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> Conservation of horticultural genetic resources in France



Michel Roux-Cuvelier, Michel Grisoni, Arnaud Bellec, Emmanuel Bloquel, Carine Charron, Magalie Delalande, Marine Delmas, Audrey Didier, Charles-Eric Durel, Charles-Henry Duval, Florence Esnault, Laurence Feugey, Emmanuel Geoffriau, Bouchaïb Khadari, Sandra Lepers-Andrzejewski, François Luro, Cécile Marchal, Alix Pernet, Jérémy Salinier, Marc Seguin, Rebecca Stevens, Bernice van Issum-Groyer and Rémi Kahane

The conservation of biodiversity, and particularly of cultivated biodiversity or agro-biodiversity, is an issue of growing importance, particularly due to the acceleration of global change that is affecting the planet, human health, and nutrition. This article presents the strategy and the diversity of conservation actions of horticultural genetic resources as currently practiced in France. It also emphasizes the international horticultural connections of the French Republic, which include temperate, Mediterranean, as well as tropical environments, particularly considering overseas departments and territories.

The role of Biological Resource Centers

Plant genetic resources are the basis for varietal innovation in agriculture. They are an important source of diversity for the evolution of food systems and the development of agro-ecological cropping systems designed for sustainability and resilience. The evolution of regulations on biodiversity management by States and the implementation of the access and benefit sharing (ABS) regime have reinforced the strategic importance of agricultural plant genetic resources (PGR), and particularly of *ex situ* collections. Horticultural genetic resources are a subset of the agro-biodiversity that allows the improvement and development of horticultural value chains in the world. Conservation and valorization of these resources make agriculture more resilient to biotic and abiotic pressures, climate change, and are essential today for the food and nutritional security of the human population. They are also a matter of food sovereignty, which has become exceedingly apparent during the COVID-19 pandemic crisis. The term “Biological Resource Center” (BRC) was coined in 1999, following work carried out by the Organization for Economic Cooperation and Development (OECD). It refers to any structure holding biological samples

and their associated data from the different kingdoms of life: human, animal, plant and micro-organisms.

BRCs and gene banks are dedicated to the collection, management, characterization, conservation, enrichment and distribution of biological samples. To do this, BRCs implement procedures, techniques and databases according to standardized and optimized quality assurance and certification approaches.

They are operated under the responsibility of public research organizations. Their role is essential for research and development of horticulture. They also play an important social and cultural role through the conservation of heritage resources and traditional knowledge.

Infrastructures and organizations servicing BRCs

GIS IBiSA

The Scientific Interest Group GIS IBiSA (Infrastructure in Biology, Health and Agronomy, <https://www.ibisa.net/>) is a public instrument for the facilities maintained by the French life science establishments, including the BRCs. It carries out national policies for labeling and funding biology, health and agronomy platforms as well as BRCs. It promotes the organization and pooling of resources and equipment necessary for life sciences. The GIS IBiSA labels BRCs according to criteria of openness to all users, implementation of a quality management system, technological evolution and training.

RARE, a French agronomic BRC infrastructure

In France, in the field of agronomic and environmental research, BRCs are organized into networks (Mougin et al., 2018). The infrastructure AgroBRC RARE (Agronomic Resources for Research) brings together five networks of BRCs that conserve genetic, genomic, and biological resources:

- Microbial biological resources (<https://doi.org/10.15454/1.5613788897481968E12>);
- Crop resources and plant genomics – BRC4Plants (https://www.agrobrc-rare.org/agrobrc-rare_eng/Presentation/Plant-pillar);
- Forest resources (<https://doi.org/10.15454/1.5613761929199846E12>);
- Genomic or reproductive animal resources (<https://doi.org/10.15454/1.5613785622827378e12>);
- Resources associated with an environmental matrix (<https://doi.org/10.15454/TRBJTB>).

The AgroBRC RARE aims to pool skills, harmonize practices, encourage comparative biology projects and offers a single-entry web portal to facilitate access to well-documented samples. These objectives take into account the regulatory context that varies with the biological nature of the resources, both for sanitary and legal aspects. AgroBRC RARE provides organizational support to its members in the implementation of the Convention on Biological Diversity (CBD) in 1993, the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGR) in 2004, and the Nagoya Protocol in 2010. France has implemented the rules of access to genetic resources and the fair and equitable sharing of benefits arising from their utilization through the European regulation N° 511/2014 and French law No. 2016-1087 for the recovery of biodiversity, nature and landscapes.

The BRCs are under the responsibility of French research organizations working in the field of agronomy and biology: Research Institute for Agriculture, Food and Environment (INRAE), Research Centre for International Development (CIRAD), Research Institute for Sustainable Development (IRD), National Centre for Scientific Research (CNRS), and their partners – the technical institutes and higher education institutions. The BRC4Plants includes 47 living collections of species or groups of species shared between 18 BRCs.

Box 1. Plant collections, a window on viral biodiversity. Contributions of the Safe PGR project - EraNet Netbiome Program (2012-2015)¹

Vegetatively propagated crops are prone to the accumulation of viruses because they do not benefit from viral sanitation following reproduction by seed. On the other hand, Biological Resource Centers (BRCs) bring together a large number of species and varieties from diverse geographical origins.

The general objective of the Safe PGR project was to improve the knowledge of the diversity of viruses infecting the crops addressed by the partners' BRCs, to develop or optimize diagnostic techniques. Ultimately, it aimed to limit the risk of spreading viral diseases through the exchange of tropical plant germplasm.

To reach its objective, the BRC partners from Guadeloupe, Madeira, Azores, and Reunion, combined classical molecular biology and next generation sequencing (NGS) approaches, leading to unprecedented virus discovery in the targeted crops (banana and plantain, sugarcane, yam, sweet potato, garlic and vanilla).

A total of 21 new virus species were discovered (1 in banana, 3 in garlic, 3 in sweet potato, 4 in sugarcane, 3 in vanilla and 7 in yam) and their molecular diversity was explored. This gave the possibility of setting up diagnostic methods for these new agents as well as the optimization of methods for 10 already known viruses. Thus, the knowledge of the viral status of the germplasm collections was improved, which helps to prevent the spread or emergence of diseases.

¹For more information, see Pavis (2015).

In total, more than 103,000 accessions are conserved in the French plant BRCs, of which about 57,000 are horticultural resources belonging to 62 genera.

This organization of the plant BRCs into a network makes it possible to think about and carry out research projects around common issues specific to the BRCs such as genetic diversity and plant genome analysis or viral diversity of vegetatively propagated species (Box 1) or, more generally, the application of national and international phytosanitary regulations.

