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SCIENTIFIC REPORT

Sixty Years of the Crop Research Institute and its Impact on Plant Breeding

Ilja Tom PRÁŠIL, Jana CHRPOVÁ and Zdeněk STEHNO

Crop Research Institute, Prague-Ruzyně, Czech Republic

Research workers at the Crop Research Institute (CRI) Prague-Ruzyně, Division of Plant Genetics, Breeding and Product Quality, have contributed significantly to the breeding of field and vegetable crops over the existence of the Institute. On the occasion of the sixtieth anniversary of CRI we would like to offer a short review of the history and future prospects of these activities. Over sixty years of research (1951–2011) workers of the Institute were developers or co-developers of 66 cultivars (Table 1). Research contributions had practical impacts on the development of cereal breeding in the following areas in particular: the accumulation and characterization of genetic resources from around the World, the study of the quality and identity of cultivars, studies of crop growth habit and earliness genes, the resistance of cultivars to important diseases and abiotic stresses. In vegetables and *Brassica* crops the research contributed, in particular, to the introduction of hybrid breeding methods using cytoplasmic male sterility (CMS) and self-incompatibility, the production of doubled haploids (DH) by means of microspore culture, and the development of somatic hybridization by protoplast fusion. Since the early 1990s molecular biology methodologies have been developed.

Figure 1. Crop Research Institute in Prague-Ruzyně

Table 1. List of cultivars developed by the workers of the Crop Research Institute Prague-Ruzyně often in cooperation with other breeding institutions

Crop	Cultivar/year of registration
Winter wheat	Lena (1975), Juna (1979), Vala (1980), Odra (1981), Zdar (1983), Mara (1984), Hana (1985), Sparta (1988), Sofia (1990), Vega (1992), Samanta (1993), Siria (1994), Asta (1994), Šárka (1997), Vlasta (1999), Rheia (2002), Raduza (2006), Sakura (2007), Sultan (2008), Secese (2009)
Spring wheat	Ruzyňská II (1959), Praga (1968), Jara (1975), Rena (1978), Maja (1990), Septima (2008), Seance (2008), Izzy (2011)
Winter barley	Lunet (1990), Okal (1992)
Winter rapeseed	Aplaus (2007), Opus (2007), Oxana (2007), Oponent (2006), Benefit (2009)
Spelt wheat	Rubiota (2001)
Emmer wheat	Rudico (2006)
Poppy	Hybrid HD (1959)
Stalk type tomato	Start F1 (1981), Start S (1981)
Gherkin	Triga F1 (1979), Lyra F1 (1982), Vega F1 (1985), Petra F1 (1988), Korona F1 (1991), Alena F1 (1993), Partena F1
Cauliflower	Praga (1964), Expres (1965), Brilant (1982), Bolero (1982), Bora (1983), Regent (1988), Diplomat (1988), Fontana (1992), Dalibor (1993), Delta (1999)
Spinach	Besta (1965)
Winter onion	Augusta (1984)
White kohlrabi	Sparta F1 (1986), Luna (1993)
Bean	Pluto (1967)
Zucchini	Diamant F1 (1971)
Cabbage	Langendijská (1964)
Lettuce	Pražan (1971)
Field pea	Vesna (1967)

Genetic resources as the starting material for breeding of cultivars

The Crop Research Institute in Prague-Ruzyně (Figure 1) has dealt with plant genetic resources (PGR) for many years, and thus it carries on the activities of predecessor research stations from the period before the Second World War (Dotlačil *et al.* 2008). In the 1950s until the beginning of the 1990s, the Gene Bank Department significantly facilitated the access of breeders to foreign cultivars. The use of genetic resources led to the release of the wheat cultivars Ruzyňská 2 (1959) and Praga (1968). At present, the Gene Bank Department investigates characters and traits of important cereals and minor crops besides the main activities such as the coordination, documentation and conservation of seed propagated plant genetic resources. These crops comprise wheat, triticale, winter barley, buckwheat, millet, sorghum, amaranth, foxtail millet and some other crops that are under the custodianship of CRI within the framework of the National Programme of Conservation and Utilization of Plant Genetic Resources and Agrobiodiversity. The National Programme was established according to the Law 148/2003 Sb., and the CRI is the national guarantor accredited by the Ministry of Agriculture of the Czech Republic for coordination. Results of annual evaluations are passed to plant breeders of the relevant crops in the Czech Republic. Data, in the form of averages over several years, so called description data, are deposited in the national information system EVIGEZ http://genbank.vurv.cz/genetic/resources/. This information allows for the selection of suitable genetic resources for the breeding programmes, or for research that precedes practical application. Frequently sought-after data is on pedigrees of wheat and barley cultivars, and in the case of wheat, information on gene locations.

