiological stability and hygienic quality is a considerable issue during the chocolate production, even despite its low water activity. Outbreaks of various diseases from chocolate are just the proof of importance of this issue to be considered when examining the chocolate quality. The aim of this study was therefore to analyze samples of chocolates produced in Slovak republic for their microbiological stability and to identify present microbiota on the mass spectroscopy principle. MALDI-TOF mass spectrometry has changed the process of microbial identification, allowing a more precise and faster result than the conventional methods. Total viable counts were lower than $2.32 \log \text{CFU.g}^{-1}$ in all samples. Lactic acid bacteria were found only in one sample, with number of count $0.69 \log \text{CFU.g}^{-1}$. Yeasts were present in the samples only up to $2.51 \log \text{CFU.g}^{-1}$, whereas microscopic filamentous fungi were detected only in one sample with the addition of almonds, which could probably affected this result. Nevertheless, none of the samples examined included coliform microorganisms which are an indicator of hygienic quality. Based on the study of morphological features, we have identified the microscopic filamentous fungi as *Aspergillus* sp. The other microorganisms were identified by mass spectrometry using the MALDI-TOF MS Biotyper equipment. In samples we further identified the yeast *Candida parapsilosis*, which is most often isolated from the hands of healthy people. Commonly occurring genus was *Bacillus* genus and from these microorganisms *B. pumilus*, *B. subtilis* and *B. mojavensis* species. *Bacillus* spore-forming bacteria are able to survive technological steps such as fermentation and drying of cocoa beans and even roasting at temperatures above $100 \text{ }^\circ\text{C}$.

Acknowledge: The study was supported by the European Community project No 26220220180: Building Research Centre „AgroBioTech“ and by grant of Slovak Research and Development Agency No. VEGA 1/0411/17. The authors are sincerely grateful to two Slovak chocolate producing factories for providing samples.

Key words: chocolate, stability, safety, quality, microorganisms, MALDI-TOF mass spectrometry

IDENTIFICATION ON *LISTERIA MONOCYTOGENES* IN CHICKEN AND PORK MEAT AFTER HEAT TREATMENT WITH MALDI-TOF MASS SPECTROMETRY

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Foodborne pathogen *L. monocytogenes* is a widely dispersed in the environment; it is found in soil, water, plant material and is the cause of listeriosis. *L. monocytogenes* can growth at refrigeration temperature and at unfavorable conditions of pH (up to pH 4.7) and salt (up to 10%). It can persist in the harsh conditions of the food processing environment from which it can contaminate food.

The aim of this study was to verify the survival of *L. monocytogenes* in chicken and pork meat after heat treatment. Quality was determined using classical microbiology methods during growth on selection media. The microbiota of the examined chicken and pork meat was determined using the MALDI-TOF MS Biotyper.

Materials and Methods

In our study chicken and pork meat were used. Ten gram (± 0.5 g) of samples were placed in PA/PE film bags and 100 μ l of *L. monocytogenes* CCM 4699 suspension was added. The samples were vacuum packed and incubated at 37 °C for 18 hours. Samples of meat were subjected to thermal treatment at 55 °C, 60 °C, 65 °C, at various times: 5, 15, 30 and 60 min in a water bath. After homogenization at 0.87 % NaCl, Trypticasein Soy Lab-Agar TSA (Biocorp) and Listeria acc. Palcam Lab-Agar Base (Biocorp) were inoculate. Samples were tested on day 1 and after 3 days. The numbers of colony forming units per gram (cfu/g) were determined and identified using mass spectrometry equipment using MALDI-TOF MS Biotyper (Bruker, Germany).

Results

The numbers of colony forming units of *L. monocytogenes* after different temperature and time treatment in the selective medium were detected. 482 isolates were identified, 349 (72.40%) of isolates were highly probably species identification, 42 (8.71%) isolates were probably genus identification, 91 (18.88%) isolates were not has reliable identification. The analysis of samples from the selection medium confirmed the presence of *L. monocytogenes* bacteria. Studies on the survival of *L. monocytogenes* show appropriate heat treatment time to eliminate the bacteria. One hour for 55 °C it is necessary for killing of bacteria. Thirty minutes with temperature of 60 °C is necessary for chicken meat and 15 min heat treatment is necessary for pork meat. Five and fifteen min with temperature 65 °C heat treatment of *L. monocytogenes* kill bacteria in chicken meat and five min with temperature 65 °C kill bacteria in chicken meat. In chicken meat is the majority microflora gram negative bacteria and in pork meat it is gram positive bacteria. The heat-treated products should be prepared with care, under hygienic conditions and at the right temperature and for the right period of time.

