



LEGATO

LEGumes for the Agriculture of TOMorrow

Collaborative project

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SEVENTH FRAMEWORK PROGRAMME

THEME [KBBE.2013.1.2-02]

**[Legume breeding and management for sustainable
agriculture as well as protein supply for food and feed]**

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Stakeholder topic meeting report n°4

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1. Focus on the topic chosen

The LEGATO stakeholder topic meetings are related to the task 6.2 - sharing expertise to orientate and evaluate possible levers of improvement offered by the project results.

The fourth stakeholder meeting took place on the 17th and the 18th May 2017 at Wellness Hotel Diana, Velké Losiny, Czech Republic. It was organized by the company Agritec. The programme of the meeting was focused on breeding grain legumes and registration of legume varieties with particular attention to the Czech Republic. It was shown how the use of biotechnology and Marker-Assisted selection could help pea and faba bean plant breeding programs. It was also highlighted that specific cropping systems such as intercropping of legumes with cereals may limit insect pests. A report on the two-year pan-European trials network managed in the WP6 was presented, the methods of analysis of the results emerging was debated.

The meeting was specifically addressed to legume stakeholders, principally breeders, then agricultural, food and feed industries, environmentalists, technicians, administration officers, scientists, etc....

The meeting attracted 71 participants including 62 % from SME, 14 % academic researchers, 7 % policy, 5.6 % industry and 5.6 % of interprofessional bodies, coming from Europe (See figure 1 below). 54 % of the participants belong to a breeding company.

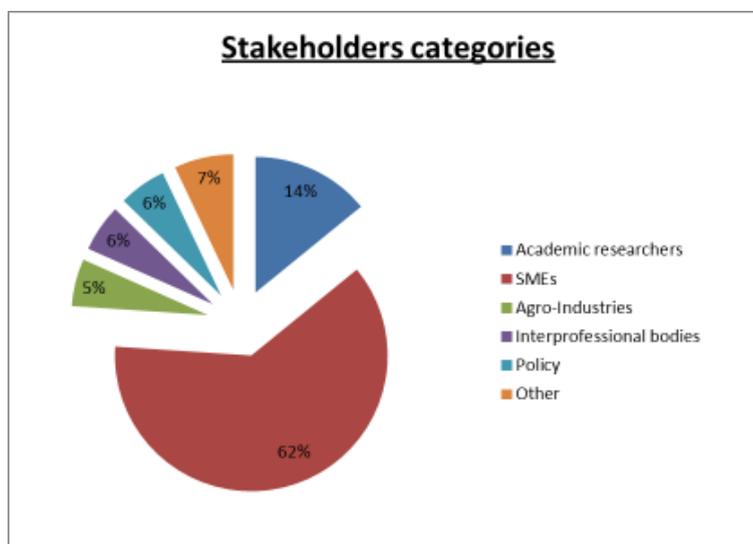


Figure 1: Stakeholder categories

2. Objectives (what we asked of the speakers, why this topic)

The content of the stakeholder meetings was defined in the DOW. Previous stakeholder meetings have concentrated on science, techniques and economy of production; science, techniques and economy of uses. This final stakeholder meeting was synthetic: based on the input of results from the different WPs and of the agro-economic context, definition of adapted ideotypes (genotypes x cropping management systems) for different European regions, and estimation of their potential development, in areas and volumes. Furthermore, there was extensive discussion about the results of the pan-European trials carried out in WP6 and the general question of definition of varieties adapted to different European areas.