This data was gathered in cooperation with Russian colleagues and published first in print and now on the website http://genbank.vurv.cz/wheat/pedigree/ or http://genbank.vurv.cz/barley/pedigree/.

The extension of diversity at the crop level is important for breeders of minor crops and small scale farmers who cannot compete with big companies in the production of staple crops. In the last 10 years, the spread of some less cultivated cereal species from the collection of the Gene Bank can be mentioned. The following cultivars were developed from them: the spelt wheat cultivar Rubiota, and the emmer wheat cultivar Rudico, which have certificates of legal protection (Stehno 2007).

Due to the interest of farmers in spring cereals for organic farming, research on spring wheat focused on traits important for that type of farming. In contrast to the cultivars for traditional farming some "non traditional" traits are important such as the type of establishment after emergence that enables a higher stand density and enhances the competitive ability of the crop. A larger distance between the flag leaf and the spike limits the transfer of diseases to the spike. Other important traits are disease resistance and grain quality.

Molecular marker techniques such as SSR markers allow the exact identification of genetic resources. Within the framework of the collection of genetic resources, 985 wheat and barley accessions were characterized by this method (Roussel *et al.* 2005).

The general aim of gathering and evaluating new genetic resources is to offer alternative genotypes for modern cultivars by introducing new genetic diversity, allowing further genetic improvements in productivity and quality, and the effective utilization of current growing technologies. The application of molecular genetics methods for the study of genetic diversity in collections and the selection of suitable donors for breeding is being extended. At the same time, the current needs of the users from breeding and research are taken into account. Every year, the Gene Bank offers about two thousand genetic resources to users in the Czech Republic and abroad.

Utilization of storage and enzymatic proteins for the verification of cultivar authenticity and quality

In the 1980s, methods of cultivar identification based on electrophoresis of protein markers began to be intensively developed at the CRI. In cooperation with wheat breeders they were successfully introduced into breeding practice. Storage and enzymatic proteins of wheat and barley that fulfil the criteria as useful genetic markers (signal genes) were intensively studied in relation to plant breeding as well as to other activities such as seed production, variety testing and in the food processing industry. Electrophoresis of gliadins and glutenins in wheat, and hordeins in barley, enables the characterization of single genotypes (lines, cultivars) and determine their homogeneity or heterogeneity with a high accuracy (Bradová & Šašek 2005). These proteins also enable important economic traits and characters of wheat, as well as of barley, to be tracked. That is why they are applied in breeding to monitor the development of new cultivars for the indirect identification of linked known genes. They are also used for the selection of suitable parental combinations and homozygous progenies, and for the assessment of the efficacy of individual breeding methods. The linkage of gliadins and glutenins to baking quality is routinely used in breeding as a means of genetic quality testing. The detailed characterization of genetic resources by means of protein markers (e.g., gliadins, hordeins, HMW or LMW glutenins) and the detection of linkages to important traits is a basis for MAS (marker assisted selection) (Psota & Bradová 2009; Bradová & Štočková 2010). Recently, automated chip electrophoresis has been developed for electrophoretic separation of wheat and barley storage proteins (Bradová & Matějová 2008). Because of its high efficiency this modern method may become a significant contributor to breeders methods, primarily in the tests of protein composition in wheat and barley populations at early stages of the breeding process.

Disease resistance of cereals

A stem rust epidemic on rye that affected the area of Šumava in 1952 was a motivation for research of cereal rusts. Later research was primarily focused on rusts of wheat. The study of physiological races of stem-, leaf- and yellow rust was initiated in the CRI as early as the 1960s and has continued to the present time. Research covers the monitoring of physiological races in the Czech Republic, the study of the genetics of resistance, and the testing of large collections of domestic and foreign cultivars under field- and greenhouse conditions (Bartoš & Bareš 1971; Bartoš *et al.*

1996; Hanzalová 2010). At present, molecular markers are also tested for the determination of resistance genes. (Hanzalová *et al.* 2009; Sumíková & Hanzalová 2010).