THE EFFECT OF REFRIDGERATION STORAGE CONDITION ON THE MOST IMPORTANT SENSORY, CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS OF PASTIRMA

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Pastirma samples manufactured from beef and covered with a layer of fenugreek, garlic, salt and spices were collected from different commercial centers and transported to the laboratory. The samples were stored at commercial storage conditions (7 ± 2 °C), which are usually the conditions for refrigeration storage in the markets, within storage time (0, 15, 30, 60, 120, and 180 days). Some of the most important sensory, chemical and microbiological characteristics were investigated. The results of the sensory analysis showed that storage negatively affects the sensory properties of the Pastirma and leads to significant loss especially after four months of storage. The most sensitive properties were: texture, appearance and smell. Moisture content decreased marginally from 43.476% to 40.989% after 180days of cold storage. However, there was a significant (<0.05) difference in PH, acidity, fat, Peroxide number, dissolved nitrogen (non-protein) and volatile nitrogen. PH values, and fat were decreased significantly from 6.14 to 5.51 and 7.475% to 5.835% respectively after 30days of cold storage. However, acidity, peroxide number, dissolved nitrogen (non- protein) and volatile nitrogen increased significantly from 0.181 to 1.124%, 2.012 to 2.898meq/kg, 0.169 to 0.268%, 0.005 to 0.035% respectively after 30 days. The microbial results indicated that the Pastirma is free from pathogenic bacteria *salmonella* and *Escherichia coli*. The results also showed a significant ($p<0.05$) increase in the number of aerobic bacteria, yeasts and fungi (10.182 ± 0.85 , 9.899 ± 0.69) after 180days of storage respectively. However, they were within acceptable limits.

Keywords: Pastirma, storage condition, sensory qualities, chemical and microbiological parameters

BIOACTIVE COMPOUNDS AND ANTIOXIDANT ACTIVITY OF SELECTED *VITIS VINIFERA* L. VARIETIES FROM THE SMALL CARPATHIANS WINE REGION

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Vitis vinifera L. is known for its health benefits and high antioxidant activity mainly due to phenolic content. Phenolic compounds play very important role not only in human health, but also in grape, wine quality and sensory properties. The aims of the present study was to determine the antioxidant effect as well as content of phytochemicals in three red grape varieties (Alibernet, Cabernet Sauvignon, Frankovka modrá) and three white varieties (Sauvignon blanc, Rizling vlašský, Rulandské biele) produced in Small Carpathians wine region of Slovakia (locality Vrbové, 2016). The content of phytochemicals (polyphenols, phenolic acids, flavonoids, and in red varieties also anthocyanins) was measured by colorimetric methods. The detection of antioxidant activity was carried out by DPPH and phosphomolybdenum method. Results of antioxidant activity showed that from red grape varieties the sample Alibernet had the best activity tested by both methods (0.49 ± 0.02 and 23.61 ± 0.02 mg TEAC/g fresh matter; TEAC – Trolox equivalent antioxidant activity), and from white varieties the sample Rulandské biele (0.4 ± 0.01 and 6.81 ± 0.08 mg TEAC/g fresh matter). In these samples were also detected the highest content of total polyphenol (Alibernet 3.53 ± 0.42 mg GAE/g, Rulandské biele 0.33 ± 0.05 mg GAE/g; GAE – gallic acid equivalent) as well as total flavonoid content (Alibernet 1.24 ± 0.01 mg QE/g, Rulandské biele 0.23 ± 0.01 mg QE/g; QE – quercetin equivalent) and phenolic acid content (Alibernet 1.77 ± 0.03 mg CAE/g, Rulandské biele 0.21 ± 0.01 mg CAE/g; CAE – caffeic acid equivalent). In sample Alibernet was also determined the highest level of total anthocyanin content (4.39 ± 0.24 mg/g fresh matter) with compare to other tested red varieties. The obtained results confirmed that grape is a good source of phytochemicals and grape extracts should be developed as health supplement products.

Acknowledgement: This work was co-funded by VEGA project 1/0411/17

Key words: grape, fresh matter, polyphenols, flavonoids, phenolic acids

EXTRACTION OF FLAVONOIDS FROM MEDICINAL PLANTS WITH USE OF VARIOUS SOLVENTS SYSTEMS

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Flavonoids are plant secondary metabolites, exhibiting a wide spectrum of bioactivities, including antioxidant, antiinflammatory, antimicrobial or anticancer effect. They also belong to important ingredients of functional and pro-health food. Flavonoids and other polyphenolic compounds are usually obtained by solid-liquid extraction, with use of polar solvents, such as methanol, ethanol, acetone or ethyl acetate. In the described research, the micelle mediated extraction method was used to obtain flavonoid-rich extracts from several medicinal plants.

As raw materials to extraction three commercially available medicinal plants were used: yarrow (*Achillea millefolium*, herb), agrimony (*Agrimonia eupatoria*, herb) and three-lobed beggartricks (*Bidens tripartita*, herb). The extraction was carried out in ultrasonic bath with use of following solvent systems: Triton X-100, Nonidet P-40 and Tween 20 (2% v/v solutions) and acetone-water mixture (1:1) for comparison. In obtained extracts three flavonoids were quantitatively determined using UHPLC-MS method: luteolin, luteolin-7-glucoside and chrysoeriol.