The French national coordination for plant genetic resources: safeguarding "orphan species"

The national coordination works towards the official recognition of collection curators and the identification of plant resources of cultivated species and their wild relatives (other than forest trees) that are relevant for France. Set up by the Ministry of Agriculture, the coordination is composed of a national support structure (SCN) hosted within the GEVES (Group for the Study and Control of Varieties and Seeds), and a Cross Section of the CTPS (Permanent Technical Selection Committee) bringing together a diversity of stakeholders (47 members) involved in the conservation and development of these resources.

In this context, the SCN acts to safeguard species, which are relevant for French agriculture. It focuses in particular on the conservation of "orphan" species, species without an identified collection or with a collection for which the regeneration or characteriza-

tion are not carried out: for example, beans (*Phaseolus* sp.), grass pea (*Lathyrus* sp.), lentils (*Lens culinaris*) and onions (*Allium cepa*). Research has been carried out on existing collections, inviting other stakeholders who are interested in getting involved in the management of these species. For beans and onions, public and private cooperation networks are in the process of being created. For grass pea and lentils, the approach was unsuccessful due to an insufficient number of stakeholders. Nevertheless, discussions are being held with international centers to identify potential solutions for safeguarding these collections. Other species will be studied, such as buckwheat (*Fagopyrum esculentum*), radish (*Raphanus sativus*) or asparagus (*Asparagus officinalis*).

Involvement of French BRCs at European level

Though genetic resources management is organized mainly in crop-oriented networks and BRCs in France, centralized multi-species gene banks prevail in Europe. The cooperation on PGR in Europe is organized through ECPGR (The European Cooperative Programme for Plant Genetic Resources, <https://www.ecpgr.cgiar.org/>) with 33 member-countries and 23 working groups with several addressing horticultural crops: *Allium*, berries, *Brassica*, cucurbits, leafy vegetables, *Malus/Pyrus*, medicinal and aromatic plants, potato, *Prunus*, *Solanaceae*, umbellifer crops and *Vitis*. French BRCs actively participate to ECPGR. Genetic resources in Europe covering 43 countries, 400 institutes and 2 million accessions can be found in the Eurisco database (<https://eurisco.ipk-gatersleben.de>).

Biological Resource Centers in France for horticultural plants

Within the BRC4Plants, eleven BRCs manage horticultural living collections (Figure 1; Table 1). An additional BRC, the CNRGV (French Plant Genomic Resources Center), is specifically dedicated to genomic resources (Box 2). A French specificity within the European space is to have several ultramarine territories, called RUP (ultra-peripheral region) at the European level or DROM-COM (overseas departments and regions - overseas collectivities) at the French level. These territories are mainly located in tropical environments. They are home to significant biodiversity and host many collections of tropical horticultural genetic resources organized in BRCs.

The Florilège portal (<http://florilege.arcad-project.org/fr/collections>) provides a focal point for web entry into the biological resources of plants for agriculture conserved in France (metropolitan and overseas). In its



■ Figure 1. Localization of the 11 French Biological Resource Centers (BRCs) around the globe. Left: overseas BRCs, right: mainland France and Corsica BRCs. A = BrAcYSol, B = Carrot and other vegetable *Apiaceae*, C = Citrus, D = Grapevine, E = Olive trees, F = *Prunus-Juglans*, G = Rose-Pom, H = Tahitian vanilla, I = Tropical plants, J = Vatel, K = Vegetables. (Figure designed by M. Duportal, CIRAD).

Box 2. The French Plant Genomic Resources Centre (CNRGV)²

The CNRGV is a national infrastructure within the National Research Institute for Agriculture, Food and Environment (INRAE). Settled in Toulouse (France) in 2004, it is both a Biological Resource Centre (BRC) dedicated to plant genomic libraries and a service provider for plant genomics projects. A genomic library consists of collection of DNA fragments cloned into bacteria (so called BAC clones) that, altogether, represent the complete genome of a plant. The BAC clones are easy to conserve, to screen for genes of interest, to isolate DNA from and to sequence.

The missions of the CNRGV are to produce, conserve, characterize, and distribute genomic resources to, ultimately, understand the organization of plant genomes or to link biological functions or agronomical traits to the DNA sequences that govern their expressions. To fulfil these objectives, the CNRGV can either screen the genomic libraries to isolate the BAC clones of interest and sequence them, directly target and sequence large DNA fragments, using internally developed CATCH methods, or produce an assembled genome sequence of a genotype of interest in collaboration with sequencing facilities.

To date, the CNRGV conserves 392 genomic libraries corresponding to 45 plant species including 20 vegetable and horticultural species. Through collaborations on these particular species, the CNRGV has implemented various projects representative of the services available. They include the production of preliminary genomic data for species not or scarcely characterized (parsnip, passion fruit), the characterization of regions governing resistances to biotic (resistance to viruses in pepper) and abiotic (frost tolerance in pea) stresses, the production of reference genome sequences (tomato, vanilla). Recently, the CNRGV has developed tools to characterize intraspecific variations in chromosome structure (apricot).

In the future, the CNRGV aims to help to better characterize the genetic resources at the genomic level.

²For more information, see <https://cnrgv.toulouse.inrae.fr/>

current configuration, it represents the network of plant BRCs managed by INRA, CIRAD and IRD members of the national infrastructure AgroBRC RARE. It should soon be interconnected with the European portal Eurisco (<https://www.ecpgr.cgiar.org/resources/germplasm-databases/eurisco-catalogue>).

The Inter-TROP portal (<http://crb-tropicaux.com/Portail>) is dedicated to the biological resources of tropical plants for agriculture, maintained by the plant BRCs attached to French research organizations in tropical areas.

Biological Resource Center BrACySol (Figure 1A)

The BRC BrACySol preserves and characterizes collections of genetic resources of different cultivated genera: *Brassica* (cabbage, turnip, rape and mustard), *Allium* (shallot and garlic), *Cynara* (artichoke) and *Solanum* (potato and related species). It is supported by two INRAE units: the Joint Research Unit Institute of Genetics, Environment and Plant Protection (IGEPP) and the Experimental Unit Genetic Resources in Oceanic Conditions (RGCO), and is based in Ploudaniel in Brittany. The BRC BrACySol is involved in various European (H2020 G2P-Sol; Prima Bras Explor) or national (PIA Rapsodyn; CASDAR Brassidel, PoTStaR, GeCoNem) projects, and projects funded by professional partners (Promo-

sol, GIE Colza, Association des Créateurs de Variétés Nouvelles de Pomme de Terre, Fédération Nationale des Producteurs de Plants de Pomme de Terre). The objectives of these projects are i) to explore, describe, and structure genetic diversity, ii) to develop core collections, iii) to carry out genetic association analyses to identify the regions of the genome involved in resistance traits to different pests or abiotic constraints, iv) to introduce this diversity into pre-selection material by exploiting recombination, and v) to develop markers that can be used in marker-assisted selection.