3. Short summaries of the presentations

- **Wellcome and commencement – Information about the company Agritec** (Director Dr. Prokop Smirous, Dr Martin Pavelek)

The meeting was introduced by the Director of Agritec (Dr. Prokop Smirous), who welcomed the participants. Then, Dr. Martin Pavelek made a presentation entitled: "AGRITEC, 75 years tradition in agricultural research in Šumperk's region". A short presentation of history of Šumperk was given. It was originally a German city. Then it became a Czech center of textile and industries. This was the origin of flax breeding at the Institute, which also had experience in cattle and plant breeding. The company Agritec, Research, Breeding and Services, Ltd. was established in 1994 and continued in agricultural research, which started in Šumperk in 1942, as a Flax Research Institute. Agritec is divided in 4

departments: Department of Grain Legumes and Technical Crops, Department of Plant Biotechnology, Department of Plant Protection, Department of Economy and Services and is composed of 55 employees. Agritec is engaged in the research of germplasm resources of grain legumes, flax and hemp, genetic-breeding methods and biotechnology of legumes, flax, caraway and rapeseed. In addition, the company is engaged in growing technologies and breeding of peas, beans, lupine, flax, hemp, caraway integrated protection of legumes and flax, including registration experiments with plant protection products. Agritec also represents foreign companies for the purpose of variety testing, production, processing, storage and marketing of legumes and technical crops. Agritec has its own varieties of oil and fibre flax and winter caraway. Agritec produces and commercializes seeds from these crops.

- **Information about LEGATO project** (Dr. Richard Thompson, INRA)

Richard Thompson - LEGATO coordinator - presented the LEGATO structure and the objectives of the project. He focused on what is new in LEGATO and then presented the agenda of the meeting (see Annex 1).

- **Perspectives of cultivation and use of legumes in the Czech Republic** (Presentation of Ing. Martin Liska, Ministry of Agriculture of the Czech Republic)

The growing areas of legumes have increased in recent years in the Czech Republic. They reached 35 633 ha in 2016/2017, with an average yield of 2.37 t/ha, including 26 601 ha of peas (average yield: 2.58 t/ha). According to the FAO, the average annual world consumption of legumes varies from 2 kg to 20 kg per person. In the Czech Republic, it was 2.7 kg in 2014, essentially pea. Exports of peas from Czech Republic (37 818.1 t in 2015/2016) are most directed to Austria, Poland and Germany. Three types of subsidies exist in the Czech Republic: SAPS–Single Area Payment Scheme (500 M€ distributed in 2016), Greening–payment (276M€ in 2016) and aid for the production of protein crops (3.40 billions of CZK in 2016). A fourth aid is for young farmers (136M€ in 2016). Prices were 254€/t for edible peas and 194€/t for feed peas.

- **Activity of Association of Grain Legumes growers and processors in the Czech Republic** (Ing. Jan Prášil, chairman APZL, director of Semo a.s)

The association of Grain Legume growers and processors in the Czech Republic, called APZL, was Founded in 2005 and is composed of 51 members. It provides: informatics, promotion, provision of list of recommended varieties in cooperation with the ÚKZÚZ (for peas since 2007 and for soybean since 2008), co-operation with the institutions (the Agricultural ministry, UKZÚZ, other unions and associations, foreign contacts). 48 professional seminars have been organized. 140 papers were published in the agrarian press. 5 Methodology for Cultivation Practices (co-operation between Agritec s.r.o. and the Ministry of Agriculture of the Czech Republic) and book publications (LEGUMES -cultivation and use in 2009, a cookbook in 2014) were produced. Almost 50 thematic seminars including field days and exhibitions were organized.

Greening has brought a very desirable development of legumes since 2013. But the consequences of prohibition of use of pesticides in greening mode will be: Immediate dramatic decline of the legumes, loss of positive effects, possible shifting of the conditions of use by using catch crops, in particular mustard with a less favorable C: N ratio in the biomass of the catch crop, reducing biodiversity, replacement of legumes with other species -more demanding for treatment, increase in pesticide consumption, increase of the consumption of N (Nitrogen) of fertilizers and increase of CO2 emissions.

Breeders, Agrochemical companies, farmers are members of the association APZL. Membership is inexpensive.

Czech company SEMO s.r.o. was founded in 1994. It is based on more than 50 years tradition of breeding, connected with the former Vegetable Breeding Station. It is engaged in breeding, production and sale of seeds.