The results clearly indicate, that aqueous surfactant solutions could be effective solvents for flavonoid aglycone extraction, luteolin glucoside was absent or present in trace amounts in these extracts. The highest content of luteolin was recorded in *Agrimonia* and *Bidens* micellar extracts made with Triton X-100 and Nonidet P-40 (up to 50 µg per one milliliter of extract in case of agrimony, Triton X-100). Large amounts of chrysoeriol (3'-methylluteolin) were detected in yarrow (the most in Triton and Nonidet extracts, about 12 µg/ml), slightly less in agrimony and three-lobed beggartricks. In contrast, in water-acetone extracts significant amount of luteolin glycoside was found (up to 50 µg/ml of *Herba Bidentis* extract), with much less content of flavonoid aglycones. Moreover, chrysoeriol was first time detected in *Bidens tripartita* and *Agrimonia eupatoria*; according to our knowledge there is no literature data on the occurrence of this compound in these plants.

Summarizing, it should be said that simple surfactant assisted extraction could be an alternative to organic solvent extraction, especially for flavonoid aglycones. Of course, it is possible to control the composition of plant extracts by selection of proper solvent system. In the case of surfactants their properties and potential risk must be taken for consideration. In the applications concerning food it should be remembered about surfactant residue removal.

Key words: flavonoids, dandelion, yarrow, agrimony, three-lobed beggartrick, micelle mediated extraction

**THE COMPARISON OF COUMARIN AND PHENOLIC COMPOUNDS CONTENT IN
WILD AND CULTIVATED SWEET CLOVER (*MELILOTUS*)**

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Melilotus L. Mill commonly known as sweet clover or melilot is a widespread plant belonging to the *Fabacea* family. There are approx. 25 species including white sweet clover (*M. albus*), yellow sweet clover (*M. officinalis*) or tall yellow sweet clover (*M. altissimus*). In its natural environment grows in a habitat of roadsides and waste places. Sweet clover are utilized as feed production, green manure and cover crop. Unfortunately, it has not been widely used because *Melilotus* can contain high levels of coumarin that can be converted into dicoumarol upon fungal contamination. Dicoumarol is an anti-coagulant, which can cause internal bleeding. The content of coumarin is very diverse depending on the variety, as well as the country of origin.

The aim of this study was to compare the content of coumarin as well as phenolic compounds in wild and cultivated sweet clover. The research material was the harvest and dried *M. albus* and *M. officinalis* flowers and leaves obtained from their natural habitat localized in the Podkarpackie Voivodeship (biennial *M. officinalis* -3, annual *M. albus* -1, biennial *M. albus* -5) and from experimental plots, where additionally different seed density was used (3, 6, and 9 g/m²) (annual and biennial *M. albus*). Samples were collected in July 2015 and 2016. Qualitative and quantitative analysis of coumarin and related compounds was carried out using high-performance liquid chromatography (HPLC) with UV- ViS detection. Additionally, o-coumaric, p-coumaric, melilotic and vanilic acid have been identified.

The samples exhibited varying amounts of coumarin. Generally, flowers contained a higher content of this compound compared to leaves, as well as biennial form was richest in coumarin than annual form. Cultivated sweet clover showed similar content of coumarin as wild plant.

Key words: sweet clover, *M. albus*, *M.officinalis*, coumarin, phenolic compounds, HPLC

RHEOLOGICAL EVALUATION OF INDUSTRIAL TOMATO IN RAW AND PROCESSED CONDITION

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Of the vegetables, the tomato has one of the biggest producing areas in the world, and its size shows an increasing trend not only abroad, but in Hungary as well. The vast majority of the tomato yield is processed by industry.

Nowadays, selecting the proper genotype is one of the main steps to fulfil the requirements of the industry. Six genotypes were evaluated (*Heinz 1015 F₁*, *Heinz 9478 F₁*, *Kecskeméti 407*, *NUN254 F₁*, *Prestomech F₁* and *Rustico F₁*) in the experiment for the following quality parameters – fruit shape index (length/diameter), firmness (g/cm²), force needed to tear the skin (kg⁻¹), consistency of tomato puree (g*sec).

The purpose of the experiment was to determine which genotype has the most suitable fruit quality parameter to fulfil the actual requirements of the industry.

According to the results, the *Heinz 1015 F₁* and *Heinz 9478 F₁* hybrids had shown oval shape index (1.3), while the *Kecskeméti 407* genotype was close to spherical shape (1.1). Highest firmness of fruit was detected in the samples of *Heinz 1015 F₁* and *Heinz 9478 F₁* hybrids (1602 g/cm² and 1458 g/cm²) against *Rustico F₁* (1243 g/cm²). All examined variety fulfilled the requirements of mechanical harvest, so several values were higher than 1000-1200 g/cm².

The firmness of skin and the consistency of tomato juice were measured by rheological methods. The highest value was measured in *Kecskeméti 407* genotype (0.68 kg⁻¹), which showed seven times higher values than other hybrids (0.067-0.089 kg⁻¹). It is a great advantage for mechanical harvest, however during the industrial process it can be a disadvantage because it can increase the waste ratio of raw material.