The *Brassica* collection includes 1,200 accessions of vegetable crucifers, which are pop-

ulations collected from farmers before their disappearance in favor of hybrid varieties, and 2,200 accessions of oilseed crucifers including lineage varieties representing world variability. It also includes original scientific material presenting traits of agronomic importance such as resistance to different pests, or seed quality. The accessions of this collection are stored as seeds in freezers (-18 °C) or in cryopreservation containers (-196 °C). The seed lots are regenerated every 10 to 15 years, depending on the evolution of their germination faculty.

The *Solanum* collection includes 12,000 accessions. It is composed of i) clones of potato-related species originating from South America and characterized for a certain number of traits of interest, ii) varieties representing world variability, including heritage ones that are not maintained in any other European BRC, and iii) original scientific material presenting traits of agronomic importance such as resistance to different pests, or related to the technological quality of tubers. These accessions are maintained by vegetative propagation in the form of tubers, in vitro plantlets or cryopreserved meristems. The maintenance of related species in the form of characterized clones constitutes a specificity of this collection.

The *Allium* collection includes 120 garlic accessions and 300 shallot accessions. It is composed of populations collected in France before the opening of the catalog in 1991, old and new varieties and original scientific material for the selection of agronomic characteristics such as disease resistance or dry matter content of bulbs. The accessions are maintained by vegetative multiplication in the form of bulbs and planted in the field every year.

The *Cynara* collection includes 20 artichoke accessions. It is composed of French varieties and varieties used as reference in the trials. The accessions are maintained in the form of plants in the greenhouse.

The BrACySol BRC coordinates three national networks for the conservation of plant



■ Figure 1A. A. Diversity of potato varieties. Credit: C. Maitre, INRAE. B. Diversity of cabbage populations. Credit: N. Quééré, INRAE.

■ Table 1. Detailed list of French Biological Resource Centers dealing with horticultural collections.

Code	BRC name	Town (region)	Affiliation	Contact	Family
A	BrACySol	Ploudaniel (Brittany)	INRAE	Bracysol@inrae.fr	<i>Brassicaceae</i> <i>Alliaceae</i> <i>Asteraceae</i> <i>Solanaceae</i>
B	Carrot and other vegetable <i>Apiaceae</i>	Angers (Pays de la Loire)	Agrocampus Ouest	crbcarotte@agrocampus-ouest.fr	<i>Apiaceae</i>
C	Citrus	San Giuliano (Corsica)	INRAE-CIRAD	emmanuel.bloquel@inrae.fr	<i>Rutaceae</i>
D	Grapevine	Montpellier (Occitania)	INRAE	cecile.marchal@inrae.fr	<i>Vitaceae</i>
E	Olive tree	Porquerolles (French Riviera)	CBN Med/INRAE	b.khadari@cbnmed.fr magalie.delalande@inrae.fr	<i>Oleaceae</i>
F	<i>Prunus-Juglans</i>	Avignon (Provence), Bordeaux (Aquitaine)	INRAE	marine.delmas@inrae.fr	<i>Rosaceae</i>
G	Pome fruits and roses (RosePom)	Beaucouzé (Pays de la Loire)	INRAE	Alix.Pernet@inrae.fr Laurence.Feugey@inrae.fr	<i>Rosaceae</i>
H	Tahitian vanilla	Raiatea (French Polynesia)	EVT	sandra.lepers@vanilledetahiti.pf	<i>Orchidaceae</i>
I	Tropical plants	Petit Bourg (Guadeloupe), Le Lamentin (Martinique)	CIRAD/INRAE	crb.plantes-tropicales@cirad.fr	<i>Musaceae</i> <i>Bromeliaceae</i> <i>Poaceae</i> <i>Anacardiaceae</i> <i>Dioscoreaceae</i>
J	Vatel	Saint Pierre (Reunion Island)	CIRAD	carine.charron@cirad.fr marc.seguin@cirad.fr	<i>Orchidaceae</i> <i>Amaryllidaceae</i> <i>Araceae</i> , <i>Euphorbiaceae</i> , <i>Dioscoreaceae</i> , <i>Convolvulaceae</i> <i>Fabaceae</i> <i>Cucurbitaceae</i>
K	Vegetables (Leg)	Avignon (Provence)	INRAE	rebecca.stevens@inrae.fr	<i>Solanaceae</i> <i>Cucurbitaceae</i> <i>Asteraceae</i>

genetic resources: the “vegetable crucifers” network, the “oilseed crucifers” network and the “potato” network, and it participates in the *Cynara* network. In addition, the BRC BrACySol is part of the European network ECP/GR and is a member of the *Brassica*, *Allium* and Potato working groups. The collections maintained within the BRC BrACySol are visible on the French portal Florilège.

The BRC BrACySol is engaged in a certification process according to the ISO 9001-2015 standard.

BRC Carrot and other vegetable *Apiaceae* (Figure 1B)

Based on genetic resources research and management activity initiated in 1996, the BRC “Carrot and other vegetable *Apiaceae*”

was re-established in 2011. This BRC is supported by the Institut Agro | Agrocampus-Ouest, with joint research with IRHS (Institute of Research on Horticulture and Seeds). The related activities involve carrot and wild relatives genetic diversity and evolution, the genetic determinants of compounds involved in quality and resistance to diseases in carrot, and the effect of environment,

	Main genus/ species	Common names	Number of accessions preserved	Number of accessions released (2019)	Selected scientific references
	<i>Brassica</i> <i>Allium</i> <i>Cynara</i> <i>Solanum tuberosum</i>	Cabbage, rape seed Garlic, shallot Artichoke Potato	3,400 420 20 12,000	1,310	Aissiou et al. (2018) Esnault et al. (2014, 2016)
	<i>Daucus</i> <i>Chaerophyllum</i> Other <i>Apiaceae</i>	Carrot Tuberous rooted Chervil, parsnip, fennel, etc.	4,376 706 37	5	Chevalier et al. (2021) Geoffriau (2020) Martinez Flores et al. (2019)
	<i>Citrus</i>	Citrus	1,100	420	Ahmed et al. (2019) Curk et al. (2016) Luro et al. (2017)
	<i>Vitis</i>	Table grape Grape	2,262 5,738	500	Bonhomme et al. (2020) Candresse et al. (2020) Dumas et al. (2020)
	<i>Olea</i>	Olive	63	0	El Bakkali et al. (2019) Khadari and El Bakkali (2018) Khadari et al. (2019)
	<i>Prunus</i> sp. <i>Juglans</i>	Apricot, almond, peach, cherry, plum Walnut	2960 400	NA	Bernard et al. (2020) Cirilli et al. (2020)
	<i>Malus</i> <i>Pyrus</i> <i>Cydonia</i> <i>Rosa</i>	Apple Pear Quince Rose	4,917 2,474 59 2,518	638	Lassois et al. (2016) Liorzou et al. (2016) Lopez Arias et al. (2020) Muranty et al. (2020)
	<i>Vanilla</i>	Vanilla	321	2	Lepers-Andrzejewski et al. (2012) Lubinsky et al. (2008)
	<i>Musa</i> <i>Ananas</i> <i>Saccharum</i> <i>Mangifera</i> <i>Dioscorea</i>	Banana Pineapple Sugarcane Mango Yam	403 467 335 120 430	600	Arnau et al. (2017) Martin et al. (2020) Umber et al. (2020)
	<i>Vanilla</i> Tropical garlic Roots and tubers Neglected vegetables	Vanilla Garlic Taro, Cassava, Yam, Sweet potato, etc. Beans, peas, etc. Luffa, snake gourd, etc.	700 33 82 103	300	Andriamihaja et al. (2020) Bouétard et al. (2010) Roux-Cuvelier (2017)
	<i>Solanum melongena</i> <i>Capsicum</i> <i>Solanum lycopersicum</i> <i>Cucumis melo</i> <i>Lactuca</i>	Eggplant Pepper Tomato Melon Lettuce	2,333 2,173 3,378 2,332 948	1,595 (distribution out of the research unit)	Daunay et al. (2011) Salinier et al. (2019a, b)

