

# ECPGR Activity Grant Scheme – Fourth Call, 2016 Final Report

## Common ECPGR protocols and tools available for Characterization & Evaluation of *Malus/Pyrus* genetic resources (Pome Fruit C&E)

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#### INTRODUCTION

Characterization and Evaluation (C&E) of agronomic traits is fundamental to the management and utilization of our genetic resources. Appropriate methods, protocols and tools need to be adapted and updated to allow the coordination of such activities. Alongside the 2012 meeting of the Malus/Pyrus Working Group (WG), a half-day ad hoc meeting on Malus/Pyrus phenotypic descriptors was organized and the importance of finalizing an agreed document updating phenotypic descriptors was highlighted as a priority for the WG.

Following the meeting, document drafts (Methods and Descriptor Lists for the Characterization and Evaluation of Biodiversity) for both apple and pear were produced, but these remained in early draft form and the WG had been unable to produce a final version for adoption within ECPGR.

The value of using harmonized molecular markers for tracking errors and redundancy within and between collections was also highlighted during the 2012 WG meeting. In an equivalent context, several European apple collections had been, or were currently being, genotyped with SSR markers within the framework of national or European projects (e.g. 'Fingerprinting the [UK] National Apple & Pear Collections' Defra project GC0140; 'FruitBreedomics' EU 7<sup>th</sup> Framework Programme N°265582). For pear, initial work to coordinate efforts had been done, to a lesser extent (limited to 140 accessions) in the ECPGR EcoHisPy project. Several groups, with ECPGR representation, had already published results of SSR genotyping in larger apple and pear collections.

Whilst a number of the genotyping efforts had aligned with the basic <u>ECPGR recommendations for the use of standard markers</u>, considerable work was still required to adjust and align datasets in a coordinated way. With the view to building a common tool for comparing data across collections, further standardization work still needed to be done to harmonize marker data for them to potentially be stored in EURISCO as a centralized open-access repository.

The project thus aimed to have two complementary objectives with activities planned over a 24-month period: (i) The first objective was to finalize and implement the ECPGR adopted 'Characterization and Evaluation Descriptors for Apple and Pear Biodiversity'; (ii) The second objective was to harmonize SSR data across genotyped collections of European pear, in order to make them available for users and suitable for potential submission to EURISCO.

For the first objective, a coordinator (Marc Lateur, Belgium) and a co-coordinator for each crop (apple: Matthew Ordidge, UK; pear: Monika Höfer, Germany) were designated to ensure that efforts were coordinated in a timely manner. Electronic coordination meetings were planned to be organized by co-coordinators at least each trimester. The work plan proposed was as follows: (i) circulate the existing versions of ECPGR apple and pear descriptors to partners and crop experts for revision and comment; (ii) compile comments and suggestions, and recirculate; (iii) organize a 2-day meeting with one day per crop for agreeing the final content of the document; during the meeting, a common characterization and evaluation test would be agreed for studying the applicability/reproducibility of assessments of selected traits on a limited set of common cultivars; (iv) implement the common test and collate data in a standardized file; (v) analyze data and compile a summary report; (vi) adopt the final version of ECPGR 'Methods and Descriptor Lists for Apple & Pear' and send to the ECPGR Secretariat for final editing in electronic format.

The second objective was coordinated by C.E. Durel (France) who had previously coordinated similar work on apple (Urrestarazu et al., 2016) with the help of Matthew Ordidge (UK) as co-coordinator. The proposed work plan was as follows: (i) contact scientists who had already published or produced pear SSR data within ECPGR countries; (ii) establish a collaborative agreement favouring the sharing of SSR data while guaranteeing preliminary publication by the data owners; (iii) collect SSR and passport data from partners having signed the agreement; (iv) collect leaf samples from representatives of each dataset for additional genotyping to aid with allele alignment; (v) harmonize data across the collections

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to identify redundancy, putative synonyms and mutants or errors that can be further checked in each collection; (vi) prepare an SSR data file suitable for upload into the EURISCO database and to be made available through an online searchable function to be developed (in kind) as an update to the ECPGR *Malus/Pyrus* database; (vii) finalize and submit for publication a genetic diversity and structure analysis of European pear. In line with the analysis, a subset of the (8) most discriminant SSR markers that can be associated in "multiplex" will be identified and recommended for simplified identification/distinction of new collections. In parallel, since the work done previously by INRAE Angers to harmonize SSR marker data for apple was mostly finalized across several thousand European accessions from ten different collections, an apple SSR data file would be produced ready for upload into the EURISCO database as well as being made available through the ECPGR *Malus/Pyrus* database tool.

