SUSTAINABLE EXPLOITATION OF BIOACTIVE COMPONENTS FROM THE BLACK SEA AREA TRADITIONAL FOODS

BASEFOOD

NEWSLETTER

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Organisation and management

BaSeFood duration extended

Due to the complexity of some experimental and survey activities, especially connected with on-site traditional food surveys and documentation, intervention studies, and technology- chain experiments, it was unanimously decided to apply for a project's time extension, that was approved.
New BaSeFood end date is October 31, 2012.

5th Coordination and Project management board meeting

The meeting was held in Athens, Greece, on February 22-24, 2012. It was organised by Hellenic Health Fundation staff, in the nice context of the Kostis Palamas Building, of the Cultural Centre, Athens University Club.
The meeting was an important milestone, in which the point of the situation of BaSeFood activities was set, and the scheduling of the relevant residual activities, to the extended project's end date was done.
The meeting was organised in three days:

- Day 1. Three parallel sessions were devoted to discuss activities related to:
  - large scale, on site, surveys of foods of the Black sea region;
  - bioactivity assays and intervention studies using selected plant materials or extracts;
  - technological innovation in traditional food chains, bioactive retention determinations, bioactive characterisation of plant raw materials.
- Day 2. This was a plenary meeting day, in which the point of the situation was made by Workpackage and task leaders.
- Day3 was devoted to the Project management board meeting.

The Athens meeting participants.

Plants and foods reports

The general strategy of BaSeFood on-site surveys obeys to a need stemming from the rapid decline in the opportunity to study traditional plants and related foods, connected to the still on-going rapid societal changes, in the most sensitive areas.
In this context, qualitative descriptive surveys are effective tool aimed at:

- Investigating facts in natural, rather than in experimental settings
- Retrieving evidence-based information, that is latent in diffuse knowledge but often absent in official, formal scientific knowledge
- Helping in generating, rather than testing hypotheses
- Helping in programming and tuning possible future quantitative and experimental research

The broad topics on which concentrate the attention have been defined in the course of the project and several reports have been published in the previous issues of this newsletter.
As a general reflection, it is pretty clear that traditional foods were mainly foods of hard periods, in which the ingredients were those available. Very difficult, indeed, to write a standard recipe ! Tradition belongs to people, not to regulators, not even to scientists !

Surveys in Italy, autumn and winter 2011

L. Filippo D'Antuono (UNIBO)

Kales. The documentation of kale recipes and uses in Italy was almost concluded.
Surveys were carried out in the three selected areas of north-central Italy: western Liguria, with special respect to

Broccolo lavagnino on sale, Chiavari market (GE), January 2012.

val di Vara, the area of Lucca and Pisa and the area of Pratomagno, province of Arezzo.
A full picture of traditional and modern uses of kale has been built, ranging from isolated plants used for home consumption, to specialised production, addressed to supermarkets.
Twelve traditional recipes were documented, and critically reviewed in comparison with what published in popular literature.

Palm tree kale, Cembrano (SP), January 2012.

Plam tree kale leaves, packed, Peccioli (PI), January 2012.

Val di Vara black kale, homeyard, Cembrano (SP), January 2012.

As a whole, it clearly appeared how the present interest on kales is generated by its strong link with local food traditions, that are more and more considered by consumers as added value determinants of food preference.

Local corn and corn products. The typical Italian meal made of corn flour is the polenta. There is a rich story about the way rural populations were sustained by corn polenta, in the plains and lowlands and of chestnuts, in mountainous areas.

Red and yellow Ottofile corn, Gallicano (LU), December 2011.

Corn has however been among the crops subject to bigger changes by social and technological evolution: during the last century it changed from a human food cereal to an animal feed staple. In parallel the varietal panorama also completely changed, with the introduction of high yielding hybrids, aimed at maximising energy production for animal diets, so rich in starch, poor in protein and with floury kernels. These types are little suitable to produce good polenta, requiring vitreous kernels, that were typical of local populations. The recovery of the interest on polenta was accompanied by the interest of a parallel recover of good quality local varieties, some of which have been characterised and selected, especially in the North east of Italy, to set up local food production chains. Some other were also used for breeding good quality hybrids for human food.
Although being more typical of northern Italy, the use of local corn varieties, especially of the Quarantino family (40 days cycle), was widespread also in central Italy, where it is however less well documented.

Local milling plant for Ottofile corn, Gallicano (LU), December 2011.

BaSeFood is carrying out surveys in some of these areas, following the usual scheme of interviews with local informants and stakeholders, to document the uses of local resources, people perceptions and relevant traditional recipes.
An interesting case is the one of "Ottofile" (eight rows) corn of Garfagnana.

Ottofile corn packs, Gallicano (LU), December 2011.

It is a local population, once widespread all along the river Serchio valley until the Pisa plain, but then reduced to the mountainous areas of the upper Serchio valley. The residual populations were recovered by local amateurs and subject to a mass selection, separating the yellow kernel from the red pigmented kernel plant, that now represent two fairly stable groups, that are cultivated and processed separately. The kernel are milled in local plants and marketed under an approved and controlled label.
The polenta made from Ottofile is considered a delicatesse and all the product is sold, with sometime demand exceeding offer, raising problems of safeguarding the authenticity of the production.
Other corn foods are prepared in several places. In the Romagna area, the "piadina", the local popular flat bread, used to be prepared with a mixture of corn and wheat flour (and often other starchy materials), to save precious wheat. This use is nowadays almost abandoned, in favour of wheat only product.
In the mountain areas of western Liguria, one of BaSeFood target areas, the "fogasette de mega", (corn round, small size flat bread) are still prepared and used at home level or in traditional restaurants and agritouristic farms.
Also in this case the past value of corn as energy supplier is clear.

Local Quarantino corn spikes, Comuneglia (SP), January 2012.

Culinary herbs. Wild fennel (*Foeniculum vulgare* L.) is the ancestor of all the cultivated fennel forms: from the aromatic plant for seed production for high anethol essential oil extraction, to the sweet or Florence fennel, which parenchymatous leaf sheaths are eaten as a popular vegetable.
Wild fennel is widespread in the Mediterranean areas and, in Italy, it is very common in the centre and the south. In this places, wild fennel represents a real staple for many traditional dishes.
It is a fundamental ingredient for the preparation of porchetta (a piglet cooked whole), it is used in the seasoning of many types of pasta, soups (renowned is a chestnut soup in central Italy), fish , mushrooms etc.
A peculiarity of wild fennel is that almost all the aerial parts are picked and used, depending on the season. The stems and older leaves are the typical filling herb for porchetta.

Dried wild fennel fruits and florets, Forlì market, January 2012.

Mature fruits are also dried and used for seasoning. The younger leaves are used mostly dried. But the very special product is represented by the not yet developed florets, that are gathered in late spring, from wild stands, especially in Southern Toscana and Northern Latium, air dried and stored.
The aroma is extremely intense and they are used in very small amounts. Even since the price can be as high as 150 Euros per kilogram.
Apart from this very special product, other uses seem quite similar to those of dill in eastern European countries.

Wild fennel flavoured tuna fish under glass, from Lampedusa island, Forlì market, January 2012.