practices and genetic interactions on quality and resistance. The BRC collaborates internationally (e.g., with European and Tunisian gene banks, ECPGR, University of Wisconsin). This BRC focuses on carrot genetic resources with 1,376 patrimonial accessions (among which 350 wild relatives) and nearly 3,000 scientific accessions resulting from research activities (inbreds, segregating populations,

intercrossing populations). It holds a unique collection of tuberous-rooted chervil (23 patrimonial and 683 scientific accessions) and a few accessions of other *Apiaceae* crops. The BRC Carrot and other vegetable *Apiaceae* coordinates the national network of carrots and other *Daucus* genetic resources (including 7 companies and 3 professional organizations), hosts and manages the net-

work collection. It provides expertise and resources in the CTPS (variety registration and maintenance controls). This BRC runs projects in partnership with seed companies, and provides scientific support to participatory breeding process of organic vegetable growers. Knowledge management of wild carrot populations is done in collaboration with national botanical conservatories (e.g.,



■ Figure 1B. A. Diversity of carrot root color. Credit: E. Geoffriau. B. Wild carrot umbels for seed regeneration. Credit: E. Geoffriau.

Bailleul, Porquerolles, Corsica) and a taxonomy expert (Via Apia). At European level, the BRC Carrot and other vegetable *Apiaceae* is the French representative on the ECPGR working group *Umbelliferae* (coordination 2008-2013), and is involved in ECPGR Carrot Diverse and EVA carrot projects. It coordinates collecting missions.

The national patrimonial carrot collection can be accessed through <https://crb-carotte-cn.agrocampus-ouest.fr/>. The BRC Carrot and other vegetable *Apiaceae* is engaged towards ISO 9001-2015 certification.

BRC Citrus (Figure 1C)

In 1959, the BRC Citrus was created by the introduction of material initially from the Mediterranean area, then from other growing areas such as Southeast Asia, the region of origin of citrus. It is located in Corsica, on the INRAE station of San Giuliano, and managed jointly by INRAE and CIRAD. Today, the Citrus BRC has more than 1,100 accessions from more than 50 countries, which constitutes one of the five most important citrus collections in the world. All groups or cultivated species are represented but the group of mandarins and their hybrids are a special-

ty core collection (about ¼ of the BRC). Citrus is conserved in the form of grafted trees (3 or 4 trees per accession) planted in orchards on 14 ha in total. Today, about 25% of the collection is duplicated and maintained in an insect-proof greenhouse. More secure forms of conservation are being studied such as cryopreservation of polyembryonic seeds (apomictic multiplication by somatic embryo). From 2014 to 2020, the Citrus BRC was certified according to the NFS 96-900 standard. In 2020, this was switched to the international quality system ISO 9001-2015. A management software has been developed and consists of modules dedicated to each major activity: introduction, conservation,

Box 3. The origin of citrus fruit – F. Curk (INRAE) and P. Ollitrault (CIRAD)

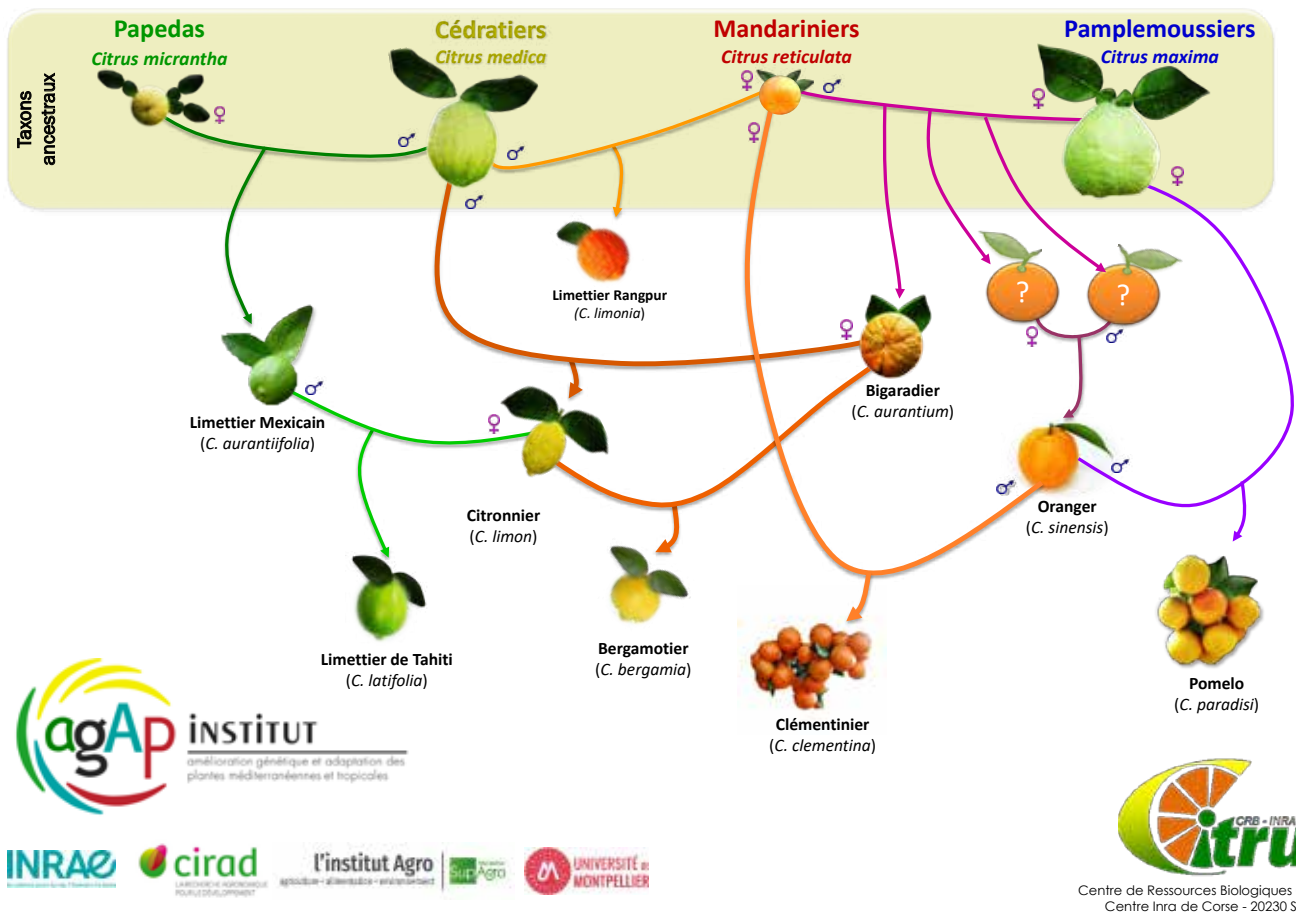
Citrus ancestors first diversified in different regions from the southern Himalayas to Oceania. This so-called allopatric evolution differentiated a number of taxa, of which four (*C. maxima* (Burm.) Merr [pummelos], *C. reticulata* Blanco [mandarins], *C. medica* L. [citrons] and *C. micrantha* Wester [papedas]) are at the origin of the main horticultural groups (Figure 2). Hybridization between these ancestral taxa has occurred as a result of overlapping geographical ranges, certainly linked to major climatic changes. These hybridizations have generated new species, some of which modern man has inherited after identification and selection by humans over millennia. Recent phylogenetic studies have uncovered and confirmed this history.