#### MATERIALS AND METHODS/APPROACH

## Finalize and implement characterization and evaluation descriptors for apple and pear

During the expert meeting in April 2017 at Gembloux (attended by 15 participants from 12 countries), a set of updates and amendments were agreed to standardize both within and between the descriptor documents for apple and pear. These amendments were based on comments added to the originally circulated documents and all comments were discussed in detail during the meeting. All amendments were subsequently incorporated into a final draft.

During the expert meeting, a common characterization and evaluation test was also planned in order to assess the applicability/reproducibility of commonly defined priority descriptors across a range of proposed reference cultivars during the 2017 season. Consequently, a working document updated with the agreed amendments to the priority descriptors to be included in the test was sent to all partners during summer 2017. A summary of the agreed recommendations on gathering standard photographs was also made available to all.

For pear, a total of 12 traits (5 for characterization and 7 for evaluation) and 18 descriptors were agreed to be applied in each collection on a common set of reference cultivars. Where reference cultivars were missing, it was agreed that each collection should propose alternative replacements (Table 1). For apple, a similar approach was agreed across a total of 14 traits (5 for characterization and 9 for evaluation) with 20 descriptors on common reference cultivars (**Table 1**). A common template was circulated in order for partners to encode their data into a single file for collation.

Due to high frost conditions during the 2017 flowering period, it was not possible to collect data on some accessions/collections and it was agreed to extend the approach into 2018/19.

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**Table 1** Common apple & pear reference cultivars (as proposed in descriptor documents). Cultivars in bold indicate the preferred name.

Apple	Pear
'Alkmene'	'Abate Fetel' (syn. 'Abbé Fétel')
'Åkerö'	'Beurré Alexandre Lucas' (syn. 'Alexander Lucas' 'Alexander Lucas Butterbirne', 'Lucasova Maslovka')
'Ananas Reinette' (syn. 'Reinette Ananas')	<b>'Beurré de Mérode'</b> (syn. Double Philippe, Doyenné Boussoch)
'Discovery'	'Beurré d'Hardenpont' (syn. 'Glou Morceau', 'Beurré d'Arenberg' – in France,'Ardenpont d'Inverno', 'Butirra d'Hardenpont d'Inverno', 'Hardenpontova Maslovka', 'Hardenpont's Winterbutterbirne')
'Golden Delicious'	'Beurre Hardy' (syn. 'Hardy', 'Gellerts Butterbirne', 'Butirra Hardy')
'Ingrid Marie'	'Beurré Superfin' (syn. 'Butirra Sopraffina', 'Hochfeine Butterbirne')
'James Grieve'	'Blanquilla' (syn. 'Spadona')
'Jonathan'	'Clapp's Favourite'
'King of the Pippins' (syn. 'Reine des Reinettes', 'Winter Goldparmäne')	'Comtesse de Paris' (syn. 'Gräfin von Paris', 'Paris')
'Reinette de Champagne'	'Conference'
'Winter Banana'	'Doyenne du Comice'
'White Transparent' (syn. 'Transparente Blanche')	'Durondeau' (syn. 'Tongre', 'Poire de Tongre', 'Tongern', Beurré Durondeau')
	'Kontoula'
	'Kristalli'
	'Légipont' (syn. 'Fondante de Charneux', 'Köstliche von Charneux')
	'Louise Bonne d'Avranches'
	'Nec Plus Meuris' (syn. 'Beurré d'Anjou' 'Anjou Pear', 'Butirra d'Anjou')
	'Nouveau Poiteau' (syn. 'Neue Poiteau', 'Patawinka Poiteau',
	'Précoce de Trévoux' (syn. 'Frühe aus Trévoux')
	'Williams' Bon Chrétien' (syn. 'Bartlett', 'Williams', 'Williams Christbirne')

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## Harmonize SSR data across genotyped collections of European pear and apple

#### Pear:

The initial approach upon contacting researchers who held data for potential alignment was to try to establish a formal collaborative agreement for data sharing. However, due to the wide range of partners that became involved, and the tendency for the partners with concerns over the timing of data sharing (for reasons of publication) to want to delay sharing data regardless, it was deemed that a formal agreement would not be helpful, or necessary. Data were subsequently collected on the basis of an informal bilateral agreement between INRAE (representing the project) and each data provider documented by e-mail. At that stage, the bilateral agreement was limited to making the SSR data available for genetic profile comparison and assignment into duplicate groups (PUNQ/MUNQ assignment, see below). The global sharing of each individual SSR data was not part of the agreement and should be confirmed later according to the wishes of the data owners to have first their own preliminary publication.