Research at CRI has contributed significantly to the study of cereal rusts at an international level (Mesterházy *et al.* 2000), and to the application of knowledge in cereal breeding, including studying resistance from *Triticum monococum* (Valkoun *et al.* 1989). Results of the research also found direct applications such that wheat cultivars susceptible to yellow rust in the field were withdrawn from the registration procedure. This secured effective protection against that rust for many years. At present, more attention is again being paid to stem rust because of the incidence and epidemic caused by the physiological race Ug99 and relevant mutations. Growing cultivars with various stem rust resistance genes significantly limits potential epidemics, and the total loss of resistance can lead to enormous economic losses. Central Europe is not immediately endangered but risks cannot be excluded. Because of climatic changes present research at CRI aims at the investigation of the over wintering possibilities of stem rust in the Czech Republic, and the possible transfer of graminicolous stem rust forms to wheat. Rust research also includes studies on stem rust virulence, and on the possibilities of applying molecular methods for the characterization of isolates/pathotypes of this rust, and the detection of resistance sources in related species of the tribe *Triticeae.*

Studies of bunt resistance have a long lasting tradition at the CRI. Since 1988 common bunt resistance in wheat has been studied by means of seed inoculation with a mixture of *Tilletia tritici* and *Tilletia laevis* teliospores from different locations of the Czech Republic (Dumalasová & Bartoš 2006, 2010a, b). Resistance to common bunt is often neglected because it is expected that bunt is eliminated by seed treatments. However, due to reduced seed exchange and limited seed treatments, common bunt and dwarf bunt have again become an important winter wheat disease in the Czech Republic. Though seed treatment usually provides sufficient control, in particular to common bunt, in many countries breeding for bunt resistance is going on. Research workers at CRI possess a great deal of data on the resistance of winter wheat, and also study bunt resistance levels in triticale and in minor crops like emmer wheat or spelt wheat. In addition to research on the detection of bunt resistance genes and variety resistance, research workers offer technical consultancy services directly to agricultural practice with respect to bunt incidence, as well as of soil contamination with dwarf bunt teliospores.

The Crop Research Institute is one of few workplaces where research on barley yellow dwarf (BYDV) is conducted in a comprehensive way (cooperation between the Division of Plant Genetics, Breeding and Product Quality with the Division of Plant Health). Research comprises study of the pathogen, i.e. the frequency of BYDV incidence in the Czech Republic, as well as the frequency and aggressiveness of different isolates. Special attention is paid to the study of resistance. This includes the detection of resistance sources, validation of the effectiveness of resistance genes, as well as participation in the development of wheat and barley breeding material with a higher level of resistance (Veškrna *et al.* 2009). Special attention was also devoted to the application of the effective gene *Yd2* that can be safely detected by the marker *Ylp* (Šíp *et al.* 2004) in the breeding programme. An example of a recent success achieved in a European programme aimed at pyramiding resistance genes is the registration of the Italian feed barley cultivar Doria, carrying resistance genes *Ryd2*, *rym4* and *Rdg1*. Release of this cultivar resulted from cooperation between EI CR, Fiorenzuola d'Arda, and CRI in Prague-Ruzyně (Kosová *et al.* 2008). In the course of the 1990s, the research programme on fungal diseases was extended to Fusarium head blight resistance, and to causal agents of leaf spots. From the beginning, research focused on the enhancement of Fusarium head blight resistance was carried out in close cooperation with plant breeding organizations. Besides the development of methods for resistance scoring, attention was paid, in particular, to the detection and subsequent introduction of new resistance sources. In cooperation with the State Phytosanitary Administration, monitoring of Fusarium head blight incidence is continuing. Since 2004, the resistance of wheat cultivars recommended for growing in the Czech Republic has been evaluated in cooperation with the Central Institute for Supervising and Testing in Agriculture (CISTA). The evaluation system is based on the combination of data on resistance obtained in artificially inoculated trials, and data from trials with natural infection under favourable conditions. This was accepted by CISTA, as described in the publication by Chrpová *et al.* (2008). Specialists are able to transfer data on variety resistance to agricultural practice. The data are also used for further research focused on the evaluation of the contribution of suitable parental combinations, and on effective selection criteria, including the effectiveness of molecular markers. (Chrpová *et al.* 2011). Another permanently disease interest are pathogens causing brown leaf spot diseases in wheat, *Septoria tritici*, *Stagonospora nodorum*, *Pyrenophora tritici-repentis* (Palicová & Hanzalová 2006). Research workers of the CRI have participated in the development of methods for resistance evaluation. At present, particular attention is paid to brown leaf spot diseases caused by the prevailing fungal pathogen *Septoria tritici.*