Spring sprouts. Wild greens were largely used in the past as staple foods, in periods when naturally available plants represented important dietary components. The range was very ample, and seasonally variable, depending on availability. Typical spring resources were the young sprouts of several plants. In Italy, among these:
Black bryony (*Tamus communis* L.)
European greenbrier (*Smilax aspera* L.)
Butcher's broom (*Ruscus aculeatus* L.)
Wild hops (*Humulus lupulus* L.)
Traveller's joy (*Clematis vitalba* L.)
Blackberry plant (*Rubus* sp. pl.)
Black bryony is being used as an example, in Italy, and a documentation study is being carried out, reporting its ways of exploitation and traditional food uses in selected areas.
Similar investigation is being developed on Georgian greenbriers.

Black bryony sprouts, Civitavecchia market (Rome), Italy.

A traditional soup with black bryony sprouts, Tolfa (Rome), Italy.

Kvass in the southern region of Russia

Elena Beteva, Dmitry Karpenko, Alexandra Krechetnikova (MSUFP)

In August 2011 preliminary case studies of the situation with the production and consumption of kvass in southern Russia, near the city of Anapa (Krasnodar region) were carried out. First, an introductory survey was conducted, which identified key respondents for deep survey. Beside it, the typical respondents were surveyed on issues relating to the preparation/production and consumption of kvass.
Among the respondents were researchers of local history museums, workers of libraries, municipal officials and representatives of urban and regional Cossack communities. As a result, information was collected regarding the history, traditions and culture of preparation and consumption of kvass in the region.

Street kiosks of kvass, southern Russia.

The Black Sea region of Russia is not the native region of kvass making. The production and consumption of kvass began here in the second half of the 19th century, brought there by Cossacks of the Kuban Cossack Army.
Subsequently, these traditions were reinforced by settlers from the central provinces of Russia (Kursk, Voronezh and others), who brought with them into the newly formed settlements – stanitsas (Cossack villages) - recipes and cooking techniques of kvass, specific to their native places.
With the development of new settlements, traditional recipes evolved over time due to the fact that the species composition of local flora differed from the previous places of residence of migrants. Kvasses were enriched by the local plant raw materials, particularly adding juniper to the kvass, which is widespread in this region.

Kvass selling in touristic resorts.

Traditional kvass continued to be prepared from rye bread dry crusts, and in cases of its shortages also dry crusts from wheat bread started being used.
In order to achieve the color characteristic of rye bread kvass, in some cases the addition of caramelized sugar during preparation also started.
Traditions of the home kvass making are still alive. However, the survey revealed a variety of age groups different attitude to home preparation and use of kvass.
Currently kvass at home is usually prepared in families where children are growing up. In families where the children grew up and went away, home kvass is still prepared mainly in cases when the whole family is getting together. Young people who did not want to put time and energy to cook a home kvass are willing to buy industrial produced kvass.
Special demand for kvass occurs during summer, when the population of the Black Sea region, which is a resort area, is sharply increasing by people coming to rest and for medical treatment from other regions of Russia.
In addition, as a result of interviewing of the Black Sea coast residents it was found that in recent years the structure of consumption of soft drinks had changed. More and more consumers are giving preference to kvass instead of others soft drinks. As a consequence, in the last three years kvass industrial production has increased dramatically.
In the resort area adjacent to Anapa, the presence of several major producers of kvass was detected. Their products are sold in many places, including those visited by tourists. In particular, kvass is selling in many specialized kiosks (see photos). Unlike home-made kvass, for which starting raw material are dry crusts, industrial scale kvass is prepared mainly from kvass wort concentrate.
Home-made manufacturing however does not lose its positions, and it is carried out not only for self consumption, but also for sale (see photos).

A pancake bar, where home made kvass is sold.

The overwhelming majority of respondents indicated that they are willing to drink kvass because it is an excellent thirst-quenching and refreshing drink. In rare cases, respondents indicated that they drink a kvass because of its health-promoting effects.

We're on the Web!
See us at:
http://www.basefood-fp7.eu/

Traditional Romanian dishes highlights

Carmen Costea, Adrian Vasile (ASE)

In a recent effort to identify and at the same time confirm the presence of certain traditional dishes in the official recordings of Romanian cuisine, the ASE Basefood team organized an event involving project members and students.
By the efforts of ASE in collaboration with the State National Archives, recipes, menus and announcements crafted and used by local restaurants from the beginning of the twentieth century were obtained and presented in a special gallery.
The interest of the ASE Basefood team was to highlight the traditional dishes, included in the 30 food items in the Basefood documented files that could be identified in the archive documents, thus underlining the proof of their existence, use and appreciation by the middle and upper classes during the first decades of the previous century.
Through a process of detective work the following Romanian traditional foods were identified as ingredients or actual dishes in that period's menus:

- As ingredients: garlic, onion, bors, celery, corn flour, carrots, dried fruits, nettles and prune jam;
- As food items: polenta, prune jam, vegetable dish, dried fruits, celery dish.

Of these, the items that were the most common on the menus and special events recipes were the polenta (mamaliga), the vegetable dish (mancare de verdeturi) and the dried fruits (fructe uscate).

Mamaliga (polenta). Appears as a common substitute of bread in many of the menus, including those of restaurants where the royal family of Romania celebrated certain events. It can be identified in lists of foods contained in the menus for lunch and dinner, on ordinary days as well as for special occasions. The abundance of documented evidence for its use proves the fact that at the start of the previous century, polenta was viewed as a noteworthy dish in mainstream restaurants, not merely confined to the diet of the peasants.
Another noteworthy aspect is that the recipe for making mamaliga has remained the same, the process being identical to that appearing in the households of the land working class.

Mancare de verdeturi (vegetable dish). Can be identified in the menus of the second and third decades of the last century, being served on many occasions, but most commonly in periods of religious fasting. The ingredients for its preparation are seen to vary from one document to another, yet the most common vegetables appearing are: celery, onion and carrots.
The presence of this dish in the great restaurants of Bucharest and other major cities proves that it was not used solely in the countryside as a necessity induced by the lack of other foods, but rather as a special item that helped to diversify the diet of urban inhabitants.

Fructe uscate (dried fruits). These fruits represented both ingredients for special deserts and pies as well as distinct dishes on the menus of the time. The fruits that were more commonly used were the prunes, the cherries and the sour cherries. They were served as deserts and sometimes as appetizers with certain drinks.
This work was organized by the ASE Basefood team coordinated by Prof. Costea and was based on the previous efforts of the Faculty of Commerce through the Department of Tourism and Geography and the Romanian State Archives. The results serve as further proof for the selection process of the 30 Romanian food items included in the project and also of their consistency and continuity in the menus and diets of mainstream Romanian cuisine.

Analytical and microbiological characterisation of prioritised foods

Mineral and trace elements content of Black Sea Area traditional foods

Tânia Gonçalves Albuquerque, Ana Sanches-Silva, Helena Soares Costa (INSA) and Paul Finglas (IFR).

In the last years great attention has been devoted to the health effects of minerals and trace elements. They are essential for biological processes and play a vital role in normal growth and development. Low intake or reduced bioavailability of minerals may lead to deficiencies, which causes impairment of body functions.
The aim of this study was to produce new analytical data of sodium (Na), Zinc (Zn), Iron (Fe) and Selenium (Se) content in traditional foods in order to highlight their potential positive health effects. Zinc is essential for growth and development, neurological function, wound healing and immunocompetence. Iron functions as a component of a number of proteins, including enzymes and haemoglobin.

Table 1. Dietary Reference Intakes (DRIs). (Values represent Recommended Dietary Allowances or Adequate Intake for minerals and trace elements).

Element	DRIs	
	Male	Female
Sodium	1.5 (g/day) [a)]	1.5 (g/day) [a)]
Zinc	11.0 (mg/day)	8.0 (mg/day)
Selenium	55.0 (µg/day)	55.0 (µg/day)
Iron	8.0 (mg/day)	8.0 (mg/day) 18 (mg/day)[b)]

[a)] *Adequate intake*
[b)] *Premenopausal women*

Food and Nutrition Board, Institute of Medicine, National Academies, available online, Accessed March 01, 2012).

Iron deficiency is the most common and widespread nutritional disorder in the world. Selenium is essential for the defence against oxidative stress and regulation of thyroid hormone, and the reduction and oxidation status of vitamin C and other molecules. Also, some studies have reported positive association between sodium intake and stroke, and when sodium intake is reduced, blood pressure is also lower.
Table 1 shows the dietary reference intakes for adults of Na, Zn, Se and Fe.
All the prioritised minerals and trace elements were analysed in the selected traditional foods from Black Sea Area countries by inductively coupled plasma - optical emission spectrometry (ICP-OES), except for selenium which was determined by graphite atomic absorption spectroscopy. The methods used in this study are accredited

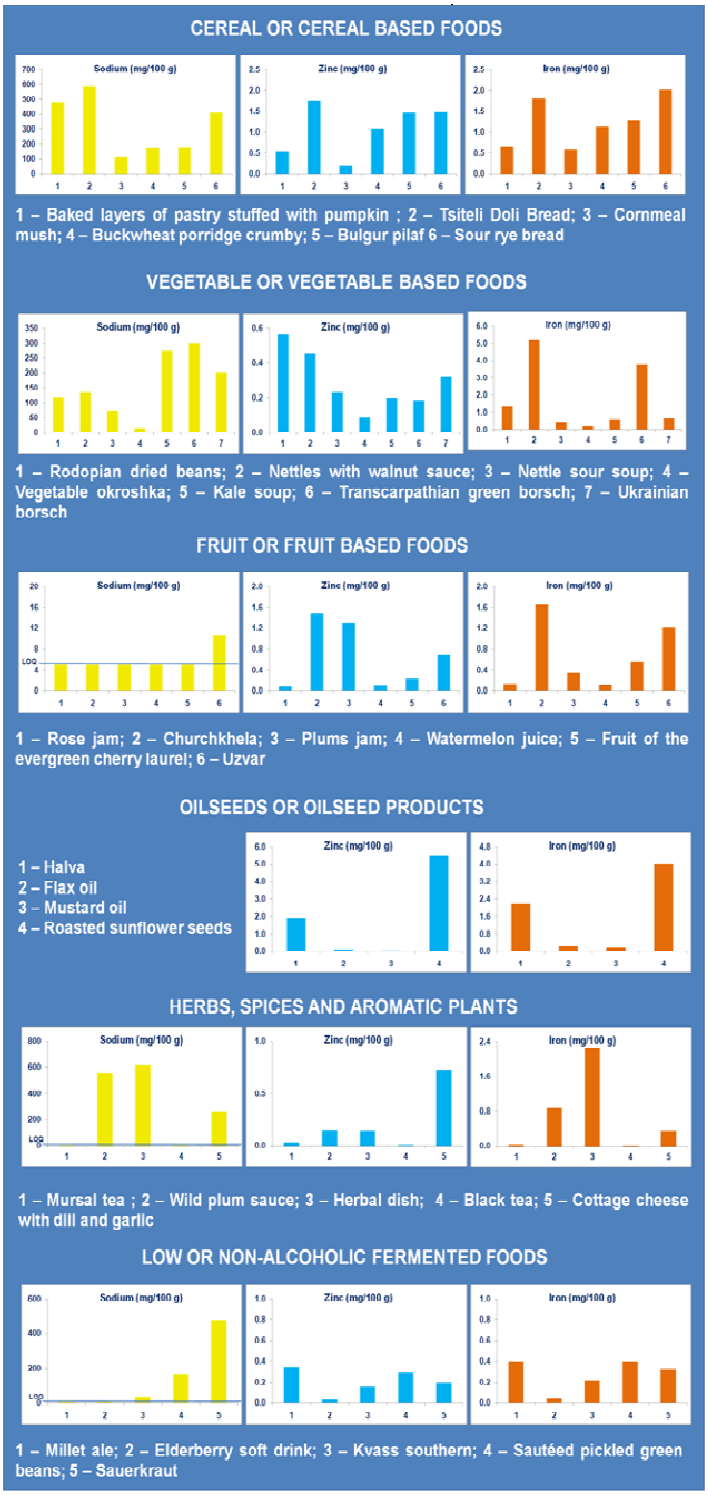


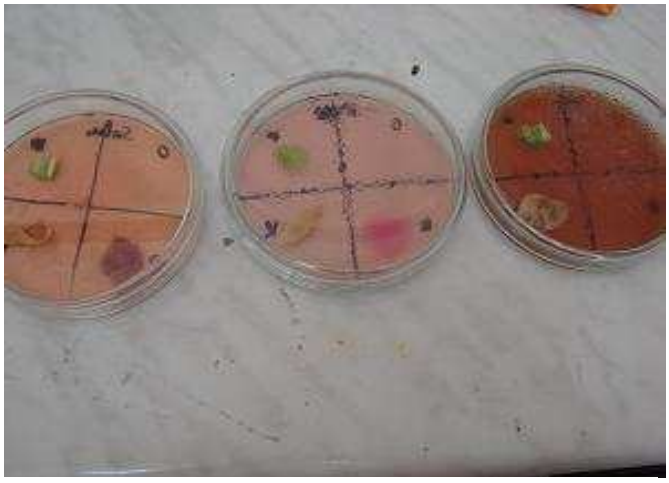
Figure 1. Mineral and trace elements content of traditional foods from Black Sea Area countries.

methods by ISO/IEC/17025 or the laboratory participates successfully in proficiency testing schemes.
The obtained results are shown in Figure 1 and they are given per 100 g of edible portion. For Na the highest level was found in tsiteli doli bread from Georgia (586 mg/100 g) and in herbal dish from Romania (619 mg/100 g). Nettles with walnut sauce from Georgia presented the highest level in Fe (5.2 mg/100 g). Se content of churchkhela and rose jam was higher than 31.0 µg/100 g. Roasted sunflower seeds had the highest Zn content (5.47 mg/100 g). Great variability of minerals content was found in the analyzed traditional foods from Black Sea Area countries. Careful assessment is required in order to evaluate the outcomes that these foods might have in human health taking into account the recommended daily intakes of minerals.

Micro-organisms of traditional foods characterised by Uzhhorod team

Ivan Kutchak, Olga Levchuk, Yuriy Rusyn, Nadiya Boyko (UZHNU)

37 traditional dishes Ukrainian, Russian, Bulgarian, Georgian and Turkish traditional foods and 110 major plant raw material samples were analysed for contamination with three groups of bacteria: beneficial, potentially pathogenic and food borne pathogens. Dr. Tana Sapundzhieva (UFT, Plovdiv) Prof. Osman Hayran and Bike Koçaoglu (YEDITEPE, Istanbul), Prof. Dmitry Karpenko (MSUFP, Moscow) provided samples and organised local pre-analytical work.
Ivan Kutchak student of faculty of medicine at the Uzhhorod National University visited the Bulgarian team in August; at the same time Ing. Yuriy Rusyn had tested the samples, raw material, plants components and dishes in Russia. Dr. Olga Levchuk visited the food technology department in Turkey, YEDITEPE, in September, 2011.
All the isolated microorganisms have been further identified and studied in the microbiological laboratory of the faculty of medicine at Uzhhorod National University, with the participation of research fellows Viktor Petrov and Viktoria Bati, two PhD students of BaSeFood project. Because of the complexity of the task more people was involved to complete it in time:
Anzhela Dolgikh, Ivan Kutchak, Andriy

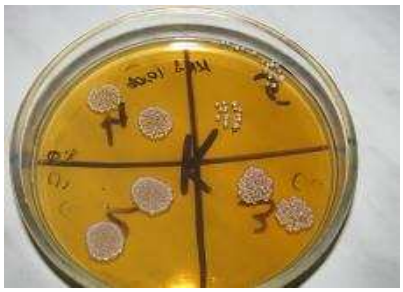


Isolation of microorganisms on different selective media.

Pauk, Vitaliia Dutova, Oleg Solopchuk (students), Larysa Bugyna and Yuriy Rusyn (Ing.); Mariia Mudryk, PhD student and Dr. Olga Levchuk.

Viktoria Bati at work.

A huge variety of bacterial and fungi were isolated from all the tested foods/plans samples, most of which are results of environmental or human contamination during plant collection, washing and preparation of ready-to-eat dishes and beverages.
We were able to find bifidobacteria associated with parsley, dill and celery, but only on samples collected in yards, and isolate lactobacilli species from garlic, cabbage, carrot, beet, cucumber, beans, onion and nettle. This is an important observation since these plants, components of traditional foods of countries from Black Sea region, can be used as source of probiotic microbial strains. The amount of potentially valuable probiotic bacteria was significantly higher in original traditional fermented products (Bulgarian boza, Romanian socata, Russian kvass, Ukrainian sauerkraut and Turkish sautéed pickled green beans),



Cultivation and identification of isolated anaerobes.

from more then 20 strains, participating in fermentation processes were isolated; their biological properties are currently under investigation.
Potentially pathogenic bacteria, represented by "typical" enterobacteria (*Klebsiella*, *Enterobacter*, *Escherichia*, *Proteus*) and *Pseudomonas* were often the dominant micro-organisms. Some of the isolated species are interesting for their possible role in etiology of human opportunistic infections (*Pantoea agglomerans*, *Eikenella corrodens* and *Serratia odorifera* biogroup 1). The coagulase negative staphylococci and enterococci have also been detected.
A highlight of our investigation was that varieties of bacteria and microscopic fungi on plants surfaces collected from home yards or street markets are significantly higher compared to samples from industrial city markets, which are often less contaminated, but sometimes with the more dangerous species of bacteria. Classical food borne pathogens were rarely found: *Salmonella typhi* in Ukrainian sorrel and *Shigella flexneri* ABC in Turkish kale, and *Staphylococcus aureus* in green beans.
Due to the literature evidence about of ability of some opportunistic pathogens to cause nosocomial outbreaks, the contamination of fresh food should be prevented, and not only safety but also quality of the food should be investigated.
The results had been presented by principal investigator of project from UzhNU Prof. Nadiya Boyko at the BaSeFood annual Meeting, Athens, 23-24th February, 2012. They will also be disseminated at the Food Science Symposium, in Kiel, Germany, 22-23 May, 2012, and at the International Pre and Probiotic Conference, Kosice, Slovak Republic, 12-14 June, 2012.

Bioactivity of traditional foods

Major plants in selected traditional foods can modulate the gut microbiota: results of in vitro and in vivo study

Viktor Petrov, Olga Levchuk and Nadiya Boyko (UZHNU)

Methanol or hot water extracts of 36 prioritised foods, 6 lyophilized plant extracts and 45 fresh major plants components were tested for inhibitory effect on 14 pathogenic and 16 potentially pathogenic bacterial strains and for the stimulating effect on 10 beneficial and 11 commensal bacterial/microbial strains.
Only a limited number of them were able to stimulate the growth of beneficial for human health bacteria in vitro. For example Georgian plum sauce demonstrated strong stimulating effect on association of *Bacillus subtilis* and *B. licheniformis*, which are construct commercial probiotics. Pumpkin, cumin tea and fresh (green) cabbage juice caused significant increase of lactobacilli (*L. salivarius*), while sauerkraut stimulated *L. acidophilus*.
Buckwheat porridge crumby, a traditional Russian dish, modulated the commensal gut micro-organism *Morganella morganii*, and local cherries were able to stimulate two strains of tested microscopic fungi: *Saccharomyces cerevisae* and *Candida albicans*.
Other positive results indicate that tsiteli doli bread (Georgia), socata (Romania), garlic, grape, boza (Bulgaria), currants, blue berries, cranberries, bread kvass (Russia) can inhibit the growth of very dangerous bacterium as methicillin resistant *Staphylococcus aureus* (MRSA: stains were isolated from patients in clinic as agent of nosocomial diseases).

Viktor Petrov in the lab.

Ukrainian sauerkraut and cranberries, Georgian plum sauces red and green, tsiteli doli bread, Romanian socata, Bulgarian boza and tomato juice and Russian okroshka can kill *E. coli* EPEC, recently reported as etiological factor of toxic outbreak originated from green part of plants in Germany.

Georgian plum sauces, tsiteli doli bread with makhobeli and nettles with walnut, Bulgarian boza and rhodopian/smilyan bean soup, Ukrainian local cherries and elderberry flowers were active against *Salmonella* and *Shigella* clinical isolates obtained from patients with toxic diarrhea and dysentery. Finally, Ukrainian local elderberry flowers, cherries, sweet pepper, rye bread and tsiteli doli bread with makhobeli from Georgia, Romanian socata, Russian bread kvass were able to inhibit *Listeria monocytogenes*.
There is an interesting observation of comparison of pro- and antibacterial properties of methanol vs. fresh extracts from the same plants: their effect was similar but methanol extracts act less specifically, for example all the tested methanol extracts had stimulation effect on *Lactobacillus ssp.*, but also on some *Candida ssp.*, while pomegranate and persimmon inhibited *B. subtilis*, dill, nettle, kale, and *Sideritis*, the clinical nosocomial agent *Enterobacter cloacae*, dill acted actively against MRSA, and persimmon against *E. coli*.
A parallel study was conducted to determine whether plant ingredients from the traditional foods are able to stimulate the commensal microbes *in vivo* and to inhibit potentially pathogenic strains.
The effect of methanol extracts of food plants (dill, kale, persimmon, *Sideritis*, pomegranate, and nettle) on intestinal microbial coenoses was evaluated by detection of quantitative changes of key gut microbes on mouse model.

Olga Levchuk electrophretically analysing mice feces.

It was shown that the dill extract inhibited two *Enterococcus* strains (*E. faecalis* and *E. faecium*), *Klebsiella pneumoniae* and *Lactobacillus spp.*, and stimulated *Bifidobacterium bifidum*. Bifidobacteria were also stimulated by the kale extract, but in this case there were no significant effects on any of the other tested gut microbiota. The nettle extract caused a non-specific stimulation of key representatives of intestinal microbes on day 3, whereas by days 14 and 24 all the indices were approximately equal to their initial levels (for *E. coli* and *K. pneumoniae*), but lactobacilli and bifidobacteria were both dramatically reduced. Persimmon was the only extract able to specifically stimulate lactobacilli, while pomegranate extract specifically stimulated bifidobacteria. The *Sideritis* extract effectively inhibited *K. pneumoniae* and commensal *E. coli*, and also induced a statistically significant increase in bifidobacteria and a moderate but non-significant increase in lactobacilli. So we were able to prove that plant components of traditional foods and fermented products rich in beneficial bacteria can be used as sources of potential probiotic strains. Plants are able to specifically modulate gut microbiota in a manner that is similar to prebiotics.
Finally in order to clarify the mechanisms by which selected plant extracts can activate the human immune response we proposed to use modern dendritic cell ("professional" antigen-presenting cells) model. Our hypotheses is based on the fact that these cells derived from human blood monocytes can be specifically stimulated by direct exposure to different plant extracts, either with pro and anti-inflammatory effect. This can be detected via the profile of self produced cytokines by means of ELISA technique.
The ability of selected food extracts to modulate and regulate host immune defence will be additionally detected and defined on BALB/c and SCID mice.

Black Sea area plant food bioactives and endothelial function

Mark Woodcock, Paul Kroon (IFR)

What is endothelial function?

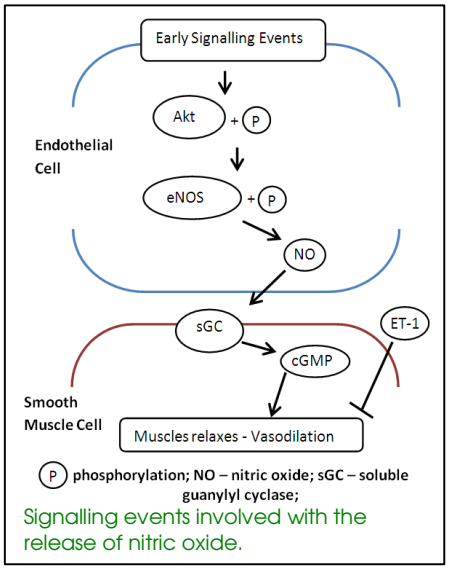
The research at IFR is focussed on establishing the potential for plant extracts and bioactives to affect endothelial function, which is a good measure of the 'healthiness' of the arteries. One important element of endothelial function is vascular tone, which is the result of a balance between vasodilating factors such as nitric oxide, which induce arterial smooth muscle relaxation, and vasoconstricting factors such as endothelin-1, which induce smooth muscle contraction.

How were effects on endothelial cell function assessed?

At IFR, we have measured vascular endothelial responses in cultured cells. The cells used, human umbilical vein endothelial cells, are widely used to study the effects of drugs, bioactives, and flow/sheer stress on the endothelium.
IFR researchers have measured the following:

- eNOS protein
- phosphorylated eNOS (phosphorylation activates eNOS → *p*-eNOS)
- phosphorylated Akt (phosphorylation activates Akt → *p*-Akt)
- Rate of cyclic GMP production
- Endothelin-1 (ET-1) protein

IFR researchers analysed the response of these signalling molecules after dosing endothelial cells with bioactive-rich extracts prepared from plant material supplied by other partners in the BaSeFood project. Dill, nettle and sideritis were supplied by the Institute of Medical Research (Partner 11) in Serbia, who also prepared the bioactive-rich extracts of each of these. Yeditepe Univeristy (Partner 13) in Turkey supplied kale and persimmon, while Elkana (Partner 10) in Georgia supplied quantities of pomegranate, and IFR researchers then prepared extracts of these plants.
Out of the extracts tested, strong, positive effects from the pomegranate and persimmon extracts were seen, suggesting that compounds they contained were able to increase nitric oxide production in endothelial cells.



Signalling events involved with the release of nitric oxide.

Which compounds are responsible for plant bioactivity, and can we learn more about their effect on cell signalling?

Work at IFR is currently underway to analyse the content of the plant extracts. A number of anthocyanins, responsible for bold red/purple colours, were found in the extract prepared from pomegranate arils. They are also found in much smaller quantities in the nettle extract, likely from the plants stems. Other flavonoids such as quercetin glycosides were found in the nettle, dill and kale extracts. Like other cruciferous vegetables, the kale extract also contains a number of glucosinolates. Persimmon has been shown to be rich in procyanidins, responsible for its astringent, sometimes bitter taste.

Research will continue, to provide further identification of the bioactive compounds contained in each of the extracts, as well as the quantities of each present.

Persimmon.

To understand the bioactivity induced by the pomegranate extract, researchers at IFR are currently seeking to identify key bioactive compounds contained in the extract, through separation of the extract into fractions containing major chemical classes of bioactive compounds.

By applying these fractions, and key compounds identified in each, to the same experiments the extract was used in, we hope to pinpoint the compounds responsible for extract's bioactivity.

The means by which these compounds mediate their activity is likely to be complex. Researchers are currently investigating two key suggestions as to how pomegranate's bioactivity is mediated in the vascular endothelial cell model. It is then hoped that this will give a better understanding as to how the active components of pomegranate mediate their bioactivity.

Bioactive-rich arils of pomegranate.

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Black Sea Area traditional plants attenuate cardiovascular risk – potential effects on disturbed platelet function of subjects with metabolic syndrome

Sandra Konic-Ristic, Marija Ranic, Nevena Kardum, Filip Stojanovic, Maria Glibetic (IMR)

The work of BaSeFOOD partner from Serbia is mainly focused on the research of Black Area traditional plants potential to attenuate cardiovascular risk by modulation of disturbed platelet function in subjects with metabolic syndrome.

Polyphenols are one of the most important group of biologically active food compounds, based on their presence in human diet and their effects on health promotion and prevention of the chronic diseases. A great number of scientific data also showed that they can be effective in modulation of platelet function.

The precise mechanism of their action regarding platelet function is not yet fully understood but it is hypothesized that it could be associated with the effect on cellular oxidative status and molecular interactions with platelet agonists.

Activities of the research team of the Institute for Medical Research were conducted within three consecutive phases.

First phase of the study was completed in October, 2011, and the main aim of this phase was to investigate:

• platelet activation status of 100 participants recruited based on the presence of different CVD risk factors (patients with metabolic syndrome).

These results were used for the further recruitment of participants for long term intervention.

• ex vivo effects of the acute consumption of four different traditional plants: nettle, dill and sideritis in the form of water extracts or pomegranate, in the form of fruit juice on activation status of platelets (expression of activation markers and aggregation with blood cells) compared with the control

In the second phase of the study we investigated the effects of larger panel of extracts of polyphenols-containing plants (nettle, dill, sideritis, kale pomegranate, persimmon), major polyphenolic compounds present in these extracts (quercetine, ellagic acid) and their major metabolites on the activation status of platelets of healthy individuals and patients with metabolic syndrome in basal conditions and after exposure to various agonists (ADP or arachidonic acid).

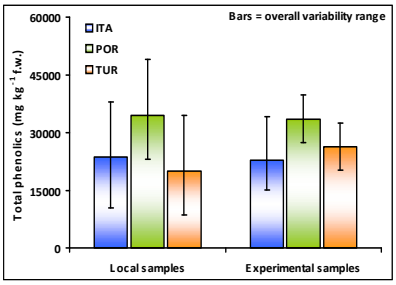
Based on the results of first and second phase of the study, but also on the findings of other BaSeFOOD partners from Institute for Food Research, UK and UZHNU from Ukraine, we selected pomegranate as the food to be tested within the study of long term consumption. We recruited 50 participants; randomly allocated within two groups, the intervention and the control, and the dietary intervention consisted of daily intake of 250 ml of pure pomegranate juice, during 6 weeks. To date, all participants started the intervention that will be finished by the end of May. Additional parameters that are monitored are traditional biomarkers and risk factors (weight and other anthropometric parameters, lipid status, glucose levels, blood pressure), inflammatory and oxidant status and antioxidant defense. Based on preliminary data, pomegranate is a traditional plant that of Black Sea Area with great potential in attenuation of cardiovascular risk, based on its effects on disturbed platelet function, as well on other risk factors.

Analytical characterisation of raw materials

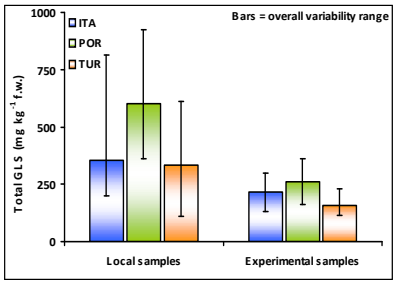
Local kale populations

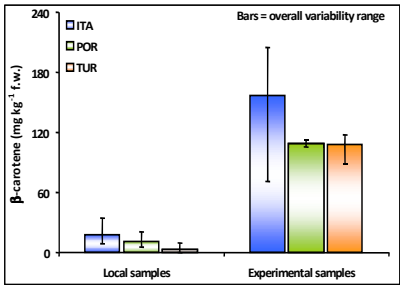
Federico Ferioli, L. Filippo D'Antuono (UNIBO)

The analytical determination for the characterisation of phytochemicals from a total of 41 kale (from Turkey, Italy and Portugal, and either sampled from local crops or from an experimental trial held in Italy, (see also BaSeFood newsletter, issue 3) was almost completed.

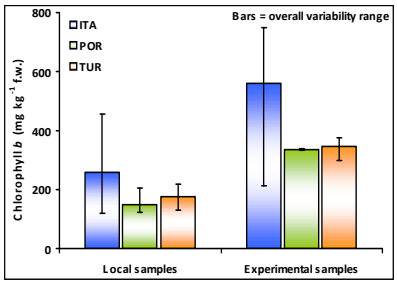


Interesting variability was detected between country of origin and growing conditions. Basically, total phenolics were hardly affected by the growing





situation, and were more abundant in the Portuguese samples. Total glucosinolates (GLS) content, as an average, was higher in the samples from local fields and again, in Portuguese samples. Interesting variation was detected also in GLS composition, with the Italian population slightly richer in glucobrassicin.



Carotenoids and chlorophyll were clearly more abundant in the samples from the experimental field, with clear connection with the higher soil fertility level. Dark leaved Italian populations had the highest contents of these components.

Ancient wheat populations

Elisa Giambanelli, L.Filippo D'Antuono (UNIBO)

During 2010-2011 a field experiment was set up in Bologna, in order to obtain material in homogeneous environmental conditions, on which carry out analytical determination of phytochemicals.

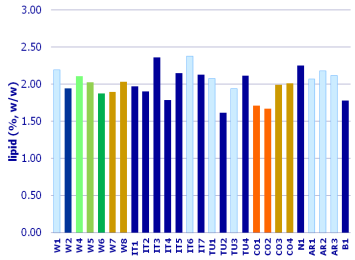
Different growth habit revealed by spring sowing.

27 samples were used:
- 7 from Georgia, belonging to the species *Triticum. monococcum, T.turgidum subsp. dicoccum, T. timopheevi, T. palaeo-colchicum, T. macha, T. turgidum subsp. durum;*
- 7 from Italy: *T .turgidum subsp. dicoccum, and T. monococcum;*
- 4 from Turkey: *T .turgidum subsp. dicoccum, and T. monococcum;*
- 4 from Armenia: *T .turgidum subsp. dicoccum;*
- 4 from Bulgaria: T .*turgidum subsp. dicoccum;*
- 4 commercial durum and bread wheat varieties, used as controls.

Harvest of experimental plots.

The trials were sown in two dates: fall (indeed very late, due to adverse climatic conditions), and late winter. This allowed to discriminate between winter habit and spring habit populations. All emmer and einkorn wheats were spring types, whereas some of the other Georgian species were winter types. Harvest took place in July, 2011. The product was cleaned and preserved in adequate conditions until analysis. Analyses 0of lipids, phenolics and carotenoids will be finished in spring 2012, and are revealing interesting differences among types.

A phase of lipid extraction.



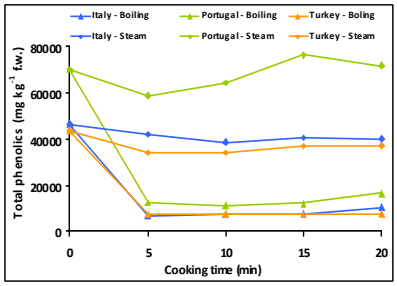
Total lipid profile of the winter sown populations.

Effects of processing on bioactive retention

Kales

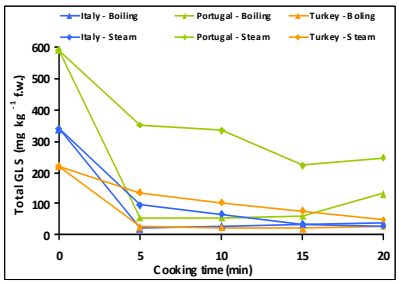
Federico Ferioli, Elisa Giambanelli, L. Filippo D'Antuono (UNIBO)

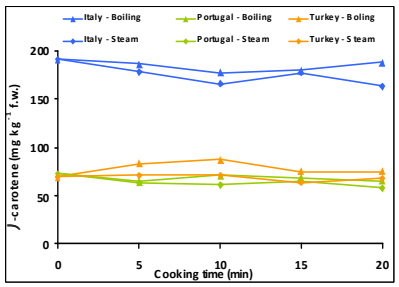
A cooking experiment was carried put using three local kale population form Italy, Portugal and Turkey, grown in Cesena (see also BaSeFood Newsletter, issue 4).



Effect of cooking on the pattern of total phenolics in kale leaves.

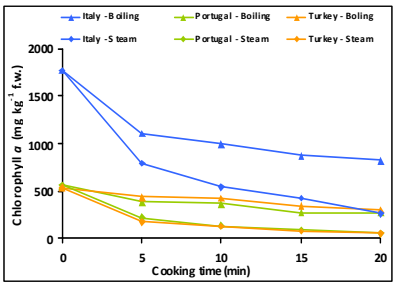
The samples were water or vapour cooked for times of 5, 10, 15 and 20 minutes. Phenolics, glucosinolates, carotenoids and chlorophylls were analysed.
Data on the evolution of these components, that will allow calculating retention factors, were for the first time obtained for this crop.





Effect of cooking on the pattern glucosinolates and β-caotene in kale leaves.

The pattern of the four classes of components was of course different, depending on their nature, but not only. Water soluble phenolics sharply decreased with water cooking, but were almost unaffected by vapour cooking.



Effect of cooking on the pattern of chlorophyll a in kale leaves.

Water soluble glucosinolates decreased with both water and vapour cooking; in this latter case, their decrease is due to degradation. Moreover, relative decrease was higher in the Italian samples, richer in indolic glucosinolates, than in Turkish and Portuguese samples. Among non-polar components, carotenoids were almost unaffected by cooking, whereas chlorophylls decreased, with a parallel increase of their degradation products (not shown).

