

Boosting innovation in breeding for the next generation of legume crops for Europe

The plan for boosting the breeding of phaseolus beans

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Legume Generation Report 5



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Legume Generation

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Background

Our project is structured and focused to directly support our breeding partners and associates as risk-taking innovators. A core principle in plant breeding is a species-specific entrepreneurial activity. Boosting should be a species-specific effort. Consequently, our six species-oriented breeder-led innovation communities (ICs) link practical breeding with the supporting research-base in a transdisciplinary platform. This operational framework enables each of the six ICs to harness the relevant science base provided by six cross-cutting science-support activities. This combines innovation focus on individual species/species groups in each IC with coherence and synergies across the project. All six ICs innovate up to the point where newly-bred germplasm and tools are demonstrated on farm at technology readiness level 7. In this way, the innovation drive within our ICs is a multi-actor force that harnesses science to improve our breeding programmes.

Phaseolus beans

The common bean (*Phaseolus vulgaris* L.) is a diploid (2n=2X=22) and self-pollinating species that was originally domesticated in two American regions (Andean and Mesoamerican), and from there the crop was distributed throughout the world. In total, five phaseolus species have been domesticated: *P. vulgaris* (common bean), *P. acutifolius* (tepary bean), *P. coccineus* (scarlet runner bean), *P. dumosus* (year bean) and *P. lunatus* (lima bean). Besides their own value as crop, these closely related species represent the secondary genetic pool for common bean as the most important *Phaseolus* species and offer valuable genetic diversity for crop improvement by interspecific crosses. For example, breeders worked on transferring high levels of resistance to biotic and abiotic stresses from the secondary gene pool into common bean.

Common beans ('beans') are the second most important cultivated legume species in the world (37.7 million ha for dry bean and 1.58 million ha for snap bean and the most important legume for human consumption. Beans are a traditional food crop grown widely in Europe with more than 500,000 ha according to the INCREASE¹ project. In fact, 18 European traditional high-quality dry bean production chains are protected by protected geographical indication (PGI) and protected designation of origin (PDO). However, despite our substantial research base in Europe, the growing market for beans for sustainable healthy diets will be met from low-cost imports unless the European crop is revived.

Extensive phenotypic variations have been reported in common beans. Concerning plant phenotyping, common beans exhibit different growth habits which vary between determinate bush types and indeterminate climbing types (pole bean). Plants with determinate growth habits have stems ending in inflorescences, whereas plants with indeterminate growth habits have stems that do not end in an inflorescence. Depending on the genotype, the beans can be consumed as pods (green or snap beans), dry seeds after rehydration (dry beans), or both. The dry bean group is the most cultivated worldwide and represents the main source of plant proteins in many parts of the world. Bean seeds contain ~22-25% protein. Bean seeds also show a wide variation in seed traits such as colour, colour distribution, shape, size, dimension, weight, and chemical composition. Snap beans, (syn. garden, French, or green beans) are a group of common bean cultivars whose

¹ INCREASE – Intelligent Collections of Food Legumes Genetic Resources for European Agrofood Systems | INCREASE



fresh pods (immature pods and seeds) are consumed as green vegetables. Fresh pods are harvested at a physiologically immature stage of development when full length has been reached, but the pod-filling process is at an early intermediate stage. One trait that is highly homogeneous among snap beans is the low content of lignin in the pods, which makes most modern snap bean cultivars indehiscent and stringless. Snap beans also vary in terms of seed traits such as colour, shape, size, cross-sections, and dimensions, and are adapted for fresh or processed consumption.

Europe has a long tradition of bean breeding, particularly snap beans, by seed companies (e.g., Vilmorin at the beginning of the 19th century). As a result of this work, a wide variety of snap bean genotypes are found in the market demonstrating diversity in colours and pod shapes. Currently, companies focus on improving resistance to diseases (anthracnose, common mosaic virus, rust, powdery mildew), pests (aphids and weevil), drought tolerance, pod quality traits (colour, shape), adaptation to processing, adaptation to mechanisation, and performance. Concerning dry beans and phenotypes covered by PGI, efforts to improve them have been minor since they are very specific phenotypes produced in limited areas. However, these crops also are affected by biotic and abiotic stresses.

The Bean Innovation Community

Figure 1. Public institutions and seed

companies in the Bean Innovative constituted

Legume Generation project

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the

The Bean Innovative Community (BIC) is a group of researchers and technologists committed to promoting the improvement of these species (Table 1; Figure 1). The BIC is made up of researchers with different backgrounds who carry out their work in public institutions and private seed companies. The BIC includes breeders, agronomists, phytopathologists, geneticists, data analysis and specialists in plant genetic resources, etc. An overview of the pre-breeding and breeding programmes can be found in Table 2. The BIC is open to the incorporation of new stakeholders (seed companies, breeders, farmer associations) to increase the impact of our research.

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Community

First name	Second name	Role Focus crop of breeding activity: major (minor)		Organisation
Lars-Gernot	Otto	Public sector researcher		IPK
Tania	Gioia	Public sector researcher		UNIBAS
Donal	Murphy-Bokern	Policy specialist; research director		
Roberto	Рара	Public sector researcher		UNIVPM
Valerio	Di Vittorio	Public sector researcher		UNIVPM
Ivo	Rieu	Public sector researcher		RU
Eric	Visser	Public sector researcher	Pole and bush dry bean	RU
Juan Jose	Ferreira	Public sector researcher and breeder	Pole and bush dry bean	SERIDA
Ana	Campa	Public sector researcher and breeder		
Roberto	Rodriguez	Public sector researcher		SERIDA
Amelie	Detterbeck	Private seed sector association		
Elizabeth	Ninou	Public sector researcher		IHU
Jasmin	Karer	Project secretariat		DS
Eveline	Adam	Private-sector plant breeder	Runner bean (P. coccineus)	SZG
Klaus	Oldach	Private-sector plant breeder	Snap bush bean	KWS
Gerthon	van de Bunt	Private-sector plant breeder	Snap bush bean	KWS
Patrice	Jeanson	Private-sector plant breeder	Snap bush bean	LIDEA
Thomas	Meyer-Lüpken	Private-sector plant breeder	Snap bush bean WAV	
David	Gaikpa	Private-sector plant Snap bush bean breeder		WAV

Table 1. Members of the Bean Innovation Community



Table 2. Pre-breeding and breeding programmes run by the members of the Bean Innovative Community

Partner	Pre- breeding	Breeding	Description
ІРК	1		Pre-breeding of bush snap bean to improve pod quality traits, biotic and abiotic stress resistance/tolerance, germination and genotyping.
UNIBAS	4		Pre-breeding of snap and dry bean (both bush and pole) to improve quality traits, biotic and abiotic stress.
SERIDA	4	4	Pre-breeding and breeding pipelines for pole and bush beans, both as dry and snap beans. Pre-breeding programmes focus on identifying and developing new sources of traits for improvement. Breeding programmes disease resistance, plant phenotypes, and seed quality.
SZG	1	3	Breeding programmes for runner bean (<i>P. coccineus</i>), but also activities for pole and bush dry bean.
KWS	1	1	Pre-breeding and breeding of bush snap bean.
LIDEA	1	1	Pre-breeding and breeding of bush snap bean.
VanW	1	2	Pre-breeding and breeding programmes for bush snap bean, a programme for pole bean in crop mixture with maize for forage.

In addition to the members with pre-breeding and breeding programmes shown in Table 2, UNIVPM carries out activities that support our breeding programmes. These focus on identification and application of strategies for conservation and characterisation of plant genetic resources in legume species, including common bean e.g., INCREASE project (phenotypic, phenomic, nutritional and molecular characterisation, development of genomic tools such as the pangenome and models for genomic predictions based on GWAS studies in multi-location field-trials). UNIVPM supports breeding with genomic, phenomic and phenotypic characterisation of germplasm, including especially locally adapted genotypes. Further research activities that support breeding include identification of common bean genotypes for snap bean production and/or to be used in breeding programmes for snap bean variety development (<u>BRESOV</u> project). Activities at the identification of suitable genotypes for intercropping with maize.



Strategies for improving phaseolus beans

Common bean yield is affected by many factors, including diseases, tolerance to abiotic stress, growth habits, seed phenotype, adaptation to specific climatic conditions, and adaptation to mechanisation. In addition, factors such as adaptation to organic farming systems, adaptation to processing, consumer preferences, quality traits, and chemical composition can impact bean crop expansion. Many of these characteristics can be improved by plant breeding methods.

To develop breeding programmes, breeders need sources of characters (genotypes and variation), knowledge of character inheritance (genetic control), and tools for selecting the best genotypes. These tools can involve phenotyping methods, markers to assist selection, and available genomes. The main objectives of the BIC are to provide resources to boost phaseolus bean breeding in Europe, both in public institutions and private companies.

The key objectives are:

- 1. to collect information about genetic resources from related projects;
- to screen panels of bean breeding lines and genetic resources, including common bean and scarlet runner bean accessions to identify sources of resistance to biotic/abiotic stress, variation in plant/root architecture and in seed quality, and yield. Yield is important especially for dry beans, wheras for snap bean as vegetable the pod quality is of elevated significance;
- 3. to develop and provide new resources and new tools such as breeding populations (biand multi-parental), inter-specific lines, and validated user-friendly genetic markers for our breeders for genomic assisted breeding;
- 4. to develop new bean genotypes and cultivars, suitable as either breeding and prebreeding material, through recombination and pyramiding using cutting-edge methods, such as precision breeding, high-throughput phenotyping and high throughput genotyping complementing classical phenotypic selection.

Our plan to boost breeding

Figure 3 shows how the Bean Innovation Community is supported in Legume Generation and Figure 4 indicates the pathways to application in breeding for the Bean Innovation Community. Basically, there are two ways to approach genetic improvement: taking advantage of existing diversity, and generating new diversity through crossing and recombination. We will use both strategies to boost the bean breeding.

Our breeding business idea is that cultivars bred from local varieties are better adapted to European growing conditions and stresses. This provides the genetic foundation for a revival of phaseolus beans in Europe. While seed companies are mainly focused on snap beans, many local varieties of dry bean are under-exploited. Supported by results from previous projects (BRESOV, BEAN_ADAPT, INCREASE, and TOOLBEAN), we will establish a bean breeding set (BBS1, ~200 lines) constituted with (1) well-known breeding lines with characterised genes or QTL controlling important traits; (2) local traditional varieties (incl. cultivars from European Protected Geographical Indication); and (3) selected snap bean lines and dry beans from previous projects. We will also establish a second panel (BBS2)



specifically focused on bush snap beans with high relevance to our central European breeders (Van Waveren, KWS, LIDEA) comprising breeders' elite material and preselected drought tolerant genotypes.

BIC partners maintain their seed collections (landraces, elite cultivars, genetic stock, and pre-breeding lines) and characterisation databases. Both bean genotypes and available data will be used to establish the BBS panels. Common bean is listed among the species for the multilateral system of access and benefit-sharing in the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA). Seeds constituting the BBS panels will be exchanged through the Standard Material Transfer Agreement (SMTA) using the **Easy-SMTA** website.



Figure 2. Location of phaseolus bean testing sites (field trials and controlled conditions). These locations include a wide spectrum of field conditions.

We will evaluate both BBS panels in field trials for two years at three localities each (BBS1: SER, UNIBAS, SZG; BBS2: Van Waveren, UNIBAS, IPK; see Figure 2) using a standardised set of morpho-agronomic descriptors. Characters related to yield will be prioritised, such as 100-seed weight and num. seeds per pod and production plot. The field trials involve several different agroclimatic conditions, farming systems (organic and conventional), a completely randomised design with three replicates per genotype, and field trials. Comparison between the panels and the combination of both will be enabled by the fact that (a) both panels are cultivated by UNIBAS, and (b) between 20-30 lines should be



common for both panels (reference genotypes). The results of these field evaluations will provide data on the adaptation, and yield stability of tested genotypes as well as identification of the best genotypes. In addition, these field trials will reveal problems with bean crops related to climate change.

Selected genotypes included in the BBS panels will also be evaluated in controlled conditions for response to common diseases in the European crops (BMV, powdery mildew, anthracnose, white mould, by SERIDA), root morphology, and the abiotic stress as drought tolerance (Lemnatec automated phenotyping platform, IPK), heat tolerance (RU), and quality analysis of seeds (e.g., protein content and seed mineral content) and pods (e.g., pod sugar content) (SERIDA and UNIBAS).

A common database will integrate all the data collected in both panels (phenotypic and genotypic) obtained in field trials and evaluations in controlled conditions. This database will be a valuable tool for breeders to select the most suitable genotypes for their improvement programs.

In parallel, we will develop segregating populations from intraspecific (*P. vulgaris* \times *P vulgaris*) and interspecific crosses (*P. vulgaris* \times *P. coccineus*) to generate recombination for forward genetic analysis and obtain new varieties and source traits. We will develop:

- An interspecific population (~ 150 lines) derived from *P. vulgaris* × (*P. vulgaris* × *P. coccineus*) to introgress in P. vulgaris interesting characters from *P. coccineus* (e.g., resistance to anthracnose and powdery mildew). This population will be genotyped and phenotyped to investigate the genomic regions controlling introgressed traits. We hope that within this population new sources of traits to biotic and abiotic stresses will be found.
- A multiparent advanced generation inter-cross (MAGIC) population from very different parents. This population was evaluated for different traits and used in association analysis (GWAS) to detect the genomic regions involved in the analysed traits.
- F2:3 populations to investigate the genetic control of new characteristics introgressed in *P vulgaris* from *P coccineus*. Bulk segregant analysis sequencing (BSA-seq) will be used for rapid mapping of major genes after phenotypic segregation analysis.

We will use cutting-edge genomic methods (e.g., whole genome sequencing, WGS; genotyping by sequencing, GBS; BSA-seq in the mapping) as well as cutting-edge phenotypic methods to connect the phenotype and the genome and to map the candidate genes that control important traits for breeding (e.g., major resistance genes). RNA-seq analysis will also be implemented to validate these genes, allowing the development of breeder-friendly DNA and functional markers. Establishing new DNA markers will increase the efficiency of breeding programmes.

Finally, we will develop a pilot breeding programme to obtain new genotypes (varieties) with new combinations of resistance genes to different diseases using the classical improvement method (e.g., backcrossing, single crosses) and precision breeding to pyramid the major resistance genes in dry and snap bean cultivars. This pilot breeding



programme will use as a model the cultivars protected by PGI 'Faba Asturiana' (https://faba-asturiana.org/) and the resistance genes Co-2, I, Pm1, and bc-3. The selection of potentially best breeding lines will be based on classical resistance tests and marker-assisted selection, taking advantage of molecular markers. This will provide new breeding lines for breeding programmes of BIC and new cultivars to be registered and marketed.

Routes to application

We will strengthen existing collaborations and develop connections with the European legume community of researchers and breeders. The knowledge shared with the breeders will ultimately contribute to the development of new bean genotypes and tools to breed for multiple disease resistances.

Dissemination

The expected results can be classified as a) plant material, and b) data and tools to accelerate the breeding programmes. The main dissemination route is within the innovation community itself – between our members.

Expected results (plant material)	Dissemination plan
Bean breeding set (BBS) well-characterized. Potential source of traits to	Seed exchange among
European breeding programs.	partners through SMTA /MTA
New materials well characterised derived from multi-parental crosses.	Seed exchange among
Potential source of traits to European breeding programmes.	partners through SMTA
New materials and genes are well characterized and derived from	Seed exchange among
interspecific crosses. Potential source of traits to European breeding	partners through SMTA
programmes.	
New materials of the market class fabada with introgressed resistance to	Distribution to local farmers
BCMNV. Potential variety to be protected and marketed.	with the help of seed
	companies

Expected results (data and tools)	Dissemination plan
Data recorded from BBS evaluated in field European trials.	Internal database
	distribution. Publications
Data recorded from BBS evaluated in controlled conditions: Response to	Internal database
biotic (anthracnose, BCM and powdery mildew) and abiotic stresses (drought	distribution. Publications
tolerance). Root phenotype	
A new set of molecular markers for the assisted selection (friendly markers);	Internal database
linked markers and functional markers	distribution. Publications
Precise data on the control of the genomic regions involved in the genetic	Internal database
control of recorded traits	distribution. Publications
Results of a pilot programme to group resistance to for bean disease;	Internal database
anthracnose, bean common mosaic virus, powdery mildew and bean common	distribution
mosaic necrotic virus	



Exploitation

We expect that the results can be exploited through:

- 1. Availability of a wide set of genotypes with superior traits for bean breeding in Europe. These materials can be used by breeders to obtain new cultivars. Material will be available through standard material transfer agreements (SMTAs).
- 2. Potential new varieties to be exploited by partners and seed companies and finally to be used by farmers. Our community will develop segregating populations to support the breeding of new varieties by specific breeding programmes. Considering the interest of the seed companies, this material will be protected and made available to those companies using specific agreements.
- 3. Availability of genes/QTLs controlling important traits for bean breeding in Europe. Public and private breeders will have access to this information to accelerate the breeding programmes, and consume fewer resources to obtain new varieties.
- 4. Molecular markers that tag genes/QTLs to accelerate breeding programmes in Europe. Public and private breeders will have access to this information to accelerate the breeding programmes, and consume fewer resources to obtain new varieties.

We expect that these results will boost the autonomy and efficiency of public and private bean breeding sectors in Europe, both in snap and dry beans.

Communication

All members of the Bean Innovation Community will actively be involved in communication tasks. Communication will focus on internal communication within the BIC and the research community and society (external communication). Internal communication will be developed through periodic online meetings and Legume Generation satellite meetings. For external communication, Tania Gioia (UNIBAS), Valerio Di Vittori (UNIVPM) and Thomas Meyer-Lüpken (van Waveren) will be our ambassadors who monitor developments in the region, identify communication opportunities, and supply content material for the Legume Generation project website (www.legumegeneration.eu) and the Legume Hub, Europe's open-access knowledge platform on legumes that guarantees a permanent availability of the project outputs. A feasible schedule of regular meetings will be developed to obtain timely input from ICs for the development of content for communication channels.

The available resources for communication in the project will be used: the project <u>website</u>, the Legume Hub and social media (<u>Linkedin</u> and <u>X</u>. In addition, the social media channels of institutions, companies, or researchers will help amplify communication to obtain a maximum impact.



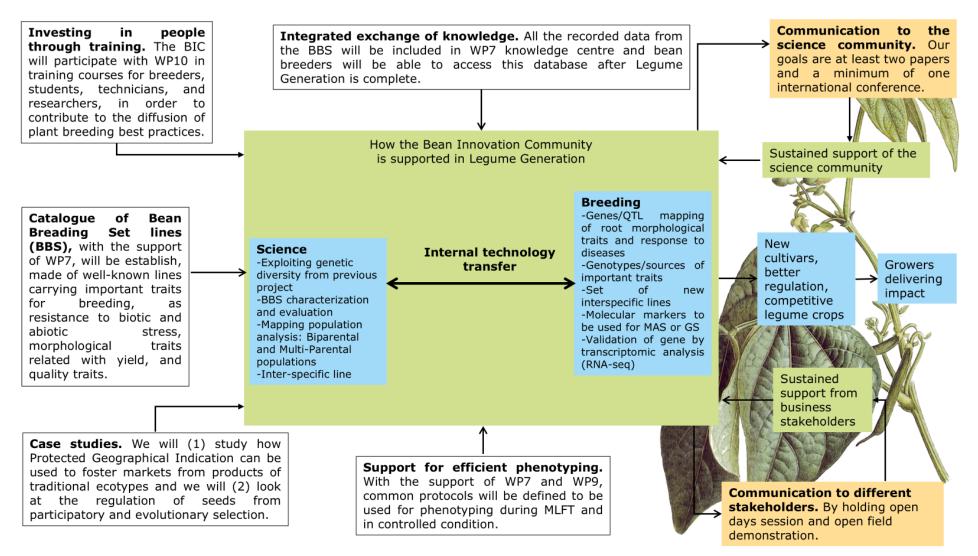


Figure 3. How the Bean Innovation Community is supported in Legume Generation

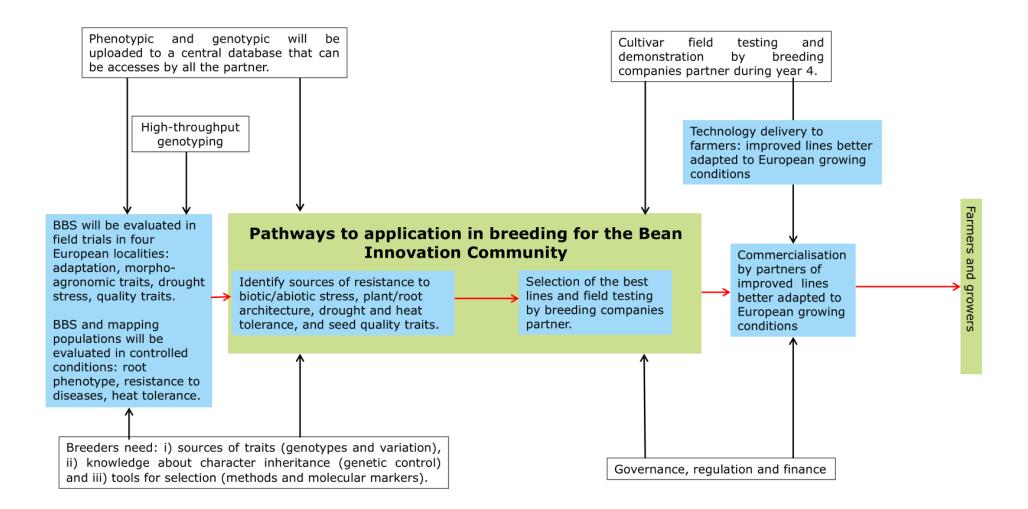


Figure 4. Pathways to application in breeding for the Bean Innovation Community