With the consistency we could evaluate the density of the juice. The *Heinz 1015 F₁* and *Heinz 9478 F₁* hybrids showed higher values (896 and 786 g*sec) compared to *Kecskeméti 407* genotype (546 g*sec). Rather high correlation was found between shape index and consistency (r = 0.891), which was similarly observed in the case of fruit firmness and juice consistency (r = 0.882). The proper juice outcome is determined by oval fruit shape (less chambers) and firmness of fruit, which means the slower breakdown of pectin content.

It can be concluded, that the most suitable hybrids for processing are *Heinz 1015 F₁* and *Heinz 9478 F₁* by the quality parameters and rheological evaluation.

Acknowledgement: The publication was supported by the EFOP-3.6.3-VEKOP-16-2017-00008 project. The project is co-financed by the European Union and the European Social Fund.

Keywords: industrial tomato, tomato genotypes, rheology

EXAMINATION OF CHANGES IN THE ANTIOXIDANT COMPOUNDS AND ACIDITY OF THE MASH OF DIFFERENT FRUITS

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Hungary is famous of its high-quality fruits and fruit products, such as jams, juices, frozen products or pálinka, which is a special Hungarian spirit. Fresh fruits contain significant amounts of healthy components, for example antioxidant compounds, organic acids, etc.

In this study, 2 apple varieties were used for preparing a mash, and their dry matter content, total phenolic content, flavonoid content and acid content were measured before and after distillation.

According to the results, it can be declared, that there are differences between the apple varieties in the parameters mentioned above, which difference can be observed during the whole mashing. The concentration of phenolic compounds and flavonoids were decreasing during the mashing. On the other hand, the acid content of the samples increased. Unfortunately, the concentration of these compounds was extremely low in the primary distillate, for instance, the concentration of flavonoids was under the limit of detection. Therefore, they are not likely to be present in the final product either.

In this study, the mash and primary distillate of two apple varieties were examined. The differences between the varieties could be observed during the mashing, and changes in the total phenolic content, flavonoid content and acid content could also be seen, but the concentration of these components were extremely low in the primary distillate. Further studies would be necessary to find compounds of the raw materials in the final product.

Key words: apple, mashing, spirit, antioxidant, acid content

GROWTH OF GREEN PEA BY EFFECT OF MOLYBDENUM TREATMENTS AT DIFFERENT DEVELOPMENTAL STAGES

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Molybdenum is one of the seven microelements which is essential for the plants. The Mo content of plants is generally low, rarely exceeding 1 mg kg⁻¹ dry weight, although there are differences among plant species. For example, legumes accumulate much more Mo than other plants do.

The main aim of the present study was to examine whether increasing molybdenum (Mo) concentration affects on dry mass of green pea plants (*Pisum sativum* L.) in pot experiment at different developmental stages. In this experiment calcareous chernozem soil (Látókép) was used and molybdenum was supplied to the soil as sodium molybdate (Na₂MoO₄·2H₂O) in five different concentrations as follow: 0, 3, 30, 90 and 270 mg kg⁻¹ Mo. The determination of the dry mass of green peas was carried out in four different stages of development which were the following: four-node condition, the beginning of flowering, green ripening, and complete maturity.

In this study we have found that the amount of molybdenum concentration has no significant effect on the dry mass product of the generative organs. In the case of vegetative organs, however, the 30 mg kg⁻¹ Mo-treatment increases the dry mass growth in a statistically verifiable way. However, above 30 mg kg⁻¹ Mo-treatment there is a decrease in the dry mass of vegetative parts, the values are similar to the control results statistically.

Key words: green pea, calcareous chernozem soil, molybdenum, dry mass

PHYSICOCHEMICAL AND ORGANOLEPTIC CHARACTERIZATION OF KEFIR WITH A DIFFERENT PROTEIN CONTENT

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Kefir is consumed around the world and has been for centuries. Kefir has several health benefits such as boosts immunity, builds bone strength, supports digestion. Additionally, improves allergies, heals skin and improves lactose intolerance symptoms. This product contains a large amount of easily absorbable calcium, potassium, phosphorus, and protein. These are ingredients that keep our bones and teeth in good shape. The Kefir market is growing at a fast pace and has huge opportunities with the ongoing rise in product innovations.

The aim of the work was to analyze the physicochemical and organoleptic properties of kefir available in the city of Rzeszów, Sub-Carpathian region, Poland. The quality of the kefir was analyzed depending on the amount of protein. The physicochemical properties such as pH, total acidity ($^{\circ}\text{SH}$), lactic acid [g/l] and syneresis effect were evaluated. All analysis was carried out in five replications. Moreover, examined consumer preferences regarding kefir consumption and sensory characteristics. A sensory profile of kefir was evaluated using a 10-point scale to assess the preferences of customers.

The research material used in the experiment were kefir purchased in various commercial establishments. Nine kefir from various producers were selected for the study. Kefir were characterized by the different protein content of 2.90g/100g to 4.10g /100g. Different contents of protein in kefir was also accompanied by significantly different fat and carbohydrate content. The studies divided kefir due to protein content into three groups. In the first group, the average protein content was 2.90%, in the second group at 3.35% and in the third group at 4.03 %. In the group with the highest protein content, the highest pH value was found pH 4.49 and the highest titratable acidity of 40°SH . Kefir with different protein content were characterized by a different content of lactic acid. In the group with the highest protein content (4-5 %), the highest lactic acid content 0.84 g/l and the lowest amount of lactic acid in the group containing <3 % protein were determined. The protein content significantly influenced the syneresis effect.