Oilseeds

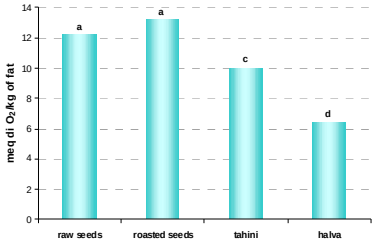
Federica Pasini, L. Filippo D'Antuono, M. Fiorenza Caboni (UNIBO)

This was a collaborative experiment between UFT, Plovdiv, and UNIBO.
For seven oilseeds and fatty nuts , the cycle of processing from raw seeds to halva was considered:

1. **Peanuts**
2. **Almonds**
3. **Walnuts**
4. **Hazelnuts**
5. **Sesame seeds**
6. **Sunflower seeds**
7. **Pumpkin seeds**

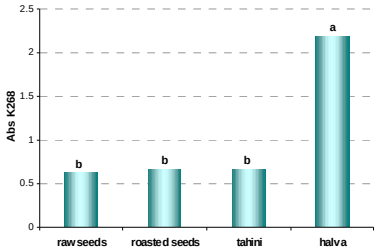
Total fats, the status of lipid oxidation, sterols, tochols and phenolics were determined at each step, and are being determined during shelf life of final products, for a period of twelve months.
The samples are in course on analysis and data are being processed.

Roasted seeds → **Tahini** → **Halva**



Effect of processing on average peroxide values of 7 oilseeds and nuts.

It can be noted that processing in some steps reduced the peroxide number of raw material, but final step, from tahini to halva, highly increased the content of conjugated trienes.



Effect of processing on average conjugated trines of 7 oilseeds and nuts.

Survey-based consumers and stakeholders investigations

A survey on health promoting traditional foods concepts

Annachiara Berardinelli, L. Filippo D'Antuono (UNIBO), on behalf of all participants

This is a part of an ample collaborative work carried out by ASE, ELKANA, MSUFP ONAFT, SPES/GEIE, UFT, UNIBO, UZHNU, YEDITEPE.
It was a survey-based investigation aimed at defining the consumers and experts ratings, and defining the potential of different combination of concepts belonging to the sphere of traditional foods with health promoting properties.
Three different concept levels were considered: carriers (foods), health claims and functional ingredients, which categories were defined a priori, during focus group sessions.
"Mini concepts" were obtained as binary combinations. As an example:

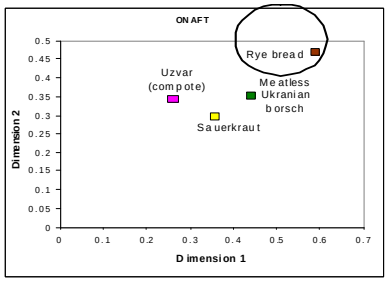
- carrier / health claim mini concept: a whole mal bread that reduces the risk of heart diseases
- carrier - functional ingredient mini concept: a whole mal bread rich in antioxidants
- functional ingredient - health claim mini concept. (a food rich in) antioxidants that reduces the risk of heart diseases.

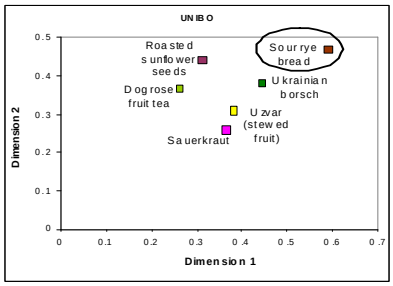
These mini concepts were rated by consumers and food experts (marketing experts, nutritionists and food technologists) as mini-concepts or "binary combinations".
Market experts seem to better reflect consumers opinion than nutritionists and food technologists.
"Spice or herb" in combination with health claims "strengthens the natural defence of body", "reduce certain types of cancer" and in combination with functional properties "rich in antioxidants" and "rich in vitamins" were the most potential carrier / health claims and carrier / functional ingredients mini-concepts.
For the mini-concept functional ingredients-health claims, high potentiality was observed for "foods rich in antioxidants" in combination with "reduces the risk of certain types of cancer", "reduces the risk of heart diseases" and "keeps your arteries healthy" and for "foods rich in vitamin" in combination with "strengthens the natural defence of the body against frequently occurring diseases like a cold", "reduces the risk of certain types of cancer" and "reduces the risk of heart diseases".

An analysis of traditional food perceptions of populations of the Black sea area

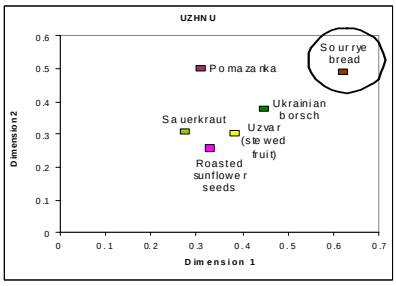
Annachiara Berardinelli, L. Filippo D'Antuono (UNIBO), on behalf of all participants

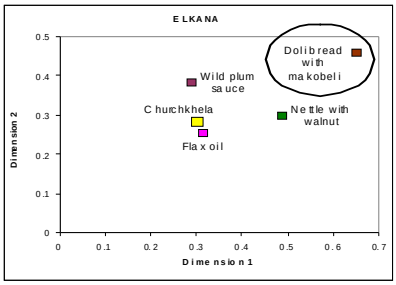
This part of the was aimed at individuating the attitude of Black sea area consumers towards traditional foods, in relation to food characters belonging to different spheres of perception.
4-8 traditional dishes were selected for each country. A total of 25 of attributes belonging to the following spheres of perception were individuated:
• sensorial (e.g.: is tasty)
• health promoting (e.g. is low in calories)
• convenience (e.g. is easy to prepare).
The foods were rated on a binary scale by consumers of each participating country and by Ukrainian migrants in Italy. The concept of a generic traditional food was also rated for the same attributes, on an ordinal scale.
A principal component analysis substantially confirmed the validity of the three a-priori defined spheres of perception, although revealing some sub-dimensions.
The perceptual map obtained by multi dimensional scaling of the rating of individual foods revealed interesting patterns. As an example, in the figures below, it can be seen that rye bread (rated in three contexts: Ukraine, Russia and Italy) or other cereals foods (e.g. the Georgian Tsiteli doli bread) are all placed in the upper right side of the first two common space coordinates from MDS, associated with health promotion perception.





Layout of traditional foods in the space of the first two multi dimensional scaling common space coordinates.





Layout of traditional foods in the space of the first two multi dimensional scaling common space coordinates.

BaSeFood dissemination

BaSeFood final meeting and International traditional and street food meeting, Cesena, 3-6 October, 2012

ANNOUNCEMENT

Traditional food international (TFI-2012)

Traditional foods: from culture, ecology, diversity, to human health and potential for exploitation

JOINTLY WITH

Street food seminar

An international forum on street food aspects and perspectives

The end of BaSeFood program will be an occasion of interacting with local stakeholders, in the organisation of a comprehensive international traditional and street food event.
The program will include:

- Wednesday October 3. BaSeFood final meeting (restricted to Consortium members).
- **Thursday 4 and Friday 5 October: Traditional food international**
  An international meeting devoted all aspects of traditional foods
  The program will include: invited speaker presentation, highlights of BaSeFood results, reports from the coordinators of recently funded EU programs about traditional foods
  The meeting is open to offered contributions!
- **Friday 5 October, afternoon. Street food seminar**
  An international forum about street food definition and facts
- Saturday 6 October, morning. Dissemination events, centred on Black Sea region traditional foods within the street food fair of Cesena.

The event web site is being activated at: tfi-2012.com

Find there all details about program, venue, registration and modalities to submit your contributions.

Presentations at scientific meetings

- *Danesi F., Pasini F., Mudryk M., Kocaoglu B., Karpenko D., Kapreliants L., Jorjadze M., Stroia A., Alexieva I., Caboni M.F., D'Antuono L.F., Bordoni A.* Bioattività degli alimenti tradizionali del Mar Nero: primo screening del possibile ruolo nella prevenzione cardiovascolare. Annual Meeting of the Italian Society of Human Nutrition on "Nutrition and cardiovascular risk". October 12-13, 2011, Naples, Italy. (poster presentation, in Italian).
- *Soares Costa H .* Traditional foods: a contribution to biodiversity and sustainable diets. 4th International Congress on Food and Nutrition and 3rd Safe Consortium International Congress on Food Safety. October 12-14, 2011, Istanbul, Turkey. (oral presentation).
- *Soares Costa H., Sanches-Silva A., Nascimento A.C., Santiago S., Trichopoulou A., D'Antuono L.F., Alexieva I., Bugyna L., Fedosova K., Hayran O., Karpenko D., Kilasonia Z., Stroia A.L., Finglas P.* Traditional foods from the Black Sea Area countries: minerals and trace elements content. 4th International Congress on Food and Nutrition and 3rd Safe Consortium International Congress on Food Safety. October 12-14, 2011, Istanbul, Turkey.
- *Soares Costa H., Albuquerque T., Fontes T., Mota C., Vasilopoulou E., D'Antuono L.F., Alexieva I., Hayran O., Kapreliants L., Karpenko D., Kilasonia Z., Pauk A., Stroia A.L., Finglas P.* Proximate composition of traditional vegetable and fruit based foods from Black Sea Area countries. 4th International Congress on Food and Nutrition and 3rd Safe Consortium International Congress on Food Safety. October 12-14, 2011, Istanbul, Turkey.
- *Woodcock M., Needs P., Kemsley K, Dainty J., Mithen R., Kroon P .* Early changes in vascular endothelian cell signalling in response to human quercetin methabolites: a phospho- array study. 5th International Conference on Polyphenols and Health, October 17-20, 2011, Sitges, Spain. (poster presentation).
- *Albuquerque T.G., Sanches-Silva A., Finglas P., Vasilopoulou E., Trichopoulou A., Alexieva I., Fedosova K., Kilasonia Z., Kocaoglu B., Koval N.V., Krechetnikova A., Pamfilie R., D'Antuono L.F., Soares Costa H.* Lipid profile of traditional foods from Black Sea Area countries. 4ª Reunião Annual PortFIR, October 27, 2011, Lisbon, Portugal. (poster presentation).
- *Soares Costa H., Albuquerque T., Sanches-Silva A., Vasilopoulou E., Trichopoulou A., Alexieva I., Costea C., Fedosova K., Hayran O., Kilasonia Z., Kolesnov A., Koval N.V., D'Antuono L.F., Finglas P.* Nutritional composition of traditional foods from Black Sea Area countries. 4ª Reunião Annual PortFIR, October 27, 2011, Lisbon, Portugal. (poster presentation).
- *Naska A., Sanches-Silva, A. Gonçalves Albuquerque T., Soares Costa H., Finglas P., Konic-Ristic A., Glibetic M. D'Antuono L.F., Trichopoulou A., on behalf of the BaSeFood Black Sea area partners .* Dietary data and mortality patterns in countries of the Black Sea region. 11th European Nutrition Conference, October 26-29, 2011, Madrid, Spain. (oral presentation).
- *Vasilopoulou E., Dilis V., Finglas Soares Costa H., D'Antuono L.F., Trichopoulou A., on behalf of the BaSeFood Black Sea area partners.* Traditional foods of Black Sea Countries. 11th European Nutrition Conference, October 26-29, 2011, Madrid, Spain. (poster presentation).
- *Boyko N., Dolgikh A., Buhyna L., Kutchak I., Mudryk M .* Traditional foods of plant origin maintain the mucosal immune response via its prebiotic effect on gut microbes . 11th European Nutrition Conference, October 26-29, 2011, Madrid, Spain. (oral presentation, abstract).
- *Mudryk M., Buhyna L., Petrov V., Pauk A., Dolgikh A., Kutchak I., Boyko N.* Key-microorganisms in food quality / food safety of traditional dishes and drinks prioritised within BaSeFood project . 11th European Nutrition Conference, October 26-29, 2011, Madrid, Spain. (poster presentation and abstract).

Editing
Lorenzo Cerretani

We're on the Web!
See us at:
http://www.basefood-fp7.eu/

Information from partners

HHF – HHF communicated that hard copies of deliverable *"Flow charts on the preparation procedure of the composite traditional foods, accompanied if possible by audiovisual material"*, that were printed for internal HHF initiative, are available: info@hhf-greece.gr.

ASE - ***Big Projects - big challenges: catalyst for reinventing societies***. *Scoala Banilor Europeni* (the School of European Money) http://www.structuralfunds.ro/Atelier/Scoala_ed5.html is an experiment started last year under the *Business Incubator Project*, by the House of Europe Eurolink, and created together with several local entities working under the new Stakeholders Platform inside the Danube and Black Sea Regional Network for Social Economic Innovation.

The last meeting of the 14th of April 2012 was dedicated to *Challenges and competition in complex transport logistics. Dr.Carmen Costea had an intervention o*n the smart food logistics, connected to the Basefood project presentation. The list of participants contained representatives of European Parliament, Ambassadors, diplomats, politicians, stakeholders.

ASE - ***FuturICT meeting Romania*** represented the launching of the social-economic Innovation Hub for the Black Sea and Danube area, under the House of Europe and Alternative Sciences Association coordination and the FuturICT Knowledge Accelerator label www.futurict.eu . It was the Black Sea and Danube Basin Social- Economic Platform of Innovation to be considered for a future vision of the robust society. The workshop hosted by the Romanian Academy and the Scientists Club (http://www.houseofeurope.ro/FuturICT/en/futurict.html) took place on the August 26th.2011.. Professor Costea's presentation underlined the Basefood research importance in identifying food traditions and using them as economic engine in promoting a better future of each economy as local knowledge and innovation remain key drivers for sustainable growth and prosperity of each country.

ASE - ***Learning Innovation***. In March 2012, ASE and Alternative Sciences Business Club continued the tradition of the periodic meetings, started this time last December the 5th, with the launch of the book *Life as a tragic optimism*. The purpose of the last meetings was to create o road of values in different fields. Following the initial symbolism appointed with the food exhibitions and continued with a vitamin exhibition, during the last meeting of the 8th of March, the discussions underlined the need of knowledge in the field of food and how to identify values in preserving traditions for emergent countries citizens.

Beyond the presentation of the role of Basefood projects within few books dissemination, the students interest was oriented towards the transition from the success to value and spread out as a ***Contest of Essays entitled Food: from success to real values***. The best three essays were awarded by relevant stakeholders in the field. Photos of the presentations will be available on the Business Club website www.science.ase.ro and youtube. The Business Club activities was enriched with another talk given on the 15th of March by PhD Laura Libardea Vladuca lawyer at Vladuca &Assoc. Involved in our Basefood team, her talk followed by large discussions were connected to the *Dangers of hidden logistics of food and its ingredients*.