SEMO breeds varieties for European conditions: Czech Republic, Slovakia, Poland and Hungary. It focused on different species (green peas, pepper, tomato, cucumber, root vegetables, lettuce, garlic and sweet corn). SEMO is proud of the high quality of its production. Any seed leaving the grounds of SEMO must comply with very high standards. Besides offering high quality seeds, SEMO provides growers with a

functional system of associated services. An advisory service is provided during the crop growth and help to solve any of growing problems.

Different technologies are used to produce seeds: Sortex (optical sorting), improving germination of seeds by effect of low temperature, measurement of chlorophyll content.

Business activities of SEMO are present in different countries: Russia, India, Pakistan,...

Garden legumes have different uses on the market: fresh market (hand harvest), processing (mechanized harvest) (freezing, canning, drying), other uses (resistant starches ..).

- **Czech agriculture pulse breeding** (Pavel Horčíčka, director of breeding, SELGEN, a.s.)

Legumes covered 36 000 ha and wheat 839 000 ha on the total of 2 463 000 ha in Czech Republic in 2016. Wheat yield is around 5.4 t/ha (average 1920-2014) when pea yield reached 2 t/ha (average 1920-2014). Yields were high in 2011: 8,30 t/ha for spring field pea and 8,04 t/ha for winter field pea. The highest yield was obtained with Enduro: 9,54 t/ha in 2010.

3 breeders are present in Czech Republic: AGRITEC, SEMO and SELGEN. SELGEN is a Czech breeding company with the biggest portfolio of crops and the longest tradition in breeding with two plant breeding stations (Chlumec nad Cidlinou and Lužany). At Lužany, is organized the beginning of breeding (maintenance of pulses, flowers, vegetable and fruit trees). The station is well known due to varieties Eso, Menhir and Bohatýr (revolutionary variety of pea marketed on 5 continents). Current activities are dedicated to spring peas, winter barley, rapeseed. At Chlumec n. Cidlinou, breeding is focussed on fodder pea, red clover, faba beans and peas. Since 1997, there is also canola breeding and foundation of „Czech rape consortium“.

Traits that are important for pea breeding are: high yield, lodging resistance, good harvest ability. Disease and pest resistance are also researched (root diseases, complex of Phoma, Ascochyta, and Mycosphaerella, mildew, viruses, aphids) and quality for feed and food (protein content).

- **Testing and registration of varieties of legumes in the Czech Republic** (Tomáš Mezlík, ÚKZÚZ, National Plant Variety Office)

The National Plant Variety Office (NPVO) is constituted by a total staff of 55 persons. The Mission of NPVO is to establish national listing of the plant varieties, to manage Plant Variety Rights Protection, to establish a recommended list of varieties and to provide farmers with the information needed to choose the varieties which will maximize their profitability in the agricultural areas of the Czech Republic.

DUS (Distinctness, Uniformity and Stability of plant varieties) and VCU (Value for Cultivation and Use) tests are organized by NPVO.

For VCU testing, each year, a series of trials is implanted: for example, 7 trials with 9 varieties for winter field pea, 8 trials with 15 varieties for spring pea, 6 trials with 3 varieties for spring faba bean. For field pea, different traits are measured: early vigour (9-1), beginning of flowering (days), flowering period (days), maturity (days), stem length (cm), crop height (cm), lodging during growing period (9-1), lodging before harvesting (9-1), resistance to diseases (9-1) (Virus diseases, Root rot, Powdery mildew, Downy mildew, Grey mold, Leaf and pod spot, Pea rust), yield, TGW (g) and seed size grading (< 6 mm, 6-7 mm, >7 mm), colour stability of seeds (%), protein content (%), starch content (%) and trypsin inhibitor activity (TIU).

The VCU is determined by performance. Earliness x yield x standing ability, resistance to pathogens, quality parameters and other agronomic characteristics are considered. Then, the decision on a variety is based on expert opinion (commission of UKZUZ experts, no index). The variety must be distinct, uniform and stable (DUS). The maintenance must be ensured and a suitable denomination must be given.

- **Effect of intercropping of legumes with cereals on distribution and abundances of insect pests and their natural enemies in crops** (Marek Seidenglanz, Igor Huňady Jaroslav Šafář, AGRITEC)

The work presented aimed to answer several questions:

- 1) Are legumes intercropped with cereals (spring wheat, spring barley, oat) less attractive for insect pests (sucking or chewing pests) than legumes grown as monocultures?
- 2) Do aphid colonies (pea aphid, broad bean aphid) develop differently on their legume host plants when they are intercropped with cereals?
- 3) Do natural enemies attack their prey (insect pests) more frequently and more effectively in plant mixtures in comparison with monocultures?
- 4) Are spring cereals strong competitors for legumes?
- 5) Are spatio-temporal distribution patterns of insect pests and their natural enemies different in legume monocultures than in mixtures?
- 6) Where are tighter temporal and spatial associations between insect pests and their natural enemies found: in crop mixtures or in monocultures?

Trials were conducted in small plots (15 m²) and in large plots (min. 2100 m²; max. 6400 m²) with different kind of crops: Field peas monoculture (100 %), Field peas + Spring wheat (50/50 %), Field peas+ Spring barley (50/50 %), Field peas+ Fababean (50/50 %), Field peas monoculture (50 %), Field peas + Fodder peas (60/40 %), Field peas+ Oat (50/50 %), Field peas + Spring wheat + Fodder peas (33/33/33 %), Field peas + Spring barley + Fodder peas (33/33/33 %), Field peas + Oat + Fodder peas (33/33/33 %).

In small plots, aphids are dispersed differently in monocultures vs. mixtures at a spatial point of view but the results show that legumes intercropped with cereals are not less attractive for insect pests than legumes grown as monocultures, at least not at the beginning of crop colonization by aphids. However, the field pea plants in mixtures become unattractive earlier as a food source for aphids. In large plots, syrphids show higher tendency for aggregation in intercrops, and also predation of aphids diminishes.

For *Bruchus pisorum*, females start egg-laying at sites which are convenient places for oviposition. They concentrate on the early flowering varieties of pea rather than on later flowering pea varieties or pea in association with barley.

Cereal plants in mixtures with legumes have larger leaves and ears with more grains but legumes do not profit from the partnership. (LER index may however be beneficial).

To conclude, there are differences between the results coming from small plot and large plot trials. Cereal plants grown in mixtures with legumes are at an advantage compared to cereals grown as sole crops. Intercrop may reduce the development of some insect pests such as aphids.

- **Legume Biotechnology in AGRITEC (2007 -2017)** (Miroslav GRIGA, AGRITEC)
- **GM approaches in legumes:**

Regeneration systems in vitro have been developed in AGRITEC for: pea, fababean, soybean, flax, white lupin, oilseed rape, caraway, hemp, poppy. Agrobacterium mediated pea transformation was used to introduce resistance to Pea Seed borne Mosaic Virus (PSbMV) in pea plants. A study was conducted to evaluate the risks associated with release of Genetically Modified (GM) peas into the environment. Outcrossing frequency was very low: 0.70% (2005), 0.57% (2006) and 0.72% (2007). These results served to obtain permission for releasing AGRITEC GM-pea lines to field tests in 2010. The effect of GM pea feeding on the growth of model animals (rat) was also studied. All animals were in clinically good healthy state and no differences in development or behavior were recorded.

A small protease inhibitor (36 amino acids) was isolated from the labial glands (producing silk fibre) of the waxmoth *Galleria mellonella* (gmSPI-2). This inhibitor is active against both bacterial, as well as fungal proteases e.g. subtilis in or proteinase K (Nirmala et al. 2001). Gradual increase of mortality of bruchus beetle (*Bruchus pisorum* L.) larvae was observed during their growth and development in pod valves and

ovules/seeds for two pea plant groups: T=plants transformed with gene for proteinase inhibitor gmSPI2; UU=non-transformed and chemically non-treated pea plants (greenhouse, 2010). The differences (statistically significant) appear during the development of larvae in cotyledons.

These techniques can also be used to produce edible vaccines in legume seeds or other heterologous proteins.

- **Non-GM approaches in legumes**

Marker-assisted selection (MAS) was used to introduce a recessive resistance gene to PSbMV and resistance to *Erysiphe pisi*. Recent doubled-haploid technology was used in the breeding programmes of AGRITEC for flax, winter rapeseed and now caraway. It is under research for pea, poppy and hemp.

• **Report on the two-year pan-European trials network** (Jana Poslušná, AGRITEC)

For the task 1 of WP6, a network with field trials with grain legumes was established at 15 locations well distributed throughout Europe (within a wide perimeter including UK, Estonia, Serbia, Greece and Portugal) on selected species, genotypes as a pure stand (monoculture) during 2 years (2015 and 2016). The aim of these trials was to observe genotype performance in the field under targeted agronomic and environmental conditions, to receive available data on soil, climate, biotic stresses from each site x year conditions of trials, in order to produce an analysis of the (species -genotype) x environment interaction on yield. Furthermore, local information on crop management (sowing, irrigation, fertilization, desiccation, harvest,...) was obtained and taken into account. A trials protocol was established, and an MS Excel template was created for data recording.

3 climatic zones were defined (Mediterranean, Maritime and Continental). 5 different species of grain legumes (pea, faba bean, lupin, chickpea and grass pea) were tested (spring and winter types). 110 crop cultivars have been evaluated during 2 years (2015 and 2016). Field trials were sown with fully randomized small plots, 3-4 repetitions. per cultivar. 30 genotypes per locality have been tested.

Some critical moments occurred during both seasons, partly due to inter-site seed transmission. Late delivery of seed affected the following countries: Spain, Germany, Serbia, France and Austria. In Serbia some seed was delayed due to customs difficulties. In 2014/2015, high precipitation in the autumn and winter in Greece resulted in late sowing and lower yields. Severe hail damage in Austria affected 50 % of the trial, resulting in lower yields. Bird damage affected some of the emerging pea at JD/GIE Prolupins, France. In the UK at the Wherry & Sons trial, spray drift affected one replication and lowered yields of affected plots of winter beans. A parcel was lost on its way to Serbia containing the spring pea cultivar K-1 (only 1 year of testing took place, 2015-2016 only). In 2015/2016, herbicide residues resulted in crop damage of winter trials, led to no results for Agri-Obtentions. In France, heavy rains in spring destroyed the spring lupin trial, providing no results at JD/GIE Prolupins. Also there was no cropping at RAGT in 2016, due to the rain for winter pea, pest damage (birds and rabbits) for faba bean, and *Aphanomyces* infestation in the soil for spring pea.

Yields obtained in the different locations and for the different species and varieties were presented.

• **Analysis of genotype x environment interactions in pea trials** (V. Biarnès, Terres Inovia ; C. Lecomte and V. Richer, INRA)

The first results obtained from the analysis of genotype x environment interaction for spring pea were detailed. The variety Kayanne was present in all the locations in the 3 climatic zones, and so for this variety, joint regression analysis was carried out. This method was first described by Yates F, Cochran WG (1938), and later Finlay K.W., Wilkinson G.N (1963). This method plots the mean yield of the variety against the mean yield of all the varieties in all the locations, ranged from the lower to the higher. The slope of the regression line measures the adaptability to the environment, while the intercept is a measure of general performance. The method can be used to compare genotype x environment interaction within a climatic zone, comparing locations and varieties with one another.

Then a method developed by INRA, called DIAGVAR was used for spring pea in the three French sites and for the 2 years (2015 and 2016). The DIAGVAR method was developed by C. Lecomte (INRA 2005), L. Prost (INRA 2008) and A. Gauffreteau (INRA 2009), and adapted to the pea crop by C. Lecomte and V.

Richer (INRA 2016) for analyzing genotype x environment interaction in multi-environmental trials for variety evaluation. The method involves three steps: first, preparation of data and calculation of agro-climatic variables, second, an agronomic diagnosis to determine and quantify the limiting factors involved in each trial site, and third, an analysis of the genotype x environment interaction with characterization of variety's behaviour (estimation of genotypic tolerance to limiting factors). Agro-climatic variables are calculated on the basis of the main limiting factors identified in pea (water excess, drought stress, frost, Nitrogen deficit, low solar radiation, high temperatures, diseases and pests, lodging) and the dates of phenological stages. The results show that solar radiation at the beginning of flowering and at beginning of seed filling are the main limiting factors. The network may be divided into two groups: the three locations in 2015 and the three locations in 2016. Then a characterization of the variety behaviour is possible. Some of them seem to tolerate water excess (in green) whereas the others are sensitive (in red).

In conclusion, joint regression may help analysis of genotype x environment interaction for all species and DIAGVAR is a method that can be used to analyze genotype x environment interaction for spring and winter pea in the European network. This method needs meteorological data on the European network (Tmin, Tmax, P, ETP and Rg).

- **Report on Marker-assisted selection in pea and faba bean plant breeding programs at three breeder's locations** (Michaela Ludvíková, AGRITEC, Bente Lund, NPZ and Eveline Adam, SZG)

Marker-assisted selection (MAS) in pea breeding program was used by AGRITEC to introduce resistances to different fungal pathogens (*Fusarium oxysporum* f. sp. Pisi race 1, *Erysiphe pisi*) and viral pathogens (Pea seed-borne mosaic virus (PSbMV) and Pea enation mosaic virus PEMV) into breeding lines. Crossings from 2014-2016 were multiplied in Chile. F2 generation was evaluated in field trials in Agritec. A standard evaluation was made for phenological traits and yield. In parallel, the foliage sampling and analysis of DNA by MAS (10 plants/sample) and plant selection and analysis for the next generation was made on F2 generation field trials. In 2017, resistant lines (according to MAS) were sown in field conditions and the standard breeding procedure will be applied. The goal is to increase the complex resistance to fungal pathogens and viruses, to get new resistant lines and new varieties from them. MAS will help to reach this goal faster and with results that are more precise.

In Norddeutsche Pflanzenzucht (NPZ), MAS was applied on Faba bean to get an *Ascochyta* resistant population and a low vicine/ convicine population. F1 plants were multiplied in Chile. The F2 generation was evaluated by NPZ both for phenological traits of resistance to *Ascochyta* and vicine/ convicine content) and DNA extraction was currently being optimized in NPZ lab. Marker application was also being established at the marker lab of RAGT/France. For vicin/convicin, eight genotypes with normal, low and intermediate vicin/convicin content were validated (according to photometric data). F2 seeds from these lines will follow after successful validation. The aim is to establish a routine application in the NPZ's laboratory.

For Saatzucht Gleisdorf, the aim of the MAS work was also to get *Ascochyta* resistant and low vicine/ convicine populations in faba bean. 4 genotypes which have been genotyped with two flanking markers (VfVc12 and VfVc13) and which have showed complementary patterns, have been crossed. The two flanking markers are now 0.9 and 3.6 cM, respectively, away from the vicine-convicine locus. Photometric measurements have been made. 67 plants were evaluated with both methods. 62/67 analyses showed results consistent with linkage of the marker to the Vc/Cvc locus. 2 plants were near our lvc limit of 3.0 Abs 274 nm/g seeds and 3 analysis showed different results (1 plant is lvc according to MAS, but not by photometric measurement (additional unknown substance registered by photometric analysis?) and 2 plants are high vc according to MAS, but lvc according to photometric measurements. Analysis of plants cultivated in 2017 with new Vc/Cvc markers will be made and the re-test of the 3 progenies with different results in markers/photometric measurements. Then a cross-check will be made of the new results with photometric measurements.

- **Debate on the follow-ups to this work** (Steve Belcher, PGRO and Richard Thompson, INRA)

The data gathered on behalf of the Legato project will continue to be analyzed through other European projects such as LegValue and TRUE, which will hopefully bring a greater understanding of how the varieties are adapted for the climatic zones. The data will be accessed through a database of trial results available on the Legato website.

4. Main conclusions and eventual actions to be taken

Thanks to the programme organized by J. Poslusna of Agritec, this meeting gave a good opportunity to have insight into the legume breeding, biotechnology, and seed production industry in the Czech Republic. Legume breeders from several central and west European regions attended the meeting, and benefited from the overview of the Czech legume breeding set-up and also the trials and their analysis being carried out within the LEGATO project. Finally the experience of four breeding companies in developing the MAS approach for fungal and viral resistance traits was informative. The value to the breeders of the analytical service provided by the SME AMBIS (G. Kiss) was evident, and may be the basis for further collaboration. The LEGATO trials results and their interpretation is the object of a paper currently being prepared for publication, and will contribute to the guidelines brochure also in preparation (Deliverable 7.8).

Annex 1: Fourth Stakeholder meeting program

17th of May 2017

08:30 – 09:00	Registration Stakeholder meeting with a local bias to the Czech Republic
09:00 – 09:30	Welcome and commencement – Information about the company Agritec, 75 years of agricultural research. Speakers: Director Dr. Prokop Šmirous, Dr. Martin Pavelek Information about LEGATO Speaker: Dr. Richard Thompson, INRA
09:30 – 10:45	Presentations of the officials from the Ministry of Agriculture Speaker: Ing. Martin <i>Liška</i> Activity of Association of Grain Legumes growers and processors in the Czech Republic Speaker: Ing. Jan Prášil, chairman APZL, director of Semo a.s. Breeding of grain Legumes in the CR Speaker: Ing. Pavel Horčíčka, director of breeding, Selgen a.s.
10:45 – 11:00	Coffee break
11:00 – 12:30	Testing and registration of varieties of legumes in the Czech Republic (Central Institute for Supervising and Testing in Agriculture) Speaker: Ing. Tomáš Mezlík, director of National Plant Variety Office Economy of growing legumes in the Czech Republic, subsidies, perspectives of the CAP Speaker: Ing. Jaroslav Humpál, ÚZEI Use of biotechnology and advances in legumes in the Czech Republic Speaker: Dr. Miroslav Griga, Agritec
12:30 – 14:00	Lunch

- 14:00 – 16:00 Report on the two-year pan-European trials network
Speaker: Ing. Jana Poslušná, Agritec
- 16:00 – 16:15 Coffee break
- 16:15 – 17:30 Report on Marker-assisted selection in pea and faba bean plant breeding programs at three breeder's locations
Speaker: Ing. Michaela Ludvíková, Ph.D., AGRITEC
Bente Lund, NPZ
Eveline Adam, SZG.

Dinner

18th of May 2017

- 09:00 – 10:30 Road-map for the preparation of reports and deliverables required by the EC
Speaker: Steve Belcher and Jana Poslušná
- 10:30 – 10:45 Coffee break
- 10:45 – 12:00 Debate on the follow-ups to this work
Speaker: Steve Belcher and Richard Thompson
- 12:00 – 14:00 Lunch
- 14:00 – 18:30 Excursion to the Legato field experiments
Excursion in Agritec
Visit of the chateau Velke Losiny with the guided tour and the spa (based on interest)

Dinner

Annex 3: Picture of part of the participants at Wellness Hotel Diana, Velké Losiny, Czech Republic