The highest syneresis effect was observed in the group with a protein content of 3.0 to 3.95 %. Sensory analysis showed that the kefir with protein content <3 % were the most preferred taking into account the consistency parameter. The different protein content in kefir did not affect other determined parameters such as taste, aroma, color and CO_2 saturation.

Key words: kefir, protein content, physicochemical properties, consumer preferences

IDENTIFICATION OF FOOD ASSOCIATED *STAPHYLOCOCCUS AUREUS* WITH MALDI-TOF MS

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Staphylococcus aureus is recognized as one of the most common pathogens responsible for food poisoning and giving rise to various infections in humans and animal. Express and dependable methods for identification of bacterial isolates are mainly dependent on phenotypic and genotypic characteristics of the bacteria.

The MALDI-TOF MS technique is based on coupling a laser ion source and a time-of-flight mass spectrometer. The energy of the laser emitted is absorbed by the matrix, which in turn ionises the target compounds of the sample, and thus they can enter the vacuum of the mass spectrometer and finally reach the detector. The mass spectra obtained by MALDI-TOF technique provides information on the protein and macromolecular profile of the bacteria. The aim is to be able to identify *Staphylococcus aureus*, cheaper and more reliably in a routine diagnostic laboratory.

In the present study 20 food samples were collected from different food and screened for food-borne Staphylococci with a selective medium (Baird-Parker). The strains collected from food were identified by MALDI BioTyper.

Acknowledge: The work/publication is supported by the EFOP 3.6.1-16-2016-00022 project. The project is co-financed by the European Union and the European Social Fund.

Key words: MALDI-TOF MS, food-borne *Staphylococcus aureus*

THE EFFECT OF REFRIDGERATION STORAGE CONDITION ON THE MOST IMPORTANT SENSORY, CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS OF PASTIRMA

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Pastirma samples manufactured from beef and covered with a layer of fenugreek, garlic, salt and spices were collected from different commercial centers and transported to the laboratory. The samples were stored at commercial storage conditions (7 ± 2 °C), which are usually the conditions for refrigeration storage in the markets, within storage time (0, 15, 30, 60, 120, and 180 days). Some of the most important sensory, chemical and microbiological characteristics were investigated. The results of the sensory analysis showed that storage negatively affects the sensory properties of the Pastirma and leads to significant loss especially after four months of storage. The most sensitive properties were: texture, appearance and smell. Moisture content decreased marginally from 43.476% to 40.989% after 180days of cold storage. However, there was a significant (<0.05) difference in PH, acidity, fat, Peroxide number, dissolved nitrogen (non-protein) and volatile nitrogen. PH values, and fat were decreased significantly from 6.14 to 5.51 and 7.475% to 5.835% respectively after 30days of cold storage. However, acidity, peroxide number, dissolved nitrogen (non- protein) and volatile nitrogen increased significantly from 0.181 to 1.124%, 2.012 to 2.898meq/kg, 0.169 to 0.268%, 0.005 to 0.035% respectively after 30 days. The microbial results indicated that the Pastirma is free from pathogenic bacteria *salmonella* and *Escherichia coli*. The results also showed a significant ($p<0.05$) increase in the number of aerobic bacteria, yeasts and fungi (10.182 ± 0.85 , 9.899 ± 0.69) after 180days of storage respectively. However, they were within acceptable limits.

Keywords: Pastirma, storage condition, sensory qualities, chemical and microbiological parameters

PRODUCTION OF SELENIZED MILK

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Selenium was regarded as a toxic element at the beginning of the 20th century, while in 1957 Schwarz and Foltz proved that selenium was essential to our body. Selenium deficiency can lead to the development of diseases that have a serious and lasting health hazard. The role of selenium has significantly appreciated over the past few years. Hungarian soils have insufficient selenium content, so we need selenium complementation. The easiest way for a consumer to take in selenium with basic foods. In our research, we investigated whether high selenium milk can be produced by animal feed. In our experiment, we fed three Simmental cows with selenium-enriched feed. We measured the Se content of the fodder and the milk samples before and during the experiment. We came to the conclusion that oral selenium supplement affects the selenium content of milk, thus enabling the production of products with high Se content. We measured the selenium content with ICP-MS, the 1st table contains the results.

1st table: Selenium content of milk

Experimental group	Selenium content of milk ($\mu\text{g}/\text{kg}$)
Control (basic fodder)	17,9
1 mg Se enrichment	31,8
2 mg Se enrichment	53,2
4 mg Se enrichment	80,9

According to our measurements, milk produced by us can be a good alternative to satisfy our daily selenium needs.

In our further research, we want to model the excretion of selenium from the body of cattle. We will investigate whether the selenium content of the experimental milk is present in the dairy product and how the cultures respond to the presence of selenium.

Key words: milk, selenium, functional food, feed, cow

DETERMINATION OF NUTRITIONAL PARAMETERS OF DIFFERENT CEREAL FLOURS' BREAD

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Bread is an important source of food for human species. This product is a source of carbohydrates, proteins, dietary fibers, vitamins, micronutrients and antioxidants.

In this study, the nutritional parameters (dry matter-, protein-, fat-, ash-, flavonoid-and TP content) of 16 breads were examined. The breads were made from the following flours: 1-wheat flour 55, 2-wheat flour 80, 3-bio whole wheat flour, 4-graham wheat flour, 5-light rye flour, 6-whole grain rye flour, 7-fine rice flour, 8-whole grain fine brown rice flour, 9-white spelt wheat flour, 10-whole spelt wheat flour, 11-oat flour, 12-oatmeal flour, 13-corn flour, 14-sorghum flour, 15-barley flour, 16-millet flour. There was no significant difference in the dry matter content.

In the protein content, the highest values (>8%) were measured in breads which were made from wheat and millet flours. These values depend on the protein content of the flour. The fat content of breads did not exceed 1.3% in any case. The result is also influenced by the raw material and flour. The ash content of the products was between 1.64-2.72%. There are several factors that influence this. The flavonoid and TP contents showed wide ranges. The highest value for both parameters was measured in the sorghum bread, and the lowest was found in the fine rice flour bread. However, it is important to note that the TPC and flavonoid values of samples 3, 4, 5, 6, 10, 11, 14 and 15 were high compared to the others.

This test was important for the selection of the four types of flour that we would use for further research based on the appearance, behaviour and nutritional parameters of the finished breads.

Key words: bread, flour, nutritional parameters

EFFECT OF PREHARVEST FLUOPYRAM TREATMENTS FOR THE SURFACE MOLD NUMBER AND THE SHELF LIFE OF SOUR CHERRY (*PRUNUS CERASUS* L.)

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The economic loss of fruits is estimated to be around 20-25%, mainly due to the postharvest diseases. In case of cherry (*Prunus avium* L.) and tart cherry (*Prunus cerasus* L.), half of the lost was estimated caused by molds. Preharvest fungicide application has been demonstrated to be effective against fungal decay of different stone fruits during storage.

Fluopiram was applied two weeks before harvest, and its efficiency was examined on three Hungarian varieties (‘Érdi bőtermő’, ‘Újfehértói fürtös’ and ‘Petri’). The CFU and the morphology based identification of molds were detected following harvest. Mold were isolated from rotted fruits during shelf-life studies and identified based on morphological and molecular characters. Disease incidence (DI) was also detected at 20°C. These tests were repeated after 6 weeks storage at 0-2°C, in normal and modified (MAP) atmospheres.

Surface mold contamination of fruits was higher at the harvest than after 6 weeks cold storage. Similar mold species were isolated from different varieties, and storage conditions did not affected tis parameter either. The applied fluopiram fungicide reduced both the number of molds and the ratio of the rotten fruits of the harvested fruits. However its positive effect for the shelf-life was not so clear after the storage. Modified atmosphere packaging could decrease rotting in some cases.

Acknowledge: The publication is supported by the EFOP-3.6.3-VEKOP-16-2017-00008 project. The project is co-financed by the European Union and the European Social Fund.

Key words: *Prunus cerasus*, preharvest, postharvest, fungicide, fluopyram, Luna Privilege

GC-MS ANALYSIS OF FLAVONOIDS: SAMPLE PREPARATION METHODS AND DERIVATIZATION

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Flavonoids are a class of diet-derived antioxidants, which contribute to the prevention against cancer, cardiovascular, and age-related diseases. As a result, considerable attention has been devoted to the analysis of flavonoids in a variety of samples.

Several chromatography techniques have underpinned many chemical analysis methods, developed for superior flavonoid separation and identification. Among these, GC-MS is one of the most powerful tools of separation technologies, providing precise measurement of a wide range of flavonoids. This summary study features developments in the application of GC-MS for the determination of flavonoids, focusing on flavonols including quercetin, myricetin, and kaempferol from the perspective of sample preparation and GC-MS analysis.

Key words: flavonoid, quercetin, myricetin, kaempferol, separation, GC-MS

ORGANIC MILK CHEESE WITH HERBS AND SPICES

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Cheese consumption in Poland is dominated by fresh cheeses. Twarog - acid curd fresh cheese, as traditional dairy product constitute a very large and diversified group of products. In the group of acid cheeses are also found flavoured, smoked, or fried products (Ziarno and Zaręba, 2013). This fresh cheese is relatively low-energy value product with a high content of valuable and easily digestible protein. Twarog contains a fraction of easily digestible milk fat and lactose as well as vitamins and minerals (Siemianowski and Szpendowski, 2014).