In 2002, CRI participated in the establishment of the Sub-commission for Wheat at the Czech-Moravian Breeding and Seed Production Association. The aim of the Sub-commission is to develop cooperation between the research and breeding organizations operating in the Czech Republic. The common programme is aimed at the selection of genotypes with combined resistances to diseases. This cooperation has resulted in the registration of 7 wheat cultivars with co-authorship of workers at the CRI (Sakura, Raduza, Sultan, Secese, Septima, Séance, Izzy) displaying a higher resistance to the diseases studied. Wheat sources of resistance to important diseases were detected and described on the basis of results of field infection tests (Šíp *et al.* 2005).

Tolerance to abiotic stresses

This research, from the 1960's, has been aimed at studying the tolerance of crops to winter stresses, particularly to frosts. The evaluation of frost tolerance in winter cereals by means of artificial tests, where plants are exposed to intensive frosts and to other factors in pots under natural conditions, have a long lasting tradition (the so called 'Ruzyně pot test'). Later, the testing of plants for cold tolerance using special laboratory equipment (automatic freezers and cold cabinets) has allowed studies of other crops such as winter vegetables, over wintering fodder crops, grasses, etc. A wide-ranging database of winter hardiness and frost tolerance, encompassing several thousand accessions of wheat, barley, rye, triticale and rapeseed, of domestic and foreign origin, has been established. These results were applied in the registration and recommendation procedure for new cultivars adapted to our climatic conditions, as well as in the selection of resistant materials for the breeding programmes (e.g., winter barley Lunet originated from the cooperation with the Plant Breeding Station in Lužany and reached one of the highest degrees of cold tolerance in the collections of winter barley cultivars). Detailed studies were focused on the physiological and biochemical changes in plants under cold, and in the last two decades under drought also. Several protective compounds (e.g. sugar, proline, and abscisic acid), traits (e.g. growth habits, root systems) and processes (e.g. chlorophyll fluorescence, water relations) have been studied to find out their use for breeding crops highly tolerant to cold and drought (Prášil *et al.* 2007; Vítámvás *et al.* 2007). Recently, the Laboratory of Genomics and Proteomics has been established for plant analyses and searches for genetic and protein markers associated with resistance to abiotic stresses, and, hence to investigate the possibilities of applications of MAS (marker assisted selection). Research is focused on the study and utilization of dehydrins – proteins induced by stress – as markers of cereal and rapeseed tolerance to low temperatures and drought (Kosová *et al.* 2007; Holková *et al.* 2010; Vítámvás *et al.* 2010). High-throughput techniques such as microarrays (e.g., Affymetrix Gene Chip) and two-dimensional differential gel electrophoresis (2D-DIGE), coupled with mass spectrometry (e.g., MALDI-TOF/TOF), have enabled us to study stress-induced changes in the plant transcriptome and proteome and to identify stress-responsive genes and proteins, respectively. The identification and functional characterization of stress-responsive proteins has significantly contributed to a better understanding of mechanisms underlying plant stress responses, and to the improvement of plant tolerance to stress factors via genetic engineering (Kosová *et al.* 2011)

Growth habit, photoperiodic response and earliness *per se* genes of wheat

Studies of plant growth habit and flowering time have been a long term topic at the CRI, due to their importance for yield potential in the field. Originally, studies were based on wheat aneuploid techniques, introduced by Dr. Jindřich Košner in 1969, who derived a monosomic series of the Czech spring variety, Zlatka (*T. aestivum* L. var. *lutescens,* then a popular local cultivar, from the cross [Janetzkis Früh × Marquis] × Heines Koga, using the complete series of aneuploids of Chinese Spring isolated by Prof. E. Sears in the 1950's. This launched an extensive long term study on the identification of important agronomic genes of wheat, including those for disease resistance, using monosomic analyses. Subsequently, intervarietal substitution lines were developed and used to understand the genes controlling flowering time (mainly *Vrn, Ppd and Eps* genes, aiming at an understanding of their effects under different environmental conditions (Košner & Pánková 1997; Pánková *et al.* 2008a).