We now know that the sour orange (*Citrus aurantium* L.) is a direct hybrid between a pummelo and a wild mandarin tree. The orange (*Citrus sinensis* L. (Osborne)) merged from a more complex combination of *C. maxima* and *C. reticulata* genomes. Lemon (*C. limon* (L.) Burm.) is a hybrid between a sour orange and a citron. The citron is the male parent of all acidic citrus, including the Mexican lime (*C. aurantiifolia* (Christm.) Swing.), a direct hybrid between *C. micrantha* and *C. medica*. The most recent results have highlighted the complex origin of the Tahitian lime (*C. latifolia* Tan.), which has a complex mosaic genome derived from the four ancestral taxa. This lime is, in fact, a hybrid between the Mexican lime and the Mediterranean lemon.

In addition to identifying the ancestral species at the origin of cultivated citrus, this information opens the way to new strategies for breeding based on a wide exploitation of the genetic resources of the species complex to generate the genotypes of tomorrow.



■ Figure 1C. A. Citrus field collection. Credit: F. Luro, BRC Citrus. B. Greenhouse citrus collection. Credit: E. Bloquel, BRC Citrus.



■ Figure 2. Genetic origin of the main citrus sub-groups.

characterization, and dissemination of plant material.

Until 1999, the conservatory had its own quarantine strategy. Thermotherapy and regeneration by somatic embryogenesis were used, in the beginning, to guarantee the sanitary status of the introduced material. Then, from 1981, apex micrografting was applied and opened the possibilities of introduction to all varieties of all geographical origins. Since the beginning of the 21st century, the National Agency of Sanitary Safety of Food, Environment and Work (ANSES) of Clermont-Ferrand (mainland France) ensures the quarantine services. Corsica has low pest pressure for citrus. Then, the regular controls enable this BRC to be one of the rare locations where all outdoor trees remain healthy.

Nearly 32% of the accessions are available for distribution through shipment of budwood or seeds (about 170 accessions each year). These forms are available for research on plants, leaves, fruits, and flowers (about 250 accessions per year). The collection supports numerous genetic, agronomic, physiological or biochemical studies, and varietal development by selection for hybridization. The important varietal diversity of mandarins, lemons, limes, and citrons has been exploit-

ed to develop phylogenetic studies and to analyze the structure of genomes (Box 3).

BRC Grapevine (Figure 1D)

The Vassal-Montpellier Grapevine BRC is the oldest and richest ampelographic conservatory in the world, both in number and diversity of accessions. The conservatory was created in 1876 at the Montpellier School of Agriculture (now the Institut Agro | Montpellier SupAgro). During the phylloxera crisis in 1949, INRAE began managing the collection at the Domaine de Vassal site. Under the scientific aegis of the research team on vine genetics in Montpellier (now the AGAP Institute (Genetic Improvement and Adaptation of Mediterranean and Tropical Plants research unit, team “Diversity, Adaptation and Improvement of the Grapevine”), the collection has been studied and enriched continuously. The documentation of the collection and associated data are also of great importance.

Today, the collection consists of over 8,000 accessions of cultivated vines, rootstocks, and wild relatives from 54 countries. The collection conserves 2,262 accessions of table grapes, representing 1,187 different varieties, of which 243 are considered “dual purpose” for wine and table.

The accessions are cultivated in the vineyard on five rootstocks. They are characterized for morphological, phenological, agronomic, technological, sanitary, genetic, and bibliographical traits. In recent years, the emphasis has been placed on behavior in the face of the main pathogens, in particular through the Vitirama project (<https://www6.montpellier.inrae.fr/vassal/Activites/Projets/Vitirama>), to support the use and creation of varieties requiring less phytosanitary treatment. Each year, an average of 500 varieties are distributed as cuttings-graft or fresh material (leaves, berries), to professionals in the sector, amateurs, or French and foreign researchers. The research topics in which the BRC Grapevine is involved are very diverse, from archaeobotany to virology, including physiology and adaptation to climate change.

The documentary collection is a resource in its own right and is being digitized thanks to a sponsorship program. It is used in particular in projects with historians, such as the historical ampelographic atlas of France (CepAtlas, <http://citeres.univ-tours.fr/spip.php?article3174>).

In addition, the BRC Grapevine collaborates with the French Wine and Vine Institute and the French Network of Vine Conservatories,



■ Figure 1D. A. Domaine de Vassal, field collection. Credit: C. Cruells. B. 'Italia Rubi' cultivar. Credit: BRC Grapevine. C. 'Malaga' cultivar. Credit: BRC Grapevine.

which brings together 36 professional partners from all wine-producing regions. The common database for this network is https://bioweb.supagro.inra.fr/collections_vigne/Home.php.

BRC Olive trees (Figure 1E)

The olive tree (*Olea europaea*) is one of the emblematic species of the Mediterranean. It is cultivated for the production of edible oil and table olive on a surface area of over 10 million hectares worldwide. It is an "immortal" tree, particularly because of its ability to regenerate after extreme weather events. *Olea* includes six subspecies, but only the olive tree *Olea europaea* subsp. *europaea* was domesticated about 6,000 years ago. This subspecies is present throughout the Mediterranean, both in its wild (var. *sylvestris*) and cultivated (var. *europaea*) forms. Domestication *sensu stricto* began with the vegetative multiplication of wild trees selected for traits probably related to fruit size and oil content. The BRC Olive trees is supported by the National Mediterranean Botanical Conservatory (CNBMed), based in Hyères-les-Palmiers in PACA (Provence-Alpes-Côte d'Azur) region, and managed jointly by INRAE, within the framework of the AGAP Institute in Montpellier.

The BRC focuses on the national olive tree collection, hosted by the Port-Cros National Park on the Porquerolles Island (PACA region). The core collection is composed of French heritage varieties and those from other Mediterranean countries. In partnership with the olive-growing professional organization (France-Olive), a French national collection has been established, constituted by 63 formally identified accessions and considered as reference of French olive varieties. This has been possible following a huge identification work based on morphological (tree, leaf, fruit, kernel), molecular (microsatellite markers) and bibliographical (validation of

names) information and according to the opinions of the France-Olive stakeholders. This identification work continues to remove any ambiguities about certain varieties for which the reference genotype has not yet been validated.

The BRC Olive trees is based on a collection located on an island within a national park, following an organic cropping system. Moreover, its management takes into account a national level through local collections managed in partnership with France-Olive stakeholders, and an international level through world collections managed within the genetic resources network of the International Olive Oil Council (IOC). BRC Olive trees organization ensures a safeguarding of French genetic resources *ex situ* in local (France-Olive network) and worldwide collections (IOC network), as well as *in situ* through trees geo-located by France-Olive in orchards.

The BRC Olive trees is considered an innovative research tool on adaptation to climate change conducted by the AGAP Institute in collaboration with national and international partners mainly in Morocco and Spain (OliveMed project funded by Agropolis Foundation, 2021-2025). Two research topics are currently implemented within this framework: i) understanding the flowering processes in relation to the cold requirements of varieties, and ii) investigating the variability of functional traits related to drought resistance in olive trees (<https://umr-agap.cirad.fr/en/research/main-projects/olivemed>). These research lines are linked to NGS (next generation sequencing) analyses in order to identify adaptive genomic variants.

BRC *Prunus*-*Juglans* (Figure 1F)

The BRC *Prunus*-*Juglans* is managed by INRAE, in orchards or in sheltered containers of fruit and related species belonging to the genera *Prunus* (stone fruit) and *Juglans* (walnut). A large part of this material is composed of tra-

ditional and modern varieties of agronomic interest. Another part is made of accessions of scientific interest, mainly resulting from INRAE research work. Related species are also included in the collections: some are of interest as rootstocks, others may carry genes of interest such as disease resistance ones. Japanese cherry (*Prunus serrulata*), highly prized for its abundant flowering, and myrobolan (*Prunus cerasifera*), widely used as rootstock for plum, apricot or almond trees, are among the best-known species of the *Prunus* collections. These trees are grown and observed in two production regions in France:

- In Provence-Alpes-Côte d'Azur (south-east of France) the INRAE-GAFL (Genetics and Breeding of Fruit and Vegetables) research unit manages exclusively stone fruit trees: apricot (600 accessions), almond (250 accessions) and related species;
- In Nouvelle-Aquitaine (south-west of France), the Fruit tree Experiment unit of INRAE manages both *Prunus* and *Juglans* trees: cherry (3 species, 500 accessions), peach (1 species, 500 accessions), plum (1 species, 350 accessions), and related *Prunus* species (30 species, 70 accessions); and walnut (15 species, 40 accessions).

The *Prunus* collections are used by research units of the "Biology and Plant Breeding" department of INRAE, involved in varietal innovation or stress resistance. This is illustrated by the CASDAR-funded project "Caress *Prunus*" (Characterization of *Prunus* genetic resources for biotic and abiotic stresses). This project characterized the available genetic resources through a multi-criteria approach targeting phenology (dormancy, flowering, maturity and senescence) and sensitivity to pests and diseases. The *Juglans* collections are used both for breeding purposes in a partnership with the Interprofessional Fruit and Vegetable Technical Centre (CTIFL), and for research purposes, particularly in animal



■ Figure 1E. A. Turning color olives of ‘Petit Ribier’ cultivar. Credit: J.-P. Roger, CBNMed. B. Organic clay treatment against the fly *Bactrocera olea* in the French national collection located in the Parc National de Port-Cros Porquerolles. Credit: M. Delalande, INRAE.



■ Figure 1F. A. Flowering *Prunus* from the Rose-pom collection. Credit: BRC Rose-pom. B. Flowering of walnut in new plantation. Credit BRC *Prunus-Juglans*.

physiology in a research project developing plant hormonal compounds in goat breeding.

BRC Pome fruits and roses (RosePom) (Figure 1G)

The BRC “Pome fruits and roses” (BRC RosePom) is hosted by INRAE near Angers, in the region Pays de la Loire, on two main sites: Beaucouzé and La Rétuzière. It is managed by the joint research unit Research Institute in Horticulture and Seeds (IRHS), and by the experimental unit Horti. It includes collections of i) apple, pear, and quince accessions, mostly in the field and as DNA samples, and ii) rose accessions. Scientific accessions of the *Rosa* genus are preserved in the field and as DNA samples, whereas patrimonial accessions are mostly as DNA samples, the heirloom field roses being preserved by different private and public rose gardens in France. The missions of the BRC RosePom include:

- The preservation of biological resources of pome fruit and roses, including improvement and rationalization of the collections;

- The phenotypic and genotypic characterizations of these resources, to be able to supply samples and data for research and breeding purposes, mainly in the framework of collaborative projects;
 - The provision of expertise and available materials from the pome fruit collections for distinctness, uniformity and stability (DUS) testing;
 - Data analyses associated with these resources in collaboration with research teams and external germplasm managers.
- BRC RosePom also leads or contributes to pome fruit and rose genetic resources networks at national, European and international levels. The BRC RosePom currently preserves nearly 10,000 accessions in the field (4,917 *Malus*, 2,474 *Pyrus*, 59 *Cydonia*, and 2,518 *Rosa*) and several thousand additional accessions as DNA samples only. To efficiently store all associated data, a database is under construction and should soon be available. This resource, whether patrimonial or scientific, is the basis of many research projects. For

example, the CorePom project funded by the Foundation for Research on Biodiversity produced an unprecedented increase in knowledge about the accession identity and uniqueness/duplicate status of the French apple germplasm genotyped with SSR. The genetic diversity and structure were analyzed and helped in constructing core collections. A first parentage analysis was also performed. In the EU-funded FruitBreedomics project (<https://cordis.europa.eu/project/id/265582>), more than 2,400 accessions from 14 European collections, including that of the BRC RosePom, were further analyzed and revealed a prominent gene flow in apple at the European level. The same accession set was also used in a genome-wide association study on flowering and ripening periods and for the reconstruction of an extra-large, multi-generation, highly-connected pedigree. For rose, the DNA stored in the BRC of French cultivars of the 19th century allowed the FlorHiGe project (2013-2016) to show introgression of the European genetic background by the Asian one. Rose mapping prog-



■ Figure 1G. A. An overview of the different colors and fruit shapes of the apple varieties conserved in the BRC RosePom. Credit: L. Feugey, INRAE. B. Scientific genetic resource collection of *Rosa* at INRAE, Beaucouzé, France. Credit: Thouroude, INRAE.

enies present in the BRC RosePom fields are the support of several quantitative trait loci studies on different traits, including scent of roses, prickle and disease resistance. Other projects on rose diversity such as those on *Rosa gallica*, or the RosesMonde project (2015-2019) highly contributed to enrich the DNA collection of the BRC RosePom.