The aim of this study was to evaluate the sensory quality and physicochemical parameters of cheese flavoured with herbs and spices produced from organic milk. Milk was collected in Spring from organic farm from Podkarpacie. Twarog cheese was produced from pasteurized (72°C, 15 s) reduced fat milk (A – 0,6% fat, B – 1,6% fat) by the acid method of coagulation (26°C, 18 h), with mesophilic cultures Flora Danica (Chr. Hansen, Poland). The spices and herbs were added to the cheese A (1% fat) and B (3% fat): I - wild garlic (0.4%), parsley (0.4%), black ground pepper (0.2%), II - onions (0.6%), chives (0.4%), III - caraway (0.5%), hot pepper (0.2%), sweet pepper (0.3%). The organoleptic evaluation was examined in 9-point scale, where "1" meant the least perceptible feature and "9" - the most intense feature. Moreover, there was determined in twarog active acidity (pH), total acidity (by Soxhlet-Henkel method), and dry matter (by oven drying method).

Differences were observed in the acidity of twarog with herbs and spices. The highest total acidity was measured in 1% fat cheese (A) with the addition of wild garlic, parsley and ground pepper (84.00°SH). Total acidity measured in 3% fat twarog (B) was 70,50°SH in all groups (I-III) of cheese. In the opinion of the sensory panel, the intense flavours such as hot pepper and black pepper were less preferred than traditional additives like onions and chives. 3% fat cheese with the addition of onions (0.6%) and chives (0.4%) was most preferred by the sensory panel. These cheese was characterized by a very characteristic light green colour with visible particles of additives.

Key words: twarog, acid curd cheese, herbs, spices, organoleptic evaluation

CHARACTERIZATION OF *STAPHYLOCOCCUS AUREUS* STRAINS ISOLATED FROM CHEESES IN HUNGARY

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Consumers expect from manufacturers to produce safe and high quality products, which the manufacturers are trying to achieve by setting up and running strict quality systems. Nonetheless, there may be problems with the presence of *Staphylococcus aureus* and other microbes, for example during cheese production, and later handling of the products. Large quantities of *S. aureus* in food may present a public health hazard due to the pathogenicity of some strains.

The aim of this study was to investigate the occurrence of *S. aureus* in 8 commercially available fresh cheese in Hungary, and after identifying the *S. aureus* strains isolated from the samples, the characteristics (tellurite reducing ability, lecithinase activity, hemolysin production ability) and the antibiotic resistance of isolates were determined.

Based on the results, *S. aureus* occurred in all cheese; the mean *S. aureus* counts ranged between 1.7 and 5.1 log₁₀ CFU/g. In all cheese samples, the *S. aureus* count exceeded the limit (m=1.0 log₁₀ CFU/g) set in the regulation of the Hungarian Ministry of Agriculture and Regional Development and the Hungarian Ministry of Health, Social and Family Affairs 1/2003 (I. 8). A total of 8 *S. aureus* strains were isolated from cheese samples. All strains produced clumping factor, had tellurite reducing ability, and three strains had lecithinase activity. Among the 8 strains, 1 strain was nonhemolytic, 5 strains showed weak hemolysis, and 2 strains showed α and weak γ hemolysis on blood agar. In antibiotic resistance testing, it was found that all strains were susceptible to chloramphenicol and erythromycin, 87.5% of the strains were resistant to penicillin, 75.0% were resistant to cefoxitin, gentamicin, and trimethoprim/sulphamethoxazole, 62.5% were resistant to clindamycin, and 37.5% were resistant to tetracycline.

Based on our results, we can observe that a significant amount of *S. aureus* can occur in fresh cheeses. The contamination of the cheeses with the bacterium can occur from the raw material, or during producing the cheeses, or when handling finished products. Our results also corroborate other findings in literature, which show that in recent years the proportion of occurrence of resistant *S. aureus* strains is rising continuously in the world.

Acknowledge: The work/publication is supported by the EFOP 3.6.1-16-2016-00022 project. The project is co-financed by the European Union and the European Social Fund.

Key words: microbiology, cheese, *Staphylococcus aureus*, antibiotic resistance

TRANSFER COEFFICIENT OF MINERALS FROM RAW PROPOLIS TO THEIR TINCTURES

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Propolis tincture is usually the ethanolic extract of raw propolis, which has several beneficial effects to the human health. The organic constituent of the tinctures has been extensively studied, however the mineral content has not been analysed well. Therefore we analysed their mineral content of 27 raw propolis of Hungary collected in 2014 and their ethanolic extracts. The extracts were done by weight 0.50 g of raw propolis and add 5 ml of 80% (v/v) ethanol, and extract for a week. The raw propolis and the tinctures were prepared by microwave digestion with HNO₃ and H₂O₂ in quartz vessels. Mineral content was analysed by inductively coupled plasma optical emission spectrometry (ICP-OES) and inductively coupled plasma mass spectrometry (ICP-MS).

The transfer coefficient percent (TC%) was evaluated and it was found that the K has the highest average TC% with 61.8%. Other elements in order of decreasing TC% were Na, Cu, P, B, Co, Mg, Zn, S, Mn, Mo and Cr with 55.3, 46.3, 43.8, 43.1, 33.8, 31.8, 19.4, 18.8, 17.8, 16.4 and 11.5%, respectively. The TC% of the other elements were below 10%.

Significant linear correlation was found in the case of B, Cu, Fe, K, Na, P, S, V, Cr, Co, Ni, Mo ($p < 0.01$) and in the case of Mn and Cd ($p < 0.05$) between the raw propolis and their tinctures.

According to the results, it was obvious, that the minerals have different transfer coefficient percent from raw propolis, to their tinctures. Some elements have significant Pearson correlation coefficient between raw propolis and their tinctures.

Acknowledge:  The study was supported by the New National Excellence Program of the Ministry of Human Capacities. We are also grateful to the National Beekeepers' Association of Hungary and the beekeepers for collecting the raw propolis samples.

Key words: propolis, tincture, minerals, transfer coefficient

THE HYGIENICAL STATUS OF MILK AND ENVIRONMENTAL SAMPLES DERIVED FROM DIFFERENT SHEEP BREEDS

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Sheep milk has been used by man since the beginning of sheep domestication due to its high nutritive value, and its health potential. However, the consumption of raw milk is an actual microbiological risk to consumers due to the possible presence of undesirable food spoilage or pathogenic bacteria. The aim of this study was to examine the hygienical and microbiological status of milk and environmental samples derived from different sheep breeds. Total Plate Counts (TPC), *Enterobacteriaceae* count (EBC), *Staphylococcus aureus* count (SAC) and coagulase-negative staphylococci count (CNSC) in ewe raw milk and environmental samples (udder surface) were examined. During the experiment 24 milk samples and 24 environmental samples (udder surface) were examined from three sheep breeds (Dorper, Merino and Cigaja) at different times. The sampling, the preparation of samples and decimal dilutions were based on international standards and methods. Our results revealed that, the mean of TPC were 3.25, 3.60 and 3.81 log CFU/ml, and CNSC were 3.08, 3.62 and 3.08 log CFU/ml in milk samples from Dorper, Merino and Cigaja, respectively. Both EBC and SAC were less than 10 CFU/ml. Statistically there was no significance difference between milk samples taken from different ewe breeds. The mean TPC of udder surface samples were 3.46, 3.75, 4.04 log CFU/cm²; CNSC were 3.76, 3.95 and 3.91 log CFU/cm² and EBC were 1.71, 1.38 and 2.14 log CFU/cm² from Dorper, Merino and Cigaja breeds, respectively. There is significance difference between udder surface samples taken from different breeds for TPC, and no difference for CNSC and EBC. The result indicated that almost all values with higher udder surface microbial resulted in higher milk samples microbial number. Therefore, the environment of the sheep farm may represent an important reservoir of these microorganisms.

Acknowledge: The study was supported by the EFOP-3.6.3-VEKOP-16-2017-00008 project. The project is co-financed by the European Union and the European Social Fund.

Key words: Sheep breeds, raw milk, hygiene, microbiology

**FUNGAL CHARACTERIZATION USING MATRIX-ASSISTED LASER DESORPTION
TIME-OF-FLIGHT MASS SPECTROMETRY (MALDI-TOF MS)**

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PCR based methods are now the most current and most reliable process to identify fungi, although these procedures are time-consuming and expensive. During the last decades, matrix-assisted laser desorption time-of-flight mass spectrometry (MALDI-TOF MS) has been considered an accurate and reproducible tool for identification microorganisms. These analytical instruments measure the size of the proteins of fungi, which results using appropriate techniques, are shown species-specific spectra.

Our study aimed to examine DNA-based identified filamentous fungi, isolated from grapevine, walnut and apple tree by MALDI-TOF MS to extend the database, which we could apply to identify microorganisms in the future. We examined isolates belonging to *Alternaria*, *Botrytis*, *Didymella*, *Leptosphaerulina*, *Epicoccum*, *Diplodia*, *Lasiodiplodia*, *Trichoderma* genera, which were subjected to both molecular identification ITS gene sequencing and MALDI-TOF analysis.

The most effective protocol was to be found first. Two extraction methods were tried in which formic acid or trifluoroacetic acid was used with acetonitrile, and combined with sonication and MagNA Lyser cell disruption. Moreover, two different matrices (α -cyano-4-hydroxycinnamic acid and sinapinic acid) were applied.

The most effective disruption was performed with formic acid and acetonitrile (1:1), as well as the most significant protein profiles were obtained with α -cyano-4-hydroxycinnamic acid. Other treatments did not increase the efficiency of the extraction, therefore they were not considered to be necessary.

During the measurement, we have found, that the isolates have shown individually distinctive species-specific spectra, consequently this method is appropriate augmenting the database of filamentous fungi. We have also observed, there was a reduced set of peaks in the case of older mycelia, so it is advisable analysing younger fungal cultures.

The result of this extensive optimization opens the possibility to generate a widespread database, which offers a promising tool for an extremely rapid and reliable identification of plant pathogen and mutual endophytic fungi.

Acknowledge: We thank the WESSLING Hungary Kft. for the cooperation.

Key words: maldi-tof, endophytic fungi, identification

Name of publication

Scientific researches in food production, University of Debrecen - Proceedings of abstracts

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Publisher

Debreceni Egyetem, Mezőgazdaság-, Élelmiszertudományi és Környezetgazdálkodási Kar

Year of publication

2018

Pages

30

ISBN

978-963-490-041-2