A delay of flowering time, supported by an analysis of phenology and yield components within a series of reciprocal substitution lines for homoeologous group 5 chromosomes (carrying different *vrn* genes) between two winter wheat cultivars Mironovskaya 808 and Bezostaya 1 suggested the presence of multiple alleles at the individual loci *vrn-A1*, *vrn-B1* and *vrn- D1* (Košner & Pánková 2001). Monosomic analysis of a local alternative landrace, Česká Přesívka, showed the presence of a gene(s) influencing earliness on chromosome 3B. The substitutions of chromosome 3B of Česká Přesívka (CP3B) into the backgrounds of spring and winter wheat cultivars changed their heading time, which was independent of vernalization but responding to some extent to photoperiod. This new gene (*QFt.CRI-3B.1*) was subsequently mapped to a region near the centromere of chromosome 3B using QTL analysis of mapping populations of recombinant substitution lines (Pánková *et al.* 2008b). At present, the production of near isogenic lines aiming at a more precise study of the gene *QFt.CRI-3B.1* and phenotyping /flowering time experiments has been continued.

Recently, more advanced molecular techniques have been applied and new procedures have been developed at the laboratory of genetic markers of the CRI: A new allele, *Vrn-B1c* was identified in a set of spring wheat varieties, sequenced, and a new multiple PCR test developed for distinguishing all of the *VRN-B1* alleles simultaneously (Milec *et al.* 2011).

Utilization of growth stature genes to enhance wheat productivity and stability

In 1986, a method for the detection of dwarfing genes insensitive to applied gibberellin was introduced. Since then, CRI has initiated and secured the "Gibberellin Programme" focused on the introduction of effective dwarfing genes originating from the cultivar Norin 10 (*Rht1* and *Rht2*). This programme was used broadly, especially at the Plant Breeding Station in Úhřetice, SELGEN a.s. The programme aimed at the selection of high-yielding wheat genotypes (GA insensitive "tall – dwarfs") displaying high ear productivity. Results of the breeding gave rise to the new productive winter wheat cultivars Šárka, Vlasta, Mladka and Rheia, that found a significant place in Czech Agriculture. In the last decade, intensive studies on the use of available molecular markers have also focused on other important genes affecting plant growth type and plant development. Genes studied, in particular, were alleles of the *Xgwm261* locus, and *Ppd-D1* affecting sensitivity to photoperiod. The aim was to determine suitable plant types for different environmental conditions and growing systems. Suitable allelic combinations promising effective adaptation to particular environmental conditions were recently described in a paper (Šíp *et al.* 2011). It can be expected that, on the base of these characteristics, it will be possible to find effective utilization of breeding materials developed in various European breeding programmes. Finding the appropriate place for different wheat cultivars in different agricultural systems is also the objective of a series of experiments in which cultivar responses to different input levels and tillage systems used in agricultural practice is being studied (Šíp *et al.* 2009).

Breeding methods used in vegetable species

CRI research on vegetables started in the 1950s and at the beginning of the 1960s by gathering and evaluating world collections of selected vegetable species, focusing on those cultivar types that were missing in the existing collections. The breeding that followed used modern procedures which led to the registration of numerous cultivars. Some of these are still registered in the National List of Varieties. At the same time, new methods were developed and validated in practice for vegetables whose seed had been exclusively imported until that time.

The breeding of *Brassica* crops has been practiced since the beginning of the 1960s. The breeding of cauliflower used modern methods of line breeding later complemented by tissue cultures, and produced new varieties to replace previously grown cultivars that were mostly imported from abroad. Breeding was going on mainly in cooperation with plant breeding stations of the organization SEMPRA, later MORAVOSEED, and resulted in 10 original Czech cauliflower cultivars.

At the end of the 1960s, research on the utilization of self-sterility in the breeding of *Brassica* crops was initiated in connection with the worldwide development of hybrid breeding. Searches for sources of self-incompatibility (SI) and the development of homozygous SI lines were a basis for the breeding of the first Czechoslovak hybrids of Brussels sprouts and cabbage. Breeding of *Brassica* vegetables was focused on the development of highly uniform line cultivars and hybrids based on self incompatibility as well as cytoplasmic male sterility (CMS).