Following the initial analysis of shared data, five partners were asked to send to Angers new leaves of specific pear accessions in order to be re-genotyped to help with the alignment of scored data. Overall, complementary genotyping was performed on ~530 accessions from Belgium, UK, Spain, Germany and Sweden with 2 additional multiplexes (MP) containing 5–9 SSR to achieve 19 SSR per genotype. The additional genotyping of 288 accessions (3 x 96-wells plates) was initially agreed following a budget revision in March 2018. However, in part due to the response from collaborators, the complete genotyping (4 MP, 19 SSR) of ~500 accessions from various origins including Ukraine, Russia, Romania, Finland and Latvia was performed. This additional set included 86 accessions from the previous EcoHisPy project which needed re-genotyping for adequate data alignment. Many Eastern Europe and Baltic countries were thus represented. Unfortunately, no samples were sent from Portugal, Turkey and Caucasian countries despite initial discussions and requests.

Additional contacts were established with Nahla Bassil from the USDA Pear National Clonal Germplasm Repository at Corvallis for developing collaboration on SSR analysis. Additional genotyping (2 to 3 MP) was performed on 565 accessions from the USDA pear collection from Corvallis in an attempt to allow harmonization with this dataset.

Analysis of the overall harmonized dataset was carried out with the view to track putative synonyms, duplicates and/or errors. Specifically, a *Pyrus* UNiQue genotype code (PUNQ) analogous to the *Malus* UNiQue genotype code (MUNQ) (Muranty et al., 2020) was allocated to each unique SSR genotypic profile. Initial attempts to analyze genetic diversity were carried out using STRUCTURE software.

#### Apple:

For apple, it was established that some partners and other interest groups also held datasets that they were keen to be aligned with the existing harmonized SSR database (built by INRAE in the framework of the EU FruitBreedomics project). Specifically, three important new collections (in Denmark, Ireland and Germany) decided to share their data (2,266 accessions) on the same basis as was decided for pear. Again, re-genotyping of representative samples of accessions from those new collections was necessary to improve the alignment of scored data. Entries in the database were allocated MUNQ codes in order to be able to track and identify duplicate material.

As for pear, SSR data were received from Gayle Volk (USDA Apple National Clonal Germplasm Repository at Geneva) and were tentatively analyzed by INRAE, but no DNA was available for newly genotyping at that time.

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### RESULTS

## Finalize and implement characterization and evaluation descriptors for apple and pear

Final drafts of the descriptor documents for both <u>apple</u> and <u>pear</u> have been submitted to ECPGR Secretariat which performed a last check and an improved layout (2022) for final edit and/or distribution (ANNEX II). The descriptors are largely adapted from a series of other documents (IBPGR/CPVO/UPOV descriptors etc.). The adapted descriptors are standardized as far as possible both within the crop and between apple and pear. In addition, a level of priority (first/second/third) is allocated to each descriptor.

A dataset detailing the priority descriptors for both apple and pear was produced across a set of proposed reference cultivars in the different countries represented by the partners (**Table 2**). Between 2 and 12 reference cultivars were tested per country for apple and between 4 and 12 reference cultivars were tested per country for pear (**Table 3**). In total, 2,734 and 2,811 data points were collected for apple and pear, respectively. For apple, seven partners (Belgium, France, Germany, Latvia, Poland, Romania, United Kingdom) were able to collect data from a minimum of eight reference cultivars whilst for pear, the number of partners collecting data for a minimum of eight cultivars was five (Belgium, France, Germany, Poland, United Kingdom).

Standard photographic images were also shared by partners in Estonia, Spain, France, Norway and the UK.

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**Table 2** Amalgamated Characterization & Evaluation data on reference cultivars

APPLE	gamated Characterization & Evaluation data o		PEAR		APPLE	
		# Partners	# Data	# Partners		
Partner				12		
Variety				4 to 12		
Year				2		
F1 (	date	2	71		87	
F2 (	date		89		108	
Flowerin	gintensity		106		117	
Flowerii	ng period		97		121	
Intensity of sec	ondary flowering		64		11	
Frequency of sec	ondary flowering		46		0	
Harvest	ing date		94		123	
Optimal ripening	stage assessment		92		98	
Harvest	maturity		91		122	
Fruit	shape		87		133	
	Profile		95			
	Relative position of maximal diameter		95			
atio of fruit height to width mean	Ratio of fruit lenght to maximum diameter		96		133	
Frui		95		133		
	Depth of fruit stