



Book of Abstracts

Workshop on
Bioactive compounds in berry fruits: genetic control, breeding, cultivar, analytical aspects and human health

Zurich – Switzerland
3rd - 5th December 2008

Organised by Agroscope Changins-Wädenswil ACW and the COST-Action 863 Euroberry Research: from Genomics to Sustainable Production, Quality and Health"

and supported by
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Organising committee

Béatrice Dénoyes-Rothan, UREFV–INRA, France	WG1 Leader
Margit Laimer, Universität für Bodenkultur Wien, Austria	WG4 Leader
Bruno Mezzetti, Marche Polytechnic University, Ancona, Italy	COST863 Chairman
Christoph Carlen, Agroscope Changins-Wädenswil ACW, CH	Local organiser

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LOCATION OF EVENTS

Registration – information	
Wednesday, 3th December 13.00 to 14.00	Building ML, in front of the room F34
Plenary sessions	
Wednesday, 3th December 14.00 to 18.00 Thursday, 4th December 09.00 -13.00 14.30 – 18.00 Friday, 5th December 09.00 to 12.00	Building ML, room F34 Building CLA, room J1 Building CLA, room J1
Breaks	
Wednesday, 3th December Thursday 4th and Friday 5th December	Building ML, in front of the room F34 Building CLA, room J2+J3
Posters sessions	
Thursday 4th and Friday 5th December	Building CLA, room J2+J3

Lunch
Mensa - Polyterrasse

Social dinner – Wednesday.3th December, 20.00
Restaurant
Wirtschaft Neumarkt
Neumarkt 5
8001 Zürich
phone : 044 252 79 39

PROGRAM

Wednesday, December 3rd

Opening of the workshop and welcome

14.00 **C. Carlen,** local organiser
B. Mezzetti, Chairman of the COST Action 863
J.F. Hausman, Action rapporter
A. Maggio COST Scientific Officer

Session 1 - How do genetics and genomics contribute to a better knowledge on bioactive compound in berry fruits ?

Moderation: **Christoph Carlen** , Agroscope Changins-Wädenswil ACW, Switzerland

14:20 Strawberry genomics new impacts in basic biology and crop science
Invited speaker : K.M. Folta, T.M. Davis
Horticultural Sciences Department, University of Florida, Gainesville, FL USA

14:55 Polyploidy and its consequence on QTL detection on fruit quality compounds
B. Denoyes-Rothan, E. Lerceteau-Köhler, M. Rousseau-Gueutin, A. Moing, C. Renaud

15.20 Unraveling the regulation of flavonoid metabolism in strawberry fruits
C. Rosati, R.C.H. De Vos, G. Perrotta, A. Bovy, S. Martens

15:45 The comparison of wild growing and cultivated red raspberries by molecular and biochemical markers in Turkey
S. Ercisli

16:10 Break

Session 2 - Does variability exist in germplasm or collections and how can this variability be used for breeding programmes? (I)

Moderation: **Christoph Carlen** , Agroscope Changins-Wädenswil ACW, Switzerland

16:30 Investigating genetic diversity, nutritional quality and bioactive compounds of berry species collections grown in Russia
J.F. Hausman, D. Lamoureux, I. Lefèvre, T. Gavrilenko, S. Alexanian, P. Eyzaguire.

16.55 Breeding strawberry (Fragaria x Ananassa Duch) to increase fruit nutritional quality
J. Diamanti, F. Capocasa, S. Tulipani, M. Battino, B. Mezzetti

17:20 Wild growing berry fruits - valuable source of genetic variability in Republic of Croatia
B. Duralija, Z. Šindrak, S. Voća, D. Dujmović Purgar, A. Mešić, A.Vokurka

17:45 New blackcurrant breeding program for increasing consumption of fresh fruit with high level of bioactive compounds
S. Pluta and E. Żurawicz

18.10 End of the first day

20.00 Social dinner at the Restaurant „Wirtschaft Neumarkt“, Neumarkt 5, 8001 Zürich

Thursday, December 4th

09.00 Welcome and short overview of Agroscope Changins-Wädenswil Research Station ACW
Lukas Bertschinger, Agroscope Changins-Wädenswil ACW, Research Director, Switzerland

Session 2 - Does variability exist in germplasm or collections and how can this variability be used for breeding programmes? (II)
Moderation: Béatrice Denoyes-Rothan, INRA-Bordeaux, France

09:20 Phenotyping fruit nutritional quality parameters in the INOTALIS collection of strawberry genotypes
A. Monfort, M.J. Aranzana, D. Sánchez, M.A. Hidalgo and P. Arús

09:45 Comparison of antioxidant capacity and chemical properties of wild and cultivated red raspberries (Rubus ideaus L.)
Ç. Çekic, M. Özgen

10:10 Portuguese endemic wild blackberries as an alternative source of polyphenols and antioxidant activity
L.R. Tavares, C.N. Santos, G.J. McDougall, D. Stewart, R. B. Ferreira

10:35 Quality fruit traits and phenolic compounds characterisation of blueberry cultivars in three different cultural areas
C. Andreotti, M. Castagnoli, M.L. Maltoni, W. Faedi

11:00 Break

11:20 Antioxidant compounds in Ribes SPP. cultivars grown in Piemonte (Italy)
M. Cavanna M., G. L. Beccaro, G. Bounous

11:45 The effect of genotype and maturity at harvest on blackcurrant (Ribes nigrum L.) bioactives
J. Giné Bordonaba, L.A. Terry

12:10 Health valuable compounds in blackurrants evaluated at the research institute of pomology and floriculture (RIPF), Skierniewice, Poland
J. Markowski J., S. Pluta, M. Mieszczakowska, E. Żurawicz

12:35 Determination of the variance component of antioxidant capacity and chemical properties of strawberry (Fragaria x Ananassa)
K. Gündüz, S. Serçe, E. Özdermiri, S. Payadaş, M. Özgen

13:00 – 14.30 Lunch and Poster Session

Thursday, December 4th

Session 3 - Analysis to measure bioactive compounds in berry fruits and their agronomic improvement

Moderation: Mezzetti Bruno, SAPROV - Marche Polytechnic University, Ancona Italy

14.30 Development of high throughput analyses of polyphenol composition in berries using abbreviated mass spectrometry techniques
G. McDougall, I. Martinussen, D. Stewart

14.55 Automation of analytical methods to screen antioxidant capacity of biological matrices
J. Héritier, W. Andlauer

15.20 Effect of preharvest factors on strawberry (Fragaria x ananassa) bioactives
L.A. Terry, J. Giné Bordonaba

15.45 Understanding health-promoting bioactive compounds in blackcurrants and their agronomic improvement
R.O. Karjalainen**,** D. Stewart, G. McDougall, H. Hilz, M. Anttonen, N. Saviranta, P. Mattila, R. Törrönen

16:10 Break and Poster session

16:30 BTH induces protection against downy mildew in arctic bramble (Rubus arcticus) and the accumulation of several phenolics
H.I. Kokko, A.T. Hukkanen, K.H. Kostamo, S.O. Kärenlampi

16:55 Quality of strawberry cv. Elsanta after hot water dips of different exposures
T. Jemrić, V. Dragović Uzelac, S. Voća, D. Bursać Kovačević, B. Duralija

Section 4 - How may bioactive compounds benefit human health? (I)

Moderation: Margit Laimer, Universität für Bodenkultur Wien, Austria

17:20 Berryfruit for health research at HortResearch, New Zealand
Invited speaker : Dr R. Hurst, Leader of the Healthy Berry Program, HortResearch, Auckland, New Zealand

18:05 End of the second day

Friday, December 5th

Section 4 - How may bioactive compounds benefit human health? (II)
Moderation: Margit Laimer, Universität für Bodenkultur Wien, Austria

09:00 Polyphenols from berries : metabolism and bioavailability
Invited speaker : Prof. Dr. Gary Williamson, Functional Food, Department of Food Science, University of Leeds, England

09:35 Antioxydant capacity of portuguese endemic Rubus fruits in a neurodegeneration cell model
C. Santos, L. Tavares, V. Pontes, P. Alves, R. Ferreira

10:00 Strawberry consumption and antioxidant status in human subjects
S. Tulipani, S. Romandini, J.M. Alvarez Suarez, J. Diamant, F. Capocasa, B. Mezzetti, M. Battino

10:25 Break

10.45 Final discussion (B. Denoyes-Rothan, M. Laimer, C. Carlen)
Further research requirements and open questions related to genetics, cultivars, breeding and bioactive compounds as well as related to measurement of bioactive compounds and their benefit to human health

12.00 End of the Joint Meeting

POSTERS

I. Badjakov, R. Gevrenova, M. Niklova, V. Kondakova, E. Todorovska, A.Atanassov0	Agro Bio Institute	Bulgaria	Bioactive constituents and DNA profiling of raspberry germplasm collection in Bulgaria.
P. Crespo, A. Blanc, A. Ançay, D. Baumgartner, P. Stamp, C. Carlen	Agroscope Changins-Wädenswil ACW	Switzerland	Variation in quality traits and antioxidant capacity of ten strawberry cultivars
S. Ercisli	Ataturk University, Agricultural Faculty	Turkey	Preliminary characterisation of cornelian cherry genotypes for their physico-chemical properties.
G. Marzban, R. Nestby, A. Herndl, F. Maghuly, M. Laimer	Plant Biotechnology Unit, IAM, BOKU	Austria	Vacccinium mytrillis and bioactive compounds
S. Magnani, G. Baruzzi, L. F. D'Antuono, M. L. Maltoni, M. Ranieri,W. Faedi (C. Andreotti bring the poster)	CRA-Unità di Recerca per la Fritticoltura	Italy	Health-promoting components in old and new Italian strawberry varieties
A. Masny, E.Zurawicz, J. Markowski	Research Institute of Pomology ad Floriculture	Poland	Anthocyanins and ascorbic acid - important quality traits in strawberry breeding program at ripf skierniewice, Poland
E. Oprea, V. Rădulescu, C. Balotescu, V. Lazar, M. Bucur, P. Mladin, I. Farcasanu	University of Bucharest, Faculty of Chemistry	Romania	Essential oils form Ribes nigrum buds: GC/MS analysis and antimicrobial activities
D. Seglina,. Kampuse, I. Krasnova, G. Heideman, L. Dukalska	Latvia State Institute of Fruit Growing, Dobele	Latvia	The effect of packaging materials and technologies to the storage time of fresh black currants

ABSTRACTS

ORAL

PRESENTATIONS

STRAWBERRY GENOMICS- NEW IMPACTS IN BASIC BIOLOGY AND CROP SCIENCE

K.M. FOLTA, T.M. DAVIS

Horticultural Sciences Department, University of Florida, Gainesville, FL USA
Plant Biology Department, University of New Hampshire, Durham, NH USA

Functional Genomics, Orphan Genes, Next-gen Sequencing, Gene function

The implementation of new tools and technologies has accelerated strawberry genomics studies over the last five years. Recent goals in our laboratory target analysis of structural, functional and translational aspects of strawberry genomics. These activities enable further understanding of flavor and nutriceutical formation in strawberries, and the rapid transformation and regeneration systems make the strawberry a good representative species for functional genomics study of fruit and plant attributes. Analysis of cultivated strawberry gene and genome structure reveal that the haploid strawberry genome is remarkably small, containing characterizable repetitive regions and gene structure/spacing similiar to arabidopsis. Colinearity with other major genomes is observed, and the subgenome constituents of the octoploid genome are becoming move clear. Functional genomics efforts have targeted the strawberry "unknown-ome", attempting to identify gene function using our agile transgenic systems. We have identified novel regulators of root growth, phototropic behaviors, leaf development, and other processes. Surprisingly, some of these gene products not found in Arabidopsis thaliana have strong phenotypes when introduced to this heterologous system. Translation of photoperiodic flowering models to the crop system indicates that strawberry exhibits breaks from established models in many respects. Strawberry (*Fragaria spp*.) is a member of the Rosaceae Family, an economically important family of valued fruit, nut, ornamental and wood crops, so our rapid systems for in planta tests of gene function should translate well to these other systems. Thus, findings in the small, herbaceous, rapidly-growing plant may direct study in other crop species where such studies are not now possible.

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POLYPLOIDY AND ITS CONSEQUENCE ON QTL DETECTION ON FRUIT QUALITY COMPOUNDS

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Fragaria x ananassa, QTL, polyploidy, fruit quality, health compounds

The cultivated strawberry, *Fragaria* × *ananassa* (2n=8x=56), is an polyploid species affiliated to the Rosaceae family, which comprises important agronomical species.
Most of the traits relevant for strawberry breeding are under polygenic control, and this is partly due to its ploidy. Fruit soluble solids, sugars, titratable acidity, organic acids at maturity (Shaw et al. 1987; Shaw 1988) and ascorbic acid (Lundergan and Moore 1975; Sone et al. 2003) are quantitatively inherited. In order to have better knowledge of the inheritance and improve breeding efficiency for fruit quality traits, we developed a QTL approach.
Quantitative trait loci (QTL) were investigated using 213 full-sibling F1 progeny from a cross between the variety 'Capitola' and the genotype 'CF1116'. Progeny were evaluated for fruit components covering fruit development, colour, and the concentrations of sugars, organic acids, and ascorbic acid. QTLs were detected for all the quantitative traits analyzed, but they were mainly different according the year. This result revealed QTL instability across years, possibly due to methodological imprecision, slight differences in fruit maturity status and environmental influences. The phenotypical variation explained by each QTL was low to moderate, reflecting the high ploidy of this species. QTLs belonging to the same homoeology group, as reflected by the segregation of multiple and homoeologous locations, were detected, and in some cases their expressions were specific to the year of evaluation. These QTLs could be allelic forms of the same gene.

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UNRAVELING THE REGULATION OF FLAVONOID METABOLISM IN STRAWBERRY FRUITS

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Flavonoids, Fragaria, fruit development, gene and enzyme expression, genetic and environmental effects, metabolite analyses

Research program on strawberry at ENEA Trisaia has focused on the characterization of (poly)phenol and flavonoid genes and compounds in different genetic backgrounds, given the visual and nutraceutical properties that such compounds provide to the fruit and humans. Gene expression studies have characterized the spatial and developmental patterns of the expression of main structural genes of the pathways, in parallel to biochemical investigations on enzyme activity and main flavonoid metabolites. Comparative studies on genotypes differing in fruit flavonoid composition grown at different locations assessed the impact of genetic and environmental effects on the regulation of the flavonoid pathway in strawberry fruits. The information from such fundamental studies is being exploited for metabolic engineering, to improve the antioxidant capacity and flavonoid content of strawberries.

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THE COMPARISON OF WILD GROWING AND CULTIVATED RED RASPBERRIES BY MOLECULAR AND BIOCHEMICAL MARKERS IN TURKEY

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Raspberries, AFLP, antioxidants, FRAP, phenolics

The international tendency for growing and production of small fruits including raspberries shows a permanent increasing because these group fruits relatively has higher bioactive contents. Among raspberries, the most important is the European red raspberry, *R. idaeus* L. subsp. *idaeus* and the place of origin of red raspberry has been postulated to be the Ida Mountains of Turkey (Jennings 1988). In the present study eleven wild growing and one well known red raspberry cultivar, cv. Heritage were compared both molecular and biochemical way. The wild plants were selected before according to attractive fruit, high yield and free of pest and disease characteristics from Eastern Anatolia region of Turkey. For molecular characterization fluorescent-AFLP markers was used. A total 40 primer-enzyme combination was screened and among them 9 out of 40 was found the most effective to obtain higher polymorphism ratio. To study the health benefits of red raspberry fruits, eleven pre selected wild grown and one well known cultivar, Heritage were also evaluated for their some physico-chemical berry properties such as fruit weight, total antioxidant capacity, total phenolics, ascorbic acid, soluble solid content (SSC) and acidity. β-carotene bleaching and ferric reducing antioxidant power (FRAP) were used to determine total antioxidant capacity while Folin–Ciocalteu reagent used to determine total phenols. Fruit weight, SSC and ascorbic acid content of genotypes were between 1.61-2.18 g; 10.87-13.60 and 21-36 mg/100 g, respectively. Antioxidant activity and total phenolic content varied among genotypes and ERZ5 genotype had the highest antioxidant capacity in both methods. This genotype also had the highest total phenolic (2031 mgGAE/gDW) content. There are linear relationships between antioxidant capacities and total phenols. The present study demonstrates the potential of certain wild genotypes, notably ERZ5, for improvement of nutritional value through germplasm enhancement programmes.

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INVESTIGATING GENETIC DIVERSITY, NUTRITIONAL QUALITY AND BIOACTIVE COMPOUNDS OF BERRY SPECIES COLLECTIONS GROWN IN RUSSIA

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Fresh and processed products derived from small berries make important contributions to human nutrition and health, as well as offering economic opportunities for a decentralized and high value agricultural production. Berries in general are valuable sources of minerals, vitamins, dietary fibre, antioxidants and other nutrients, which make them an important commercial fruit crop. However, a wide diversity of phytochemical levels and antioxidant capacities exists within genotypes of these small fruits.
The N.I. Vavilov Institute of Plant Industry (VIR), one of the biggest and oldest germplasm collections worldwide, among others maintains raspberries, mountain ash, blackcurrant and honeysuckle. These collections are poorly characterised and there is a lack of information about genetic diversity and variability in terms of nutrient, micronutrient and non-nutrient phytochemicals with health functions of these berry species.
The project aims i) to investigate diversity at the DNA level in subsets of the above-mentioned species with the help of microsatellite markers, ii) to analyse their diversity in terms of biochemical compounds. Their nutritional quality will be evaluated by proximate analysis of carbohydrates, proteins and fibre. Samples will also be analysed for their total antioxidant capacity. Among all the compounds that are shown to have antioxidant potential, some will be evaluated specifically. As these berries are reported to contain significant amounts of polyphenol antioxidants such as anthocyanins, we will determine concentration of these compounds. Some of these berry fruits are known to contain high concentrations of vitamin C, thus concentration of this compound will also be assessed. In addition to these compounds, the composition and concentration of carotenoids will also be characterized.
Identification of berry accessions providing elevated concentrations of the compounds outlined can be of importance in the improvement of food quality in order to eliminate dietary deficiencies and reduce the risk of contracting chronic diseases. A major aim of the project is also to demonstrate the relevance of the conservation of plant germplasm collections as a source of valuable genetic diversity for future generations.

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BREEDING STRAWBERRY (*FRAGARIA X ANANASSA DUCH*) TO INCREASE FRUIT NUTRITIONAL QUALITY

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Fragaria, germplasm, seedlings, fruit quality, antioxidant capacity, phenols

Strawberry breeding programs are currently used to acquire new varieties improved for specific agronomic (Yield and Size), qualitative (Firmness, Sugars Content and Acidity) and sensorial (Colour and Aroma) characteristics, all combined to increased disease resistance and plant adaptability. Nowadays, to besides to all these parameters is necessary to look for the specific bioactive components well known for their effect on human health. This aspect is now highly requested by the consumer.

This study started with the evaluation of fruit nutritional quality of different varieties now available in the market and also recently released (Anita, Antea, Asia, Ciflorette, Darlisette, Daroyal, Queen Elisa, Roxana, Irma, Dora, Adria, Sveva, Onda, Clery). For all genotypes were evaluated for their standard parameters of productivity and fruit quality (°Brix, Total Acidity, Color, Firmness, fruit size and total yield) and also of nutritional value (Total Antioxidant Capacity (TEAC) and Total Phenols Content (TPH). The results are showing a wide difference among varieties for the production and fruit quality, mostly related to their adaptability to the local evaluation conditions. Regarding their nutritional values, only few varieties differed for their higher phenol content and total antioxidant capacity.

By considering these results a breeding program has been started by considering two approaches specifically addressed to increase nutritional value of fruits. A first approach was based on the use of inter-specific crosses within *F. virginiana glauca* (FVG) and different *F. x ananassa* cultivars. After a selection work were identified some selections showing a very high increase of fruit bioactive compounds, but keeping most of FVG fruit phenotype. AN94,414,52 (DON x FVG22) resulted the most interesting selection because combining the nutritional parameters with already acceptable production and quality values. Backcrossing this selection with other *F. x ananassa* varieties and selections were obtained new seedlings showing a better combination of plant yield and fruit nutritional quality.

The second approach was a breeding program based on cross combinations with *F. x ananassa* varieties or selections already identified for their high nutritional values. Also from this work was obtained new material performing with increased content of bioactive compounds already combined with high standard values of plant yield and fruit standards. These results were achieved mostly from the progenies derived from the following cross combinations: (AN04,147 (Darselect x Irma), AN05,32 (00,92,4 x Onda, AN05,48 (Roxana x Candonga), AN05,68 (CN00,142,1 x Irma), AN05,75 (Irma x Dora), AN05,77 (Record x Dora).

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WILD GROWING BERRY FRUITS - VALUABLE SOURCE OF GENETIC VARIABILITY IN REPUBLIC OF CROATIA

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Fruit quality, biodiversity, phenolic compounds, vitamin C, berry fruits

Wild growing fruits traditionally make a certain part of population diet in the Republic of Croatia. These fruits were found to be a rich source of bioactive compounds with considerable dieto-therapeutic impact on human health.
During the winter wild fruits is used as processing (dried or preserved with sugar). Otherwise those used to be eaten fresh during ripening.
The most important species of wild growing fruits suitable for utilization belong to several genera: *Rubus*, *Prunus*, *Fragaria*, *Castanea*, *Vaccinium*, *Sambucus*, *Rosa* and others.
Almost all species of mentioned genera are potentially interesting for use in breeding programs with the goal of improvement of nutritious and sensory properties (aroma and flavour) of fruits varieties – their cultivated relatives.
This research includes survey of the most important wild growing fruit species in Croatia. Also, chemical parameters (dry matter, vitamin C, anthocyanins, and phenolic compounds content), of fruits quality are presented. It was found out that wild growing fruits have a considerable higher content of the measured chemical parameters by comparison with related, but cultivated fruit species.

All of these facts indicate that wild growing fruit species, used in diet since ancient times, will have greater significance in a future scientific research.

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NEW BLACKURRANT BREEDING PROGRAM FOR INCREASING CONSUMPTION OF FRESH FRUIT WITH HIGH LEVEL OF BIOACTIVE COMPOUNDS

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Blackcurrant, Ribes nigrum L. breeding, cultivars, dessert fruit.

The Research Institute of Pomology and Floriculture (RIPF) in Skierniewice, Poland is the main centre of top and small fruits breeding in Poland. Activities in this area include genetic and methodological studies as well as development of new cultivars. Three main breeding directions are taken into consideration: breeding for resistance, breeding for fruit quality and breeding for good adaptation to the local climate conditions. The usefulness of newly released cultivars for mechanical fruit harvest is also important for the technology of blackcurrant cultivation in our country.

Since 1986, at the Plant Breeding Department of the RIPF in Skierniewice the blackcurrant breeding program has been underway aimed at obtaining new cultivars suitable for the growing technology with collecting fruits by different types of harvesters. The new cultivars should have a high productive value, with their fruit being used for the processing and freezing industries. The work has resulted in obtaining five cultivars: 'Tiben', 'Tisel', 'Ores', 'Ruben' and 'Tines'.

The new breeding program aimed at releasing of dessert-type blackcurrants was started in 1997. In the first stage the blackcurrant genetic resources were phenotypicaly evaluated and screened for the traits important for the dessert-type fruit quality. The best cultivars were selected as parental forms ('Bona', 'Ben Sarek', 'Czereszniewa', 'Lentaj', 'Storklas' and 'Big Ben') and their breeding value was assessed by using the general combining ability (GCA). Studies showed that the highest breeding value (positive and significant GCA effects) had cultivars 'Big Ben' and 'Storklas'. Both genotypes were the most promising parents in the breeding program aimed at developing dessert-type cultivars in Polish conditions.

Further studies on phenotypic evaluation of genotypes maintained in the field collection and cultivar trails were carried out in 2002-2007. The results showed that few new Polish cultivars and breeding selections as well as several foreign genotypes were productive and produced very large and attractive fruits of good taste and were resistant to fungal diseases. They were Polish genotypes ('Bona', 'Gofert', 'Tines', 'Tisel', PC-425, D13B/11.), Lithuanian ('Gagatiai'), Scottish ('Big Ben'), Russian ('Dlinnokistnaja' and 'Lentaj') and Ukrainian ('Czereszniewa' and Sofijewskaja'). They were used as potential parental forms in the second crossing program done in spring 2008. These genotypes were crossed in the factorial design using 3 male forms ('Ceres', 'Foxendown' and 'Sanjuta'). Obtained F_1 full-sib family progenies will be assessed individually for 3-4 years in terms of the traits determining dessert-type fruit quality. The GCA and SCA of parental forms will be calculated and the best crosses will be repeated in larger scale to obtain bigger population of seedlings producing the best dessert-type blackcurrant fruit.

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PHENOTYPING FRUIT NUTRITIONAL QUALITY PARAMETERS IN THE INOTALIS COLLECTION OF STRAWBERRY GENOTYPES

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Fragaria, Sugars, Polyphenols, Total Antioxidant Capacity, Genetic Association

Phenotyping a collection of strawberry genotypes for the main characters controlling the strawberry fruit nutritional quality (mainly total sugars, total acidity, total antioxidant capacity, polyphenols and vitamin C) is a fist step towards improving the nutritional quality of new cultivars. All these characters are in general under a quantitative genetic control, even if occasionally some can also be qualitatively inherited. Many of these characters are also related to the fruit ripening process. This study was carried out over 70 strawberry cultivars and breeding lines of the INOTALIS collection.
Nutritional parameters analyzed were: Total Antioxidant Capacity (TAC) measured using ferric reducing/antioxidant power method (FRAP), Polyphenol content measured by the Folin-Ciocalteu (FC) method, sugar content measured by HPLC and determining content on Fructose, Sucrose and Glucose, vitamine C by Ascorbic Acid spectrophotometer analysis and Acidity as acid/base value.
Other important fruit quality characters such as weight, shape, colour, firmness and Brix were also analyzed in fruits collected at specific ripening stages. These parameters showed different levels of variability, indicating that they are genetically controlled, and can potentially be incorporated in new cultivars with enhanced organoleptic and nutritional quality.
Parameters as CAT, Sugars or polyphenols showed different levels of variability, indicating the potential existence of genes that can be incorporated in cultivars with enhanced organoleptic and nutritional quality. This analysis permits to compare new lines to reference commercial varieties.
Genetic association was detected between some parameters and markers. Marker-trait associations identified markers adequate for marker assisted selection for Antioxidant Capacity, Polyphenols or Sugar content.

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COMPARISON OF ANTIOXIDANT CAPACITY AND CHEMICAL PROPERTIES OF WILD AND CULTIVATED RED RASPBERRIES (RUBUS IDEAUS L.)

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Anthocyanins, FRAP, TEAC, total phenolic, wild

We investigated some of the chemical properties and the antioxidant capacity of 14 wild red raspberry (Rubus ideaus L.) genotypes selected from Northern Turkey between the altitudes 1123-2043 m. In addition, 'Heritage' and 'Tulameen' cultivars were included in the study to determine the variation of antioxidant capacity and chemical properties between wild and cultivated raspberries. Fruit total phenolic (TP), total monomeric anthocyanins (TMA), soluble solids (TSS), individual organic acids and sugars were examined. The antioxidant capacities of fruits were determined by both FRAP (ferric reducing antioxidant power) and TEAC (trolox equivalent antioxidant capacity) assays. The fruit color and weight were also recorded.

All of the parameters tested were found to be significantly different among genotypes. With the help of principle component (PC) analysis, the variation among these genotypes and cultivars could be divided into three groups; A2, A9, A12, A14 formed the first group with high phytonutrient properties, while the cultivars 'Heritage' and 'Tulameen' grouped together with high phytonutrients but low color values; the rest of the genotypes formed the final group (III). The first three PCs explained 30, 25 and 16% of the variation, for a total of 71%. Among the variables tested, the phytonutrient-related parameters (TP, TACY, FRAP and TEAC) were highly correlated with PC1. The antioxidant capacity among the samples averaged 14.6 and 14.1 μmol TE/g fresh weight (fw) by the FRAP and TEAC methods, respectively. A2 displayed the highest antioxidant activity, TP and TMA content. TP means averaged 2047 mg GAE/g fw and TMA averaged 205 μg cy-3-soph/g fw. Variability among samples was C.V. 24% and 25% for TP and TMA, respectively. The major sugars in the samples were fructose (32.2 g/kg) and glucose (24.3 g/kg), and the predominant organic acid was citric acid (13.1 g/kg).

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PORTUGUESE ENDEMIC WILD BLACKBERRIES AS AN ALTERNATIVE SOURCE OF POLYPHENOLS AND ANTIOXIDANT ACTIVITY

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Wild blackberries, Rubus sp., polyphenols, antioxidant activity, HPLC-MS

Blackberries are a well known source of polyphenols with high antioxidant activity that may provide human health benefits. *Rubus* is a genera with high diversity and globally distributed (Deighton et al., 2000). In the Northeast of Portugal it has some endemic species of Rubus (wild blackberries) that could have an interesting antioxidant activity and could constitute a good source of fruits with high polyphenolic content. The aim of this study was to determine the potential of Portuguese *Rubus* endemic species as a source of phenolic compounds as a way to preserve these species.

For three *Rubus* species fruits (*R. brigantinus, R. genevieri and R. vigoi*) and a commercial blackberry cv. Apache (*Rubus rubus*) it was determined the polyphenol content (Singleton and Rossi, 1965), anthocyanin content (Deighton et al., 2000) and antioxidant activity (Cao et al., 1993, Michalska et al., 2007). It was determined the HPLC-DAD-ESI-MS profile for each species in order to identify the differences between them.

Although two endemic species (*R. brigantinus* and *R. vigoi*) have higher amount of total polyphenols than commercial blackberry cultivar, their antioxidant activity and anthocyanin content are similar to the commercial species. The total polyphenols, antioxidant activity and anthocyanins content in *R. genevieri* are lower than in the commercial blackberry. Comparing the HPLC profiles, the endemic species contain a higher amount of sanguiin H6, lambertianin C. Some *in vitro* studies mention the ellagitannins possessing anti-proliferative activity (Ross et al., 2007, Mertens-Talcott et al., 2003, Rao et al., 1991) and vasodilatory properties (Mullen et al., 2002). In the case of *R. vigoi* it has also a higher amount of quercetin derivatives that are described in bibliography as presenting some important properties such as anti-inflamatory (Read, 1995, Orsolic et al., 2004), anti-fibrotic (Lee et al., 2003), anti-coagulative (Bucki et al., 2003), anti-proliferative (Orsolic et al., 2004), anti-bacterial (Cushnie and Lamb, 2005), anti-atherogenic (Perez-Vizcaino et al., 2006) and anti-hypertensive (Perez-Vizcaino et al., 2006, Duarte et al., 2001). The use of species with higher contents in ellagitanins and quercetin derivatives such as *R. brigantinus* and *R. vigoi* could be promising in the achievement of healthier fruits for direct consumption or for addition in nutraceuticals.

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QUALITY FRUIT TRAITS AND PHENOLIC COMPOUNDS CHARACTERISATION OF BLUEBERRY CULTIVARS IN THREE DIFFERENT CULTURAL AREAS

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Vaccinium corymbosum ,variety, flavonoids, anthocyanins, cultivation systems

The Italian production of blueberry (*Vaccinium corymbosum*) is constantly increasing during the last decade and presently it is approximately of 1.000 tons. The most important regions for blueberry are Piemonte and Trentino-Alto Adige, representing more than 70% of the whole national production. In Italy the most common cultivars are characterized by high chilling unit requirement and generally they are interspecific hybrid of *V. corymbosum* with other *Vaccinium species*.
Our research investigated fruits from six blueberry cultivars: 'Blueray', 'Bluetta', 'Berkeley', 'Bluecrop', 'Toro', 'Brigitta'. Blueberry fruit samples of each cultivar were harvested from variety trials located in three different areas of Italy: North-West (Cuneo, Piemonte), North-East (Trento, Trentino-Alto Adige) and Central-East (Cesena, Emilia-Romagna). Blueberry plants from the Cuneo and Trento collections were grown in open field whereas, in Cesena, plants were grown with a soilless system (in pots). Comparison among cultivars was therefore performed within three different environmental and cultural conditions that could be considered as representative of three important small fruit production districts in Italy.
Fully ripened berries were evaluated for their biometrical characteristics (fruit weight, size and shape), for their main quality indices (soluble solids, titratable acidity, sugar/acid ratio, skin color) and phenolic characterization. The aim was to evaluate differences induced by genotype and cultural environment on these fruit quality parameters and on phenolic compounds, that are considered as the main antioxidant metabolites of fruits and therefore are responsible for the high nutraceutical value of this food. Phenolic compounds were extracted and analyzed following the methodology reported in Andreotti et al. 2008, adapted for blueberry samples. Main classes of phenolic compounds (simple phenols, hydroxycinnamic acids, flavan-3-ols, flavonols and anthocyanins) were evaluated and the concentrations were compared among all cultivars.
Preliminary results show relevant differences among berries of cultivars grown in the same cultural conditions. The more interesting genotypes as for overall quality of fruits at harvest, are finally discussed in relation to the characteristics of the three districts of production tested in this research.

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ANTIOXIDANT COMPOUNDS IN *RIBES* SPP. CULTIVARS GROWN IN PIEMONTE (ITALY)

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Key words: currants, FRAP, anthocyanins, polyphenols, HPLC

Berries with their intense black-purple and red colour have high phenolic concentrations and are also rich in vitamin C, a rich source of antioxidants (Moyer *et al*., 2002). In recent years the plant phenolics have received much attention due to their antioxidant, antimutagenic, anticarcinogenic, anti-inflammatory, antimicrobial, and other biological properties related to benefits on human health (Morton *et al*., 2000). The aim of this research was to determine the range of total anthocyanin content (ACY), total phenolic content (TPH) and antioxidant capacity in 29 cultivars of *Ribes* spp. (*Ribes nigrum*, *R. rubrum*, *R. grossularia,*) germplasm, including 2 hybrids of blackcurrant and goosberry (*Ribes* x *nidigrolaria*), using spectrophotometric analysis. Moreover the 29 samples were analysed by high-performance liquid chromatography (HPLC) with diode array detection (DAD). This technique, coupled with mass spectrometry analysis, allows to quantify and identify the different anthocyanins in the samples, being one of the most abundant class of phenolic compounds present in the genus *Ribes*. The berry samples were collected at ripeness during the first two weeks of July 2006, according to the maturity stage of the cultivar. ACY was determined by the pH differential method. TPH was determined by Folin-Ciocalteau method. Antioxidant capacity was measured by FRAP (*ferric reducing antioxidant power*) assay. The antioxidant capacity ranged from 37.09 (cv Werdavia) to 107.1 mmol Fe^{2+} equivalents/ Kg fruit (cv Black Down). TPH ranged from 22.64 (cv Rokula) to 560.48 mg gallic acid equivalents/100 g (cv Black Down). In two white mutations of *Ribes rubrum* anthocyanin pigments were not found. The highest content of anthocyanins was detected in the cultivar of *Ribes nigrum* Geant de Boskoop (373.75 mg cyanidin-3-glucoside equivalents/100 g fruit). In general blackcurrant cultivars showed the highest values for the three analyses. The most abundant anthocyanins identified and quantified with HPLC analysis in these cultivars of *Ribes nigrum* were delphinidin 3-rutinoside and cyanidin 3-rutinoside with a mean of 133.60 and 113.14 mg/100 g respectively, whereas the most abundant anthocyanin found in *Ribes rubrum* cultivars was cyanidin 3-xylosylrutinoside with an average of 21.80 mg/100 g. Finally in the cultivar Rokula (*Ribes grossularia*) and in the two hybrids analysed (Jogranda and Jostina), the most abundant anthocyanin was cyanidin 3-rutinoside, with values of 5.74, 22.29 and 10.83 mg/100 g respectively.
In conclusion, the presence of a higher content in phenolic compounds and antioxidant capacity, especially in *Ribes nigrum* (representing, therefore, an important resource of antioxidant compounds) was confirmed. The differences identified may have important implications on the studies related to human health. These cultivars represent a valuable resource of nutraceutics, because the quantiy phytochemicals is genetically related. The manipulation (by means of traditional breeding methods or advanced biotechnologichal or genetic manipulation) (Tucker *et al*., 2002; Dharmapuri *et al*., 2002) could be powerful tool to modify antioxidant fruit content.

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THE EFFECT OF GENOTYPE AND MATURITY AT HARVEST ON BLACKCURRANT (*Ribes nigrum L.*) BIOACTIVES

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Antioxidant activity, anthocyanins, ascorbic acid, total phenolics,

Blackcurrant (*Ribes nigrum* L.) berries are a rich source of antioxidants, due in part to their high content of anthocyanins and ascorbic acid. Several studies have demonstrated the health-related benefits associated with their consumption. Thus said, it is known that processing blackcurrant fruits can dramatically reduce the content and bioavailability of certain bioactives when compared to fresh fruits, and therefore, it is perhaps surprising how little information is available on characterising the effect of genotype as well as optimum maturity at harvest on fresh blackcurrant-derived bioactives.

In the present study, berries from 2006 and 2007 harvests were assessed to elucidate the genotypic variability of bioactive compounds from a wide range of UK-grown blackcurrant cultivars and better understand the temporal changes in bioactive compounds and antioxidant activity that occur during the latter stages of blackcurrant ripening. Results illustrated the crucial role that genotype has in determining the content of bioactive compounds in blackcurrant berries since significant differences were observed for most of the target analytes studied (*viz*. individual anthocyanins, total phenolics and ascorbic acid). On a FW basis, cv. 9198-1 had the highest ascorbic acid content (1.7-fold higher than the mean value) whereas cv. 871-5 showed 1.5-fold higher anthocyanins content and greater total phenolic content than the mean value of the different cultivars. Furthermore, the temporal flux in bioactive compounds during blackcurrant ripening was also strongly influenced by the genotype. Generally, overripe berries tended to have higher levels of anthocyanins, total phenolics and antioxidant activity than early ripe or fully ripe berries.

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HEALTH VALUABLE COMPOUNDS IN BLACKURRANTS EVALUATED AT THE RESEARCH INSTITUTE OF POMOLOGY AND FLORICULTURE (RIPF), SKIERNIEWICE, POLAND

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Blackcurrant, Ribes nigrum L,. anthocyanins, ascorbic acid, procyanidins

Experiments on blackcurrants conducted at the Research Institute of Pomology and Floriculture (RIPF) are focused on characterization of productivity value of the studied genotypes. However, from the health point of view the chemical composition of fruit are also of interest. During last three years (2005-2007) 22 cultivars and breeding selections were tested and basic quality parameters were determined. Investigated were soluble solids content, titratable acidity, anthocyanins and ascorbic acid. The differentiation in soluble solids content was in a range from 12.0 up to 22.0%, which indicates the impact of genetic factors on fruit composition. Large differences were also found in the case of titratable acidity - 1.9 to 4.0 g/100g. Even larger differentiation was in biologically important compounds (antioxidants), such as anthocyanins and ascorbic acid. For anthocyanins it was from 116 to 479 mg/100g while in the case of ascorbic acid from 94 to 328 mg/100g. On the average the blackcurrant fruit contained 299 mg/100g of anthocyanins and 158 mg/100g of ascorbic acid being a good source of compounds having substantial antioxidant activity.
Blackcurrant fruits are also a good source of other health valuable compounds as flavan-3-ols which in this fruit are present in the polymerized form rather than a single compounds. These compounds are called procyanidins. In three blackcurrant cultivars, which are important for blackcurrant production in Poland the procyanidins content was determined during research within ISAFRUIT project (EU funded project, contract No. FP6-FOOD 016279-2).
The content of procyanidins in fruits harvested during 2006 year was 163 mg/100g for ‘Tiben’; 200 mg/100g for ‘Tisel’; 287 mg/100g for ‘Ben Lomond’. These blackcurrant cultivars are also characterized by high degree of procyanidins polymerization depending on the measurement methods (T – thiolysis; Ph - phloroglucinolysis): 24.2 T – 26.7 Ph - ‘Tiben’; 33.9 T – 36.3 Ph - ‘Ben Lomond’; 41.3 T – 41.7 Ph - ‘Tisel’.

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DETERMINATION OF THE VARIANCE COMPONENT OF CAPACITY AND CHEMICAL PROPERTIES OF STRAWBERRY (FRAGARIA ×ANANASSA)

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Anthocyanins, FRAP, TEAC, total phenolic, variance components

There is an increasing interest in the chemical properties and antioxidant capacities of small fruit species. These characteristics are now being included among the breeding objectives. Thus, it is important to determine the genetic and environmental components of the variances for these characteristics. In this study, we grew 18 strawberry cultivars that originated from diverse breeding programs in three environments (greenhouse, plastic tunnel and open field). Fruit total phenolic (TP), total monomeric anthocyanins (TMA), and antioxidant capacity of fruits by both the FRAP (ferric reducing antioxidant power) and TEAC (trolox equivalent antioxidant capacity) assays were determined. Harvesting dates differed based on the growing conditions; however, we compared the performances of the cultivars on their highest production picks.

Environmental treatment, genotypes and their interactions were all significant for all four traits. On average, the TP and antioxidant capacities determined by both methods were lower in the greenhouse (1585, 8.0 and 6.1 µg GAE/g fresh weight (fw), respectively) than either plastic tunnel or open field treatments, which were not significantly different from each other (2337 and 2229 µg GAE/g fw; 9.2 and 9.3 µmol TE/g fw; 8.2 and 8.1 µmol TE/g fw, respectively). The greatest TMA was recovered from the open field (115 µg cy-3-soph/g fw) followed by the plastic tunnel (97 µg cy-3-soph/g fw) and greenhouse (85 µg cy-3-soph/g fw). We estimated the percentage of each component on total variation. The greatest proportion of variations for total TMA and antioxidant capacity (FRAP and TEAC) was among the genotypes (73, 41, 43%, respectively). The environmental component was largest for TP (54%) followed by genotypes (17%). These results indicate that chemical properties and antioxidant capacities are greatly affected by the environment, while breeding studies have the potential to create genotypes with high phytonutrient contents.

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DEVELOPMENT OF HIGH THROUGHPUT ANALYSES OF POLYPHENOL COMPOSITION IN BERRIES USING ABBREVIATED MASS SPECTROMETRY TECHNIQUES

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Anthocyanins, Inheritance, Polyphenols, DIMS, LC-MS, Fruit Quality.

Tannin-enriched extracts from raspberry, cloudberry and strawberry were analysed by liquid chromatography mass spectrometric (LC-MS) techniques and by an abbreviated direct infusion mass spectrometric (DIMS) technique. The DIMS spectra compared well with the polyphenol composition described by LC-MS. For example, the predominance of signals associated with ellagitannin diversity in cloudberry and raspberry extracts and the diversity of signals from proanthocyanidins in the strawberry extract could be identified. The ellagitannin-derived signals in the raspberry tannin sample showed dose-related saturation probably due to ion suppression effects. Nevertheless, negative-mode DIMS spectra of whole berry extracts described qualitative differences in ellagitannin-derived peaks in raspberry, cloudberry and strawberry samples. Positive mode DIMS spectra also illustrated qualitative differences in the anthocyanin composition of berries of progeny from a raspberry breeding population previously analysed by LC-MS. With suitable controls, DIMS can be applied to rapidly assess differences in polyphenol composition, especially in large sample sets such as the progeny from breeding programmes or environmental studies.

Short column abbreviated mass spectroscopic (SCAMS) techniques that provide rapid analysis times and overcome ion suppression effects seen in DIMS will also be described.

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AUTOMATION OF ANALYTICAL METHODS TO SCREEN ANTIOXIDANT CAPACITY OF BIOLOGICAL MATRICES

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antioxidant; automation, micro plate, photometry, fluorencence

One aspect of the initiation and development of cardiovascular diseases and cancers are oxidative processes that lead to the generation of hydroxyl radicals and peroxyl compounds. There is a growing body of evidence that chemopreventive antioxidants contribute to inactivate “reactive oxygen species”. In any case, assessment of the antioxidant capacity of food or other biological matrices is of the utmost importance.

To assess antioxidant capacity, several photometric tests are used, to give the most information on the reactivity of antioxidant mixtures and the highly complex reactions occurring. Recently, RESAC has been published, an electrochemical methods for a rapid screening of antioxidant capacity. Frequently used tests are DPPH, FRAP, ORAC or total phenols according to Folin-Ciocalteu to give an idea on antioxidant activity. These photometric tests are time consuming and spend a lot of reagents.
Therefore, we transferred these photometric tests on micro plate, to be able to analyse a multiplicity of samples in a short time using a minimum of reagents.

Trolox, gallic acid as standard compounds as well as plant extracts and human plasma have been analysed conventionally and after miniaturization on micro plates. Values show a good correlation for all the four methods tested.
The new methods have been characterised according to their limit of detection, linear domain, repeatability and reproducibility.

The described miniaturized and automated methods can be carried out routinely and permits rapid and sensitive screening of the antioxidant capacity of standard solutions as well as complex biological materials. The new micro plate methods are much quicker and environmental-gentle. Much less reagents and solvent are used. The methods are well suited for quality and specification control and may be implemented as part of an in-house quality system. Additionally, the present methods may facilitate future studies concerning the alteration of antioxidant capacity during growth, storage and processing of food or medicinal plants.

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EFFECT OF PREHARVEST FACTORS ON STRAWBERRY (*Fragaria* x *ananassa*) BIOACTIVES

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Anthocyanins, ascorbic acid, water deficit irrigation

Strawberry fruits represent an important source of bioactive-compounds including ascorbic acid, anthocyanins and phenylpropanoids. The concentration of health-related compounds in strawberry fruits is, however, known to be affected by both preharvest and postharvest factors. Exposure of strawberry plants to stress during fruit development may enhance the concentration of certain bioactives in fruit. In the present study the response of strawberry plants (cv. Elsanta) grown under both deficit irrigation (DI) and/or inoculated with *Botrytis cinerea* was evaluated to determine the effects of such imposed conditions on bioactives in harvested fruit. Inoculation with *B. cinerea* did not have an impact on any of the target analytes studied. However, despite DI detrimentally affecting berry size, drought stress had a profound effect on fruit physiology and biochemistry. Dry matter content as a proportion of fresh weight was increased by a quarter in fruit from water-stressed plants as compared to fruit harvested from plants held at or near field capacity. The concentration of the main strawberry anthocyanins (*viz*. pelargonidin 3-glucoside and pelargonidin 3-derivatives) increased with DI. This said, cyanidin 3-glucoside tended to be lower in fruit from DI-treated plants as compared to plants kept at or near field capacity. While ascorbic acid content was not affected by drought stress, total phenolics content and total antioxidant activity increased by as much as 1.4-fold in fruit from water-stressed plants. Given that the promotion of strawberry sales is being increasingly based on 'healthfulness' and flavour rather than just berry size and yield, manipulating water delivery may be a viable prospect for increasing fruit quality. The possible mechanisms of increased synthesis of strawberry-derived bioactives as a result of drought stress are discussed.

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UNDERSTANDING HEALTH-PROMOTING BIOACTIVE COMPOUNDS IN BLACKCURRANTS AND THEIR AGRONOMIC IMPROVEMENT

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Polyphenolics, Ribes ,neuroprotection, breeding, farming methods

Black currant (*Ribes nigrum* L) is in Europe an important berry for food industry mainly because of its color and organoleptic properties, which makes it suitable material for juice industry, liquors, jams, yoghurts and many other food applications. In addition to high vitamin C content, black currants contain a high level of number of bioactive compounds with potential health-promoting properties. A recent study shows that black currant is a very rich source of phenolic compounds. Among the 143 vegetable foods analyzed black currants were included in the top 10 list in terms of their polyphenol concentration. A very high content of anthocyanins (250 mg/100g of fresh fruit) have been detected from berries, and at least part of the orally administered anthocyanins are absorbed by humans and are found as intact anthocyanin glycosides in the blood. A high level of several flavonols including myricetin, quercetin, kaempferol and isorhamnetin as well as polymeric pro-anthocyanidins, has also been detected. Among hydroxycinnamic acids, caffeic acid, m-coumaric acid, p-coumaric acid, ferulic acid and sinapic acid, are the most abundant. The potential health benefits of flavonols, anthocyanins and other phenolics such as reducing risk of having cancer, cardiovascular and type II diabetes have been suggested, but specific health benefits remains to be waiting for extensive clinical trials. The phenolic extracts from black currant have recently demonstrated to provide effective neuroprotection against oxidative stress induced neuronal damages in human cell cultures. Among phenolics, anthocyanins are considered the most potent neuroprotective compounds found in soft fruits, and as a black-couloured berry, black currant contains high amount of antocyanins, four major anthocyanins (delphinidin 3-O-glucoside, delphinidin 3-O-rutinoside, cyanidin 3-O-glucoside, and 3-O-cyanidin rutinoside) are reported in black currants.

The potential health benefits of black currant based products may be greatly increased if berries from cultivars with high contents of the health-promoting compounds are used as raw materials. The high variability in the levels of flavonols in different black currant cultivars have been demonstrated showing the potential of plant breeding for taking advantage to develop special cultivars for high in health-promoting compounds in target populations. Farming methods either growing organic or conventional means, did not found to have any major influence on the phenolic contents in blackcurrants. With the help of molecular-marker techniques, introgression of defined genes or genome parts possessing health effects is a tool box of modern breeding for speeding up the breeding progress.

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BTH INDUCES PROTECTION AGAINST DOWNY MILDEW IN ARCTIC BRAMBLE (*RUBUS ARCTICUS*) AND THE ACCUMULATION OF SEVERAL PHENOLICS

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Benzothiadiazole, BTH, ellagitannins, oomycete

Arctic bramble fruits (*Rubus arcticus*) are mainly collected from nature in Finland, but there is an increasing interest in cultivating this rare, highly valued and aromatic fruit. Downy mildew (*Peronospora sparsa*) disease causes high annual fluctuation in the yield of arctic bramble particularly during rainy summers. Therefore it is important to study the performance of different arctic bramble cultivars under the downy mildew infection pressure in the changing climate. The possible molecular basis for downy mildew resistance in arctic bramble is unknown, because none of the present cultivars is completely resistant to the disease. We have studied the ability of benzothiadiazole (BTH) to induce resistance against downy mildew, and its effect on the content of different phenolic compounds and proteins. Fewer symptoms and less pathogen DNA was found in BTH-treated plants than in the controls under greenhouse conditions. Pretreatment with BTH also inhibited sporulation on the leaves detached four days after the BTH treatment and inoculated *in vitro* on water agar plates. BTH induced the accumulation of flavonol glycosides and specific ellagitannins, the amount of which correlated negatively with the amount of pathogen DNA measured from the same samples. Lower concentration of BTH caused greater accumulation of phenolics. Based on proteomic analysis by two-dimensional gel electrophoresis and mass spectrometry, BTH caused up-regulation of 79 proteins, of which 13 could be identified. PR-10, flavanone-3-hydroxylase, caffeoyl-CoA-3-*O*-methyltransferase, ACC oxidase, and alanine aminotransferase were identified as defense-related proteins. Strong induction of PR-1 was found in Western analysis, suggesting the activation of SA-mediated defense. In conclusion, the activation of SA-mediated defense pathway, large changes in the leaf proteome as well as the accumulation of specific phenolic compounds may explain the resistance to *P. sparsa* induced by BTH in arctic bramble.

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QUALITY OF STRAWBERRY CV. ELSANTA AFTER HOT WATER DIPS OF DIFFERENT EXPOSURES

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Fragaria x ananassa Duch., heat treatments, quality, phenolic compounds

Strawberry fruit (*Fragaria x ananassa* cv. Elsanta) grown in soilless culture systems at Sveta Nedjelja near Zagreb were dipped in hot water (HWD) at 48° C for 5, 10, 15 and 20 min and stored after 7 says at 0° C. Hue value was significantly lower between 15 and 20 min exposures compared to the 5-10 min. Compared to control fruits, heat treatments generally decreased quality parameters with exceptions of SSC. Flesh pH was higher after dips for 15 and 20 min at 48° C than in dips for 5 and 10 min and there was significant difference between 15 and 20 min of exposure time. Increasing exposure time from 5 min to 20 min had significant effect on invert sugars. This treatment had lowest TA and the highest SSC/TA ratio, resulting with bland taste as determined with sensory panel. Storage significantly decreased flavan-3-ols (catechin, epicatechin) and flavonol glycosides (myricetin, quercetin and kaempferol) in control fruits. Heat treatments decreased concentrations of all studied phenol compounds. Exposure time had no significant effect on p-hydroxibenzoic and caffeic acid. However, 20 min of exposure time preserved p-hydroxibenzoic acid, (-) epicatchin myricetin, and kaempferol, but reduced ellagic acid to the significantly lowest level.

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BERRYFRUIT FOR HEALTH RESEARCH AT HORTRESEARCH, NEW ZEALAND

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Berryfruit, anthocyanins, antioxidants,inflammation, human health

New Zealand has a large fruit production sector and a strong, successful berryfruit industry, particularly in blackcurrants, Boysenberries and blueberries. The industry has shown significant growth in recent years, following the increasing interest by consumers in potential health benefits of berryfruit. Research innovation has also played an important role in this growth. HortResearch is a world-leading fruit science company, recognised as providing high quality and high impact research and innovation outcomes for New Zealand's $4 billion dollar horticultural sector. HortResearch recognises and encourages collaboration between its scientists and scientists from overseas institutes or universities.

In July 2008 a major HortResearch berryfruit programme, which targeted gut health in the final year, came to a successful end. This programme was followed by a new berryfruit programme called 'New Berries', which has been funded for a further five years. The new programme has a multi-faceted approach to develop and extend the knowledge and understanding of berryfruit phytochemicals and how they may contribute to enhancement of human health and well-being. This knowledge is exploited through plant breeding, genetics, flavour standards and biological health screening (*in vitro* and *in vivo*) to develop new flavoursome berryfruit products with supporting robust scientific health evidence. The programme is particularly targeted at a combination of flavour attributes and health research for blackcurrant, blueberry and Boysenberry fruits. Health attributes being investigated include regulation of inflammation and how this affects digestive health and physical fitness.

Dr Roger Hurst from the Functional Food & Health team at HortResearch leads the new berryfruit programme and will overview here the previous and new programmes and their key scientific outcomes, with the aim of encouraging international collaboration to speed up research progress for the New Zealand and the EU berryfruit industries.

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POLYPHENOLS FROM BERRIES: METABOLISM AND BIOAVAILABLITY

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Polyphenol, flavonoid, phenolic acid, health, bioavailability

Berries contain a range of (poly)phenols, from anthocyanins to flavonols, flavanols, and phenolic acids. Understanding of the absorption and metabolism of these compounds has expanded enormously over the last 10 years, although there are still gaps in our knowledge especially for anthocyanins and phenolic acids. Although the small intestine is the major site of absorption for intact polyphenols, the colonic microflora plays a major role in metabolism of larger polyphenols to smaller compounds, which can then also be efficiently absorbed. The combination of parent polyphenols and metabolised phenolics constitute a considerable array of compounds which could have biological activity. This combination of compounds gives a much greater total concentration than any individual chemical species alone. This consideration is especially important for berries where the large number of parent compounds can give rise to a wide range of metabolites both from mammalian and microbial metabolism. For this reason, biological effects in vivo are more often observed on polyphenol-rich foods, such as berries, than on single isolated compounds. The bioavailability of polyphenols from berries will be reviewed, including their absorption, metabolism and excretion.

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ANTIOXIDANT CAPACITY OF PORTUGUESE ENDEMIC *RUBUS* FRUITS IN A NEURODEGENERATION CELL MODEL

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Rubus sp., antioxidant activity, neurodegeneration

A wide array of plant phenolic substances have been reported to have substantial neuroprotective activity, intervening on multiple biological processes such as iron chelation, radical scavenging, activation of survival genes, cell signaling pathways, regulation of mitochondrial function and possibly the ubiquitin/proteasome system. *Rubus* species are a well known source of polyphenols with high antioxidant activity that may provide human health benefits.
The aim of this work is to compare the neuroprotective antioxidant activity of *Rubus idaeus* fruits with endemic species fruits from the Northeast of Portugal.
A hydroethanolic extraction of fruits from *R. idaeus, R. brigantinus, R. sampaioanus, R. vigoi e R. genevieiri* was performed. Total phenolic content and the *in vitro* antioxidant properties of *Rubus sps.* fruits extracts were then evaluated by the Oxygen Radical Absorbance Assay. The ratio ORAC/total polyphenols is equivalent for *R. idaeus, R. brigantinus, R. sampaioanus,* and slightly higher than *R. vigoi* and *R. genevieiri* that has the lower ratio.
Toxicity tests of *Rubus sps.* fruits extracts were performed in a neuroblastoma cell line (SK-N-MC) using CellTiter-Blue® kit. A nontoxic range of concentrations was defined using the SK-N-MC cells. The intracellular radical scavenging activity of the plant extracts in an oxidative stress-induced model of neurodegeneration in SK-N-MC cells was evaluated to the nontoxic range.
The pre-treatments with the extracts of *R. idaeus, R. brigantinus, R. sampaioanus* protects the cells from the oxidative stress injury as detected by an increase in cell viability up to 50% with 62,5 μg GAE.mL^{-1} and 60-70 % with 125 μgGAE.mL^{-1} of *R. idaeus, R. brigantinus.*
These results for *in vivo* radical scavenging activity of the *Rubus sps.* fruits extracts confirm the potential evaluated by the ratio ORAC/total polyphenols. These uncharacterized fruits revealed to be a promising source of natural antioxidants. Further studies will confirm their possible future use as neuroprotective compounds.

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STRAWBERRY CONSUMPTION AND ANTIOXIDANT STATUS IN HUMAN SUBJECTS.

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Folate, plasma antioxidant capacity, phenols, oxidative damage, erythrocyte, vitamin C

Strawberry represents one of the most commonly consumed berries, and is a relevant source of dietary micronutrients and phytochemicals such as vitamin C, folate and phenolic compounds. After a fine characterization of the antioxidant, micronutrient and phytochemical composition of strawberries from plants with different genetic background, we carried on sequential feeding studies based on acute and prolonged consumption of strawberries in addition to the usua diet of the human subjects.
We focused our attention on the evaluation of the potential short-term and medium-term effects on the plasma and cellular (erythrocytes and lymphocytes) antioxidant status, which could be correlated to the consumption of strawberries. The work included the evaluation of potential quantitative variations in the hydro-and lipophilic serum antioxidants and in the plasma Total Antioxidant Capacity (TAC). We also assessed the putative improvement in the cellular mechanisms of protection against spontaneous and induced oxidative damage, measured in normal erythrocytes and lymphocytes isolated from human subjects, before and after the period of strawberry intake.
In keeping with previous findings, the plasma antioxidant capacity and the serum ascorbate contents significantly increased during both the studies. Surprisingly, the serum urate levels were unchanged, confirming that the improvement in the plasma antioxidant status after strawberries consumption was related to the absorption of dietary antioxidants, with the main relevant contribution of vitamin C, rather than to the increase of endogenous metabolic urate.
However, the in vivo contribution of the strawberry phytochemicals to the increased plasma antioxidant properties failed to be ascertained, and no traces of phenolic derivatives were observed in fasted serum collected during the prolonged strawberry consumption, indicating that there is no evidence for long bioavailable circulating phenolic metabolites after several hours from the last intake of the fruits. Together with the strawberry consumption, a progressive enhancement of the erythrocyte membrane resistance to spontaneous and AAPH-induced hemolysis was observed. These findings seem to confirm the already reported role of polyphenols in enhancing red blood cell resistance to oxidative stress, in vitro and in vivo,and suggest the erythrocyte bilayer as a possible preferential localization of these phytochemicals.
Additional investigations have been conducted in order to evaluate the putative improvement of lymphocyte resistance against oxidative stress-induced DNA damage, and future studies have already been designed to further investigate the potential of strawberries in the prevention and treatment of specific diseases in human subjects.

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ABSTRACTS

POSTERS

BIOACTIVE CONSTITUENTS AND DNA PROFILING OF RASPBERRY GERMPLASM COLLECTION IN BULGARIA

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Keywords: raspberry, phenolic acids, flavonoids, Microsatellite SSR

Recent progress in molecular analyses and agricultural biotechnologies has enormous impact on selection, technology, testing, preservation and processing of agricultural products. Metabolomic assay as a new dimension in these studies and practice focuses the attention on the biochemical contents of cells and tissues, and has a rapidly growing significance in knowledge of small fruits value for human health. Bulgaria is a traditional and important producer of small berries in Europe.

Raspberry is a very reach source of phenolic compounds such as phenolic acids and flavonoids. The content of phenolic acids (caffeic, *p*-coumaric and ferulic acids) and flavonoides (hyperoside, isoquercitrin, and tiliroside) in samples of wild and cultivars *Rubus* L. leaves and DNA profiles of 20 raspberry accessions as a part of the Bulgarian raspberry collection, have been determined.

The reversed-phase high performance liquid chromatography (RP-HPLC) separations of phenolic compounds were performed on a Microsorb-MV 100 C18 column using linear gradient elution with a mobile phase composed of 20 mM phosphate buffer (pH 3.12) and methanol, and UV detection at 254 nm.

The leaves of cultivars are characterized by greater amounts of phenolic compounds than the leaves collected from natural habitats. Caffeic acid was the dominant phenolic acid in the majotity of the samples being present in amounts between 0.04 mg/g in the wild species and 1.43 mg/g in cultivars. The highest content of isoquercitrin, hyperoside and tiliroside was found in the cultivated varieties of raspberry: 1.70 mg/g, 0.99 mg/g and 0.60 mg/g, respectively.

In addition to the metabolomic assay, DNA fingerprint profiles of raspberry accessions obtained by Single Sequence Repeat (SSR) methods were compared.

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VARIATION IN QUALITY TRAITS AND ANTIOXIDANT CAPACITY OF TEN STRAWBERRY CULTIVARS

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Ascorbic acid, Fragaria, fruit yield, antioxidant capacity, Vitamin C

In the past, breeding for new strawberry cultivars was mainly focused on yield, external appearance and transportation tolerance of the fruits. Today, the fresh market is getting more consumer-oriented and beside a good productivity and fruit firmness the demand concerning the quality of the berries is increasing. However, there is a lack of knowledge concerning quality traits like gustative properties and nutritional value of new cultivars. The aim of this work was to evaluate strawberry cultivars with commercial importance on the basis of agronomical parameters as well as quality and nutritional parameters.
For this purpose ten cultivars were chosen according to their genetic diversity and commercial importance. Frigo plants were planted in July 2007 in Conthey (Switzerland). Agronomical parameters like harvest begin, leaf area, fruit yield and mean fruit weight were measured. The fruit colour, the soluble solids content, the firmness and the titratable acidity were measured at mid harvest. The nutritional quality of the fruits was evaluated by determination of the ascorbic acid content, the antioxidant capacity (DPPH and FRAP assay), the total phenolic content (Folin-Ciocalteu assay) and the total anthocyanin contents.
A factorial discriminant analysis of the ten cultivars was done with all the measured parameters. A great variability in the nutritional quality of the selected cultivars was observed. Ascorbic acid content and harvest begin were the two main parameter allowing differentiating between the selected cultivars. This variability could be highly interesting for breeding new cultivars. However, ascorbic acid content, as well as antioxidant capacity were generally negatively correlated with the fruit yield. Further research is necessary to meet high fruit yield with high nutritional value of the fruits.

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PRELIMINARY CHARACTERISATION OF CORNELIAN CHERRY (*Cornus mas* L.) GENOTYPES FOR THEIR PHYSICO-CHEMICAL PROPERTIES

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Cornelian cherry, Cornus mas, antioxidants, FRAP, phenolics

In recent years increasing attention has been paid by consumers to the lesser known fruits such as cornelian cherry, honeysuckle, hardy kiwifruit, lingonberry, elderberry, sea buckthorn, medlar, bilberry, strawberry tree, sea buckthorn, arctic bramble, mountain ash, wild persimmon, lingonberry, cloudberry, cherry laurel, jujube, rose hip, etc., which have unusual flavors and qualities, and many of which are rich with antioxidants and anthocyanins. Fruit weight, antioxidant capacity, total anthocyanins, total phenolics, ascorbic acid, soluble solid content (SSC), reducing sugar, tannins, and acidity of a number of selected cornelian cherry (*Cornus mas* L.) genotypes of varied pigmentation were investigated. The genotypes were selected previously different part of Turkey and a germplasm collection with these selected promising cornelian cherries has been established in Malatya Fruit Research Institute in 1995 year. This study was conducted in 2007 year aiming to determine healthy benefit components of selected a number of cornelian cherry genotypes. Two methods, namely B-carotene bleaching and ferric reducing antioxidant power (FRAP) were used to determine total antioxidant capacity, while Folin–Ciocalteu reagent was used to determine total phenols. Fruit weight, SSC and ascorbic acid content of genotypes were 2.09–9.11 g; 15.83–21.17% and 31–112 mg/100 g, respectively. Antioxidant activity and total phenolic content varied among genotypes and 44-18 genotype had the highest antioxidant capacity using both methods. This genotype also had the highest total phenolic (74.8 mg GAE/g DW) and total anthocyanin (115 mg cyanidin-3-glucoside equivalents /100 g FW) content. There are linear relationships between antioxidant capacities and total phenols. The present study demonstrates the potential of certain cornelian cherry genotypes, notably 44-18 for using future breeding studies.

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VARIATIONS IN ALLERGEN CONTENT OF EUROPEAN BLUEBERRIES (*VACCINIUM SP.*) GROWING IN DIFFERENT AREAS AND CLIMATE ZONES

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Vaccinium myrtillus, allergens, nutrition, environmental parameters

In recent years there has been an increased interest in berry fruits rich in phenolic compounds including flavonols, flavones, anthocyanins and different biological active acids due to their possible beneficial effects on human health (Rieger et al. 2008). In this context the European wild blueberry (*Vaccinium myrtillus*) was shown to contain various antioxidant molecules and more antioxidant capacity even than strawberry, apple, peache, grape, tomato, or kiwifruit.
In order to evaluate the potential for industrial exploitation and to determine the nutritional quality of *Vaccinium myrtillus,* the interaction between different parameters should be established, providing clear information for breeders, growers and consumers. For this purpose in the current study climatic conditions were monitored by meteorological stations, comparing conditions for *Vaccinium* populations from South, Mid- and North Norway covering latitudes from 61°N to 68°N and from Austria at 48°N.
So far, no detailed description of *Vaccinium* allergens was available, which might be due to the fact, that blueberry consumption is not very high, compared to other fruits, e.g. apple. Allergic symptoms related to PR-10 proteins are described as mild and local, which might be due to the chemical lability of these proteins. PR-14s, however, were reported to cause anaphylaxis, after consumption of blueberries (Egger, pers. comm.). To analyse the pathogenesis/stress-related protein composition of *Vaccinium myrtillus*, in a first step Western blotting was carried out, using polyclonal antibodies raised against PR-10 and PR-14 of *Malus domestica*, which cross-reacted with similar epitopes in highly conserved protein families of other plant species (Marzban et al. 2005).
We addressed the question, whether there might be an influence of different climatic conditions on the expression of PR-10s in blueberries. Dot blots were carried out including purified Mal d 1 as control standard. PR-10s was shown as being sensitive to abiotic and biotic stresses in *Vaccinium* species (Jaakola et al. 2008). Preliminary results obtained demonstrate significant differences in PR-10s content of blueberries grown in different areas and under low temperatures over the entire ripening period. Long-term measurements are needed for a better understanding of the influence of environmental low temperatures on the expression of PR-10s in blueberries.

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HEALTH – PROMOTING COMPONENTS IN OLD AND NEW ITALIAN STRAWBERRY VARIETIES

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Antioxidant Capacity, Ascorbic acid ,Total Polyphenols, Fragaria

The fruit quality is one of the main aims of strawberry research programmes to obtain new dessert cultivars with particular attention on the health benefits or to determine the ability to enhance the content of nutritional compounds. It is known that the fruit contains many antioxidant components of different origins and amounts. Its consumption has been primarily associated with a lower incidence in degenerative diseases like cancer and heart disease, but it can also reduce blood pressure, boost the immune system and reduce inflammation (Wang *et al*., 2002). The phytochemicals responsible for the antioxidant capacity, especially of red fruits, can largely be attributed to phenolics, anthocyanins and flavonoid compounds. Strawberry is considered a source of many bioactive phytochemicals. Its fruit contains ascorbic acid and polyphenolic compounds providing protection against harmful free radicals, skin ageing (Kahkonen *et al*., 2001; Wang *et al*., 2002) and ellagic acid, a natural phenolic that is both an antimutagen and an anticarcinogen (Olsson *et al.,* 2007).

The aim of this work was to investigate on the fruit health compounds of old and new strawberry varieties (*Fragaria x ananassa* Duch) grown in the Cesena area (Emilia-Romagna region – Italy). The two-year (2007- 2008) study, was carried out using 21 strawberry cultivars: respectively in the first year, 7 old varieties coming from "Cost 836 core-collection" (Annalie, Avalon Classic, Dr. Morere, Louis Gauthier, Rabunda, Regina and Tardiva di Romagna) were compared with 8 varieties coming from Italian strawberry breeding (Argentera, Alba, Dora, Onda, Queen Elisa, Vale, Tecla and Unica). In the second year, the Cost core-collection cultivars were compared with 8 varieties (Candonga, Ciflorette, Clery, Darlisette, Daroyal, Darselect, Onda and Queen Elisa). The trials were managed using soil fumigated at pre-planting with a mixture of chloropicrin (30 g/m^2) and 1, 3 dichloropropene (25g/m^2) and 10 unit/ha of mineral N supplied by fertigation. Each year a completely randomized block design with four replications of single plots of 10 cold-stored plants per cultivar was adopted in the growing system. During the fruiting season a sample of twenty ripe fruits per plot was taken at the third picking and then it was dried and used for analysis of health–promoting properties and components (total polyphenols, antioxidant capacity and vitamin C). The results are still ongoing and will be presented later.

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ANTHOCYANINS AND ASCORBIC ACID – IMPORTANT QUALITY TRAITS IN STRAWBERRY BREEDING PROGRAM AT RIPF, SKIERNIEWICE, POLAND

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Ascorbic acid, vitamin C, anthocyanins, Fragaria, strawberry

Strawberry breeding program at RIPF, Skierniewice (Poland) is aimed at obtaining cultivars for fresh market and also for processing. Dessert cultivars should produce large fruits, with light red colour and strong glossiness, high firmness and good taste. Fruits for processing should possess high level of extract, dry matter and also very good capping ability. Additionally, both kinds of fruits should contain high level of ascorbic acid and anthocyanins, because these components are very important for maintaining good human health. The ascorbic acid is an activator of many enzymes, increases the immunity of the organism and is involved in processes of biological oxidation and ahthocyanins have the ability for dezactivation of heavy metals.

The aim of the work was evaluation of content of ascorbic acid and anthocyanins in fruits of new strawberry cultivars bred at the Research Institute of Pomology and Floriculture, Skierniewice, Poland. Analyses were done using 0,5 kg samples of fully matured fruit, selected from 3, 4 and 5 harvest. Fruits were washed, capped, packed hermetically and frozen at -25^{o}C. Before analysis the samples were disintegrated in the frozen state and mixed thoroughly to obtain uniform material. In the case of organic and ascorbic acid determination, the frozen material was homogenized in 6% HPO_3 solution and after filtration analyzed by HPLC using Hewlett-Packard 1100 chromatograph, equipped with two Supelco LC-18 25 cm columns in sequence. For the determination of anthocyanins, calculated as pelargonidine-3-glucoside/100g of raw material, extinction coefficient taken for the calculation was 22400 and molecular weight M=433 (Wrolstad, 1976). Content of ascorbic acid and anthocyanins was analyzed in two or three consecutive seasons, depending of the cultivar.

The results showed, that several new Polish cultivars possess higher content of ascorbic acid in fruits in comparison to standard cultivars: ‘Senga Sengana’ and ‘Elsanta’. The highest ascorbic acid content was found in fruits of advanced clone SK-98054-01 (86,1 mg/100g), ‘Panon’ cv. (79,75 mg/100g), ‘Elsariusz’ cv. (74,4 mg/100g) and ‘Hokent’ cv. (64,4 mg/100g). Fruits of the standard ‘Senga Sengana’ contained only 37,95 mg of ascorbic acid in 100g.

Many new Polish cultivars contained also more anthocyanins in fruits than both standard cultivars (‘Senga Sengana’ – 29,85 mg/100g and ‘Elsanta’ – 20,35 mg/100g). The highest content of anthocyanins was found in fruits of clone SK-98054-01 (65,7 mg/100g), ‘Feriusz’ cv. (48,45 mg/100g), ‘Marduk’ cv. (43,5 mg/100g), ‘Recoda’ cv. (41,75 mg/100g) and ‘Hokent’ cv. (41,2 mg/100g).

Presented results show, that through the breeding program it is possible to increase level of bioactive compounds in strawberry fruits.

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ESSENTIAL OILS FROM RIBES NIGRUM BUDS: GC/MS ANALYSIS AND ANTIMICROBIAL ACTIVITIES

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Ribes nigrum, Essential oil, GC/MS, Antimicrobial activity, Biofilm

Ribes nigrum L. (blackcurrant) is cultivated extensively in many countries of Europe and their buds are used in medicine for their diuretic, antiseptic and anti-herpesvirus activities (Suzutani, 2003) which are due to the biological activity of their volatile compounds. The scientific literature is recently citing the antimicrobial activity of some gemmoderivatives that contain extract of *Ribes nigrum* buds (Mihele, 2007).
All varieties of *Ribes nigrum* buds used in this study were obtained at Fruit Research Institute Maracineni Pitesti and they were selected for fruits productivity. Essential oils were isolated by steam distillation and were analyzed by gas chromatography coupled with mass spectrometry. The main volatile compounds identified in all varieties (Abanos, Deea and Ronix) of *Ribes* essential oils were: sabinene, α- and β-pinene, δ-3-carene, limonene, ocimene, α-terpinolene, β-caryophyllene, α-humulene, terpinen-4-ol. This chemical composition is similar with previous studies (Orav, 2002). The *Ribes nigrum* essential oils exhibited large antibacterial spectrum, acting against *Acinetobacter baumanii*, *Escherichia coli*, *Pseudomonas aeruginoasa* and *Staphylococcus aureus*, as proved by the very low MIC values observed for the respective strains. Our results have shown that the essential oils exhibited good antimicrobial activity against the majority of the Gram-negative, as well as *S. aureus* tested strains. It is maybe one of the most interesting pathogens due to its intrinsic virulence.
Concerning the influence of the essential oils tested against the adherent bacteria grown in biofilms, our study showed that the subinhibitory concentrations of *Ribes* oils induced an increase in the bacterial ability to colonize the inert substratum for *A. baumanii, E. coli* and *S. aureus*, as revealed by the higher absorbance values of the adherent bacterial cells, fixed with methanol, colored by violet crystal and resuspended in acetic acid. These effects prove that besides the bactericidal activity, the *Ribes nigrum* essential oils also have anti-pathogenic potential.

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THE EFFECT OF PACKAGING MATERIALS AND TECHNOLOGIES TO THE STORAGE TIME OF FRESH BLACK CURRANTS

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Black currant, packaging, biodegradable films

Black currants have been classified as a non-climacteric fruits, exhibiting no increase in respiration rate or ethylene production during ripening. As a typical soft fruit they have a high physiological post-harvest activity, short ripening and a senescence period.
The objective of this study was to determine the possibility of shelf life extension of black currants, maintaining as far as possible the moisture and quality of berries at the storage time by packaging in different materials.
The effect of packaging material for shelf life extension was determined by using different packages: ready-made PET/adhesive/PP containers (thermoformed containers ML 620 size of 210x148x35 mm and sealed with laminated lidding film *Tecnopack,* composition PET/adhesive/PP, thickness 64 microns, tension resistance 15.67 m^2 kg^{-1}, O_2 permeability – 110 ml m^{-2} $24h^{-1}$, H_2O permeability – 10 g m^{-2} $24h^{-1}$.); PP (polypropylene) trays (210x148x35 mm) inserted in pouches made from PLA films by different thickness 25 and 40 *μm*; pouches (size 350x230 mm) made of OPP (oriented polypropylene) film by thickness 40 *μm***;** cardboard boxes (145x120x80 mm) placed in PLA film pouches by different thickness 25 and 40. Summarizing the experimentally obtained data in year 2007 a conclusion has been carried out – for the next study cardboard boxes placed in PLA film pouches by thickness 40 *μm* could be recommended. New biodegradable packaging form – containers (130x90x70 mm) manufactured with holes (total area of hols 7.9 cm^2) have been used in the year 2008. Usually for fruit packaging used PP boxes (180x110x80 mm) manufactured with holes (total area of hols 14.3 cm^2) were used as control packaging in either experiments. All samples were stored in alight "Commercial Freezer/Cooler ELCOLD" at temperature +4 ± 1 ºC.

The experiments were performed at Latvia University of Agriculture Faculty of Food technology, Jelgava and at Latvia State Institute of Fruit Growing, Dobele. The berries harvested at the year 2007 and 2008 were tested. Contents of ascorbic acid, anthocyanins, color in L*a*b measuring system, changes of moisture content and pH were analyzed during the storage period of three weeks. Oxygen and carbon dioxide dynamics in hermetic packages was analyzed by gas analyzer.

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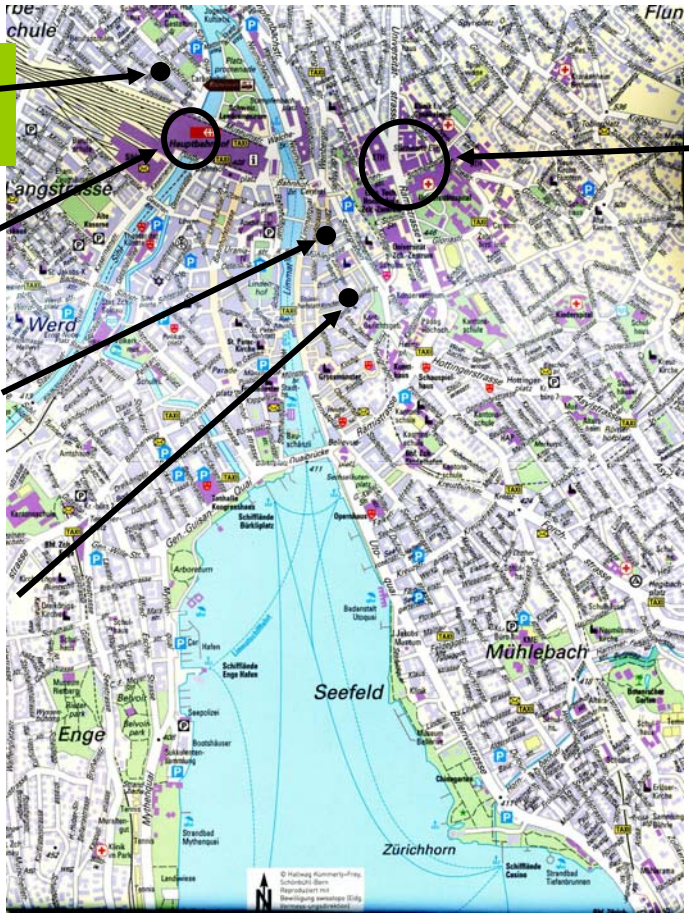
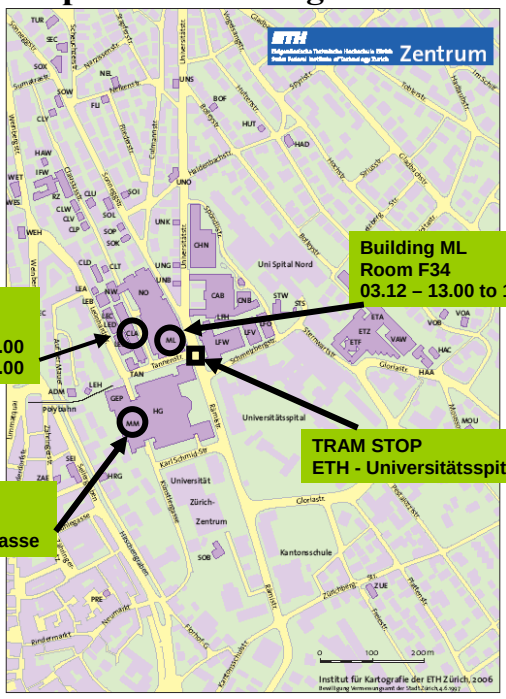
Map of Zürich Center



ETH Zentrum
Map of the buildings



Map of the Restaurant
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