Studies on hybrid breeding methods in fruit vegetables led to the first domestic F_1 hybrids of tomatoes and gherkins. Stalk type tomato Start F1 (1975–1980) was the first domestic tomato hybrid that combined earliness with high yield. By the utilization of an original discovery of the functional male sterility gene *ps2* in the domestic cultivar Vrbičanské nízké, the hybrid Start F_1 was transformed to a sterile line and substituted by the hybrid Start S F_1 (1986–present). This sterility type was also utilized in the breeding of several tomato hybrids. The gherkin Triga F_1 (1979–1985) was one of the first Czech hybrid gherkins. Partena F_1 (1993–present) was the first domestic parthenocarpic gherkin single hybrid.

At present, research on vegetables at the CRI Prague-Ruzyně aims at the application of biotechnological methods, in particular tissue culture, for the creation of specific breeding materials of vegetables resistant to abiotic and biotic stresses. Methods for the production of doubled haploid (DH) completely homozygous lines of *Brassica* vegetables by the technique of microspore culture has been optimized (Klíma *et al.* 2004), and possibilities of somatic hybridization by means of protoplast fusion are being validated. From the collection of genetic resources, potential donors of resistance to *Plasmodiophora brassicae* are being selected and crosses with the current cultivars of cabbage are carried out to develop doubled haploid lines with the desired resistances.

Breeding methods applied in rapeseed

Genetic and breeding research in rapeseed at the CRI was initiated in the second half of the 1960s. The possibilities of developing DH lines by the method of anther-, later on, microspore cultures *in vitro*, were tested, and sources of self-incompatibility were searched for to be potentially applied in hybrid breeding. In 1997, due to the initiative of the CRI the Association "Czech Rapeseed" was founded, that joins together domestic research and breeding organizations OSEVA PRO, s.r.o., SEMPRA PRAHA, a.s., SELGEN, a.s., AGRITEC Research, Breeding and Services s.r.o. and VÚRV Praha v.v.i. (CRI), dealing with winter rapeseed breeding. This aims to apply the most modern equipment and new technological procedures to rapeseed breeding. Since 2000, member organizations of the Association have been engaged in the execution of the third Ministry of Agriculture Research Project focused on the development of rapeseed genetic resources with high yield potential, desired seed and oil quality, and high resistance to biotic and abiotic factors. With respect to the close connection between research and breeding organizations, this Association is a model example of a successful cooperation. At the CRI, a method for the development of DH rapeseed lines by the technique of microspore culture was optimized (Klíma *et al.* 2008). It is now applied especially for the development of homozygous DH lines from F_1 crosses of prospective initial materials, self-incompatible DH lines with improved quality, stabilized lines with fertility restoration genes for hybrid systems CMS Ogu-INRA and Shaan 2A, and yellow seed materials with lower fibre content in the seed. At the CRI, an analytical method for the determination of seed quality by means of near-infrared spectroscopy (NIRS) was also introduced. This method enables effective, fast and non-destructive selection of undesirable glucosinolate content, and selected fatty acids, in oil on a large number of seed samples of rapeseed breeding materials. The existing activities of the Association have resulted in five new Czech line cultivars of winter rapeseed (Oponent, Oksana, Opus, Aplaus, and Benefit). The new line cultivar Cortes will be registered next year. The CRI has co-authorship of all these cultivars.

CONCLUSION

Breeding for improved crop quality and tolerance to a wide range of abiotic and biotic stress factors has become a major task under conditions of changing climate and an increasing demand for versatile, high-yielding crops. The research of the Division of Plant Genetics, Breeding and Product Quality (DPGBPQ) has contributed significantly to breeding new varieties of cereal, vegetable and oilseed rape cultivars, by itself or in cooperation with breeding institutions. In cooperation with state organizations such as the State Phytosanitary Administration or Central Institute for Testing and Supervising in Agriculture, DPGBPQ has contributed significantly to the improved assessment of quality and tolerance of the cultivars grown in the Czech Republic. For future success, a mutual collaboration between Czech and foreign researchers aimed at elucidating plant genetics, plant material testing, and the development of cultivars based on the utilization of scientific knowledge (e.g., genetic markers – marker assisted selection MAS, gene pyramiding) is crucial. The role of the Crop Research Institute should lie in research aimed at an improvement of crop quality, yield production and practical application of the scientific knowledge in crop breeding.

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Correspnding author:

RNDr. Ilja Tom Prášil, CSc., Výzkumný ústav rostlinné výroby, v.v.i., Drnovská 507, 161 06 Praha-Ruzyně, Česká republika
e-mail: cropscience@vurv.cz; www.vurv.cz