BRC Tahitian vanilla (Figure 1H)

The Tahitian vanilla BRC is located on the island of Raiatea, in French Polynesia. It is managed by the Etablissement Vanille de Tahiti (EVT) and was labelled by the GIS IBISA in 2015. Its quality management system is based on the ISO 9001 standard.

This BRC includes 71 vanilla accessions (*Vanilla × tahitensis*, *V. planifolia*, *V. pompona* and hybrids) conserved in two shaded greenhouses, plus 250 varietal creations conserved in vitro. Out of these 321 accessions, 316 are only present at the BRC Tahitian vanilla (Polynesian vanillas and hybrids created by the EVT). Five accessions are also present in the BRC Vatel: *V. planifolia*, *V. pompona* and *V. × tahitensis* 'Haapape'.

The analysis of the genetic diversity of the vanilla plants in the collection has provided insight into the secondary diversification of vanilla plants in French Polynesia and unrav-

eled their hybrid origin. Moreover, genetic analyses have shown that the majority of varieties found in Polynesian plantations result from spontaneous germination of seeds of the 'Tahiti' cultivar, or by natural polyploidisation (doubling of the number of chromosomes) for the 'Haapape' and 'Tiarei' cultivars, for example. These elements highlight the specificity of Polynesian vanilla plants. They have made it possible to define protection and development strategies such as local regulations and the PDO certification process currently underway.

The most widely grown cultivar in French Polynesia is *V. × tahitensis* 'Haapape', which is vigorous but not very floriferous and of average aromatic quality. In the Tahitian vanilla BRC, various vanilla varieties have been evaluated as part of a program to select better performing varieties, i.e., which are better adapted to climatic hazards and emerging diseases. The selection is made by monitoring agronomic traits in the EVT greenhouse, at Polynesian producers' farms, and in laboratory by in vitro tests of susceptibility to fusariosis. Some accessions selected in this way are now offered to producers. These are varieties with excellent aromatic quality and high flowering capacity, or more tolerant to fusariosis.

BRC Tropical plants (Figure 1I)

In Guadeloupe and Martinique, the French Caribbean Islands, CIRAD and INRAE have been building up large collections of tropical crop genetic resources for several decades. In 2010, the two organizations joined forces to create the Biological Resource Center "Tropical Plants-Antilles" (BRC-TP). Four collections of vegetatively propagated horticultural species, conserved in the field, make up the BRC: banana, mango, pineapple and yam. These collections are managed under quality assurance, BRC being certified according to the NF-S 96 900 standard and in the process of being certified for the ISO 9001-2015 standard. The BRC also hosts the Guadeloupe Herbarium, which contains more than 12,000 botanical plates. It actively participates in the BRC4Plants networking activities at national level.

The banana collection comprises 403 accessions representing a significant proportion of the world's banana diversity. It is maintained in the field and secured in vitro for the most fragile accessions. The following types of banana are represented: plantain, cooking, dessert, ornamental and wild bananas, including *Musa acuminata* and *Musa balbisiana*, the ancestral relatives of cultivated banana. This collection is part of the Musa-LAC (Latin America and Caribbean) network supported by Bioversity International. It is involved in numerous research projects in genetics and banana breeding. It is one of the most important banana field collections in the world in terms of the genetic diversity and number of accessions conserved.

The yam collection, managed by INRAE, is composed of 430 accessions conserved in vitro, part of which is renewed in the field each year. The main species represented are *Dioscorea alata*, *D. trifida*, *D. cayenensis-rotundata*, *D. bulbifera* and *D. esculenta*. Virology research is conducted on this collection, which is also used in international projects (e.g., RTBfoods, <https://rtbfoods.cirad.fr/>; Africayam, <http://africayam.org/>).

The mango collection is a historical CIRAD collection. It comprises 120 accessions kept in the field, mainly of the *Mangifera indica*



■ Figure 1H. A. Cured beans of Tahitian vanilla. Credit: T. McKenna. B. Tahitian vanilla flower. Credit: EVT.



■ Figure 1. A. Pineapple field collection in Martinique. Credit: M. Roux-Cuvelier. B. Diversity of banana fruit. Credit: BRC Tropical Plants. C. In vitro plantlet (yam collection). Credit: M. Roux-Cuvelier. D. Diversity of mango fruit. Credit: BRC Tropical Plants.

species. Seventy percent of the genotypes come from Africa. Recently enriched with heritage varieties from the French Caribbean Islands, this collection is used to diversify mango production.

The pineapple collection, located in Martinique, includes 467 wild and cultivated accessions representative of the diversity of the genus and of the Amazon Basin, the pineapple area of origin. The pineapple collection is currently conserved in the field, but should be secured by cryopreservation in a near future. It is one of the most important pineapple collections in the world.

The BRC is responsible for the development of OLGA software (local accession management tool), a computer tool used by many BRCs in France to manage stocks of genetic resources and associated data. The collections maintained within the BRC are listed on the Florilège web portal.

BRC Vatel (Figure 1)

The BRC Vatel (vanilla, garlic, tubers and vegetables) is located on Reunion Island, in the Indian Ocean. It is managed by CIRAD as part of the Peuplements Végétaux et Bio-agresseurs en Milieu Tropical research unit (CIRAD/University of Reunion Island). Four collections of agricultural plant genetic resources are conserved in the BRC Vatel:

- Vanilla: 700 accessions, 25 species of the genus *Vanilla* (Orchidaceae);
- Tropical garlic: 33 varieties of *Allium sativum* (Amaryllidaceae);

- Root and tuber vegetables: 82 accessions, 11 species (cassava, taro, yam, sweet potato, etc.), 7 families (e.g., *Euphorbiaceae*, *Araceae*, *Dioscoreaceae*, and *Convolvulaceae*);
- Seed vegetables: 103 accessions, 30 species (including squashes, pigeon peas, cowpeas), 9 families (mainly *Cucurbitaceae* and *Fabaceae*).

The vanilla collection includes accessions of the three cultivated species (*Vanilla planifolia*, *V. pompona* and *V. × tahitensis*) as well as 25 wild relatives and interspecific hybrids. It is one of the most important collections worldwide for the genus *Vanilla*. In recent years, the BRC has contributed to major work on the genetic diversity and evolution of cultivated and wild vanillas, on the elucidation of the mechanisms of biosynthesis and storage of aromatic compounds in the fruits, and on resistance to fusariosis. It is also involved in the creation and selection of improved vanilla genotypes. Since 2018, the BRC Vatel has been coordinating the VaniSeq project carried out by a French public-private consortium to decipher the genome of the main cultivated vanilla species *V. planifolia*. The three collections of garlic and vegetables are made up of traditional varieties from the southwestern Indian Ocean region, most of which were collected in Reunion Island. These collections meet the challenge of conserving an agricultural and cultural heritage (“legume lontan”), but are also of interest in terms of food diversification in the region.

The BRC Vatel is using four CIRAD facilities in Reunion Island: the laboratories of the Plant Protection Platform and three field stations. The accessions are kept in the form of living collections cultivated in the field or in greenhouses (vanilla, garlic, root and tuber vegetables) or seed collections in cold storage (seed vegetables). Part of the accessions are duplicated by in vitro tissue culture for vanilla, cassava, yam and sweet potato.

Of the 918 living accessions conserved at the BRC Vatel, about 200 are available for distribution. Seed samples (seeds, lianas, bulbs or tubers depending on the species) can be ordered online on the portal of the French tropical BRC network <http://intertrop.antilles.inra.fr/Portail/pages/crb-vatel>. On average each year, 300 accessions are distributed to around 30 clients, for agricultural, educational, research or recreational use.

The Vatel BRC has been awarded BRC by the GIS IBISA label since 2009. The collections are managed according to a quality assurance approach and certified under the French standard NFS 96-900 since 2016, then ISO 9001-2015 standard since 2019.

BRC Vegetables (Figure 1K)

The Centre for Vegetable Germplasm (BRC-Leg) is part of the INRAE-GAFL research unit situated in Avignon, Provence (PACA region) (https://www6.paca.inrae.fr/gafl_eng/Vegetable-Germplasm-Centre). It maintains over 10,000 accessions spread across five collections, as follows:

- Eggplant (*Solanum melongena* and relatives, 2,333 accessions): the national collection is part of the multilateral system since the species is contained in Annex 1 of the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA);
- Pepper (*Capsicum annuum* and related species, 2,173 accessions);
- Tomato (*Solanum lycopersicum* and related species, 3,778 accessions);
- Melon (*Cucumis melo*, 2,332 accessions);
- Lettuce (*Lactuca* spp., 948 accessions).

The genetic resources held by the BRC-Leg are mostly used by French public institutions (research and development), and by breeders of seed companies, who actively collaborate with each other. The accessions are regenerated and described by network associations of public and private partnerships. For each crop, collections are split into two parts: i) the freely available national collection, and ii) the networking collection, access to which is subject to specific conditions. The BRC-Leg coordinates three networks: *Solanaceae* (three crops), melon, and *Lactuca*. It also provides free samples to researchers from foreign public institutions, growers, NGOs and other bona fide users who request the acces-



■ Figure 1J. A. Field maintenance of the garlic collection at a farm in Petite Ile (Reunion Island). Credit: M. Seguin, CRB Vatel. B. Display of the diversity of vegetable seeds for educational purposes. Credit: C. Charron, CRB Vatel. C. Maintenance of the vanilla collection under insect-proof shade-house. Credit: M. Grisoni, CRB Vatel.



■ Figure 1K. A. An example of the fruit diversity of *Solanum melongena* (eggplant) and related species maintained at the "CRB-Leg". Credit: CRB-Leg. B. A pepper (*Capsicum*) flower from one of the accessions of the *Capsicum* collection maintained by the BRC "CRB-Leg". Credit: CRB-Leg. C. An example of melon (*Cucurbitaceae*) fruit diversity in the collections of the "CRB-Leg". Credit: CRB-Leg.

sions through the INRAE database (<https://urgi.versailles.inra.fr/siregal/siregal/grc.do>). This database contains passport data and descriptions of each accession. It is linked to other databases such as Florilège (national database) and Eurisco (European database). The duty of the BRC-Leg is to carry out morphological descriptions in the open field or in greenhouse. Historically, this activity has mostly included traits related to the harvested product (fruit or leaves). Evaluation has so far typically focused on resistance to pathogens and fruit quality. GAFL scientists are developing research programs to include root traits in the plant descriptions. Both the patrimonial collections and the scientific genetic resources (segregating populations, mutants, etc.) of the BRC-Leg are used for research topics currently including: salt resistance in tomato (ERA-NET root project,

<https://www.suscrop.eu/projects-first-call/root>), or *Bremia* resistance in lettuce (<https://www.ecpgr.cgiar.org/european-evaluation-network-eva/eva-networks/lettuce>). The BRC-Leg genetic resources are part of the European G2PSol project on biodiversity in the *Solanaceae* (www.g2p-sol.eu).

All seeds are conserved at 5 °C under controlled humidity, and the daily work includes the transfer of the collections to -20 °C for long-term storage. The BRC-Leg is also currently moving towards ISO 9001 quality certification.

Conclusion

In recent years, and more particularly since the implementation of the CBD, the ITPGR, and the Nagoya Protocol, the holders of genetic resources in France have organized themselves around the constitution of net-

works allowing for the global consideration of common problems and the easy sharing of information.

The management of French horticultural genetic resources has benefited from recent technological advances that have enabled the acquisition of mass data, particularly in the fields of genotyping and phenotyping, thus enriching the intrinsic value of conserved genetic resources. The horticultural BRCs and collections now play an essential role in French agronomic research, particularly for the preservation and enhancement of agrobiodiversity and the agro-ecological transition of agriculture.

Like most agricultural genetic resources, public funding for the conservation of horticultural diversity is often precarious. However, it has tended to gain in sustainability in recent years thanks to a growing institutional awareness of its crucial importance for the food of tomorrow.

The horticultural genetic resources conserved by the French BRCs are under the responsibility of national public organizations that use them for agricultural research and development. They are also widely used in breeding and varietal improvement programs conducted by the private seed and plant sector. The French horticultural BRCs have made particularly significant contributions to the development of banana, clementine, apple and vegetable *Solanaceae* and *Apiaceae* by several seed companies.

Finally, the French BRCs located in tropical environments in the French overseas departments and territories, play a critical role in the preservation of tropical agricultural genetic resources. They contribute to the food security and sovereignty of territories that are particularly threatened by climate change and the standardization of eating habits. This point will be debated during the United Nations Food Systems Summit in October 2021, and also discussed during the 31st International Horticultural Congress (www.ihc2022.org) to be held in Angers (France) from 14 to 20 August 2022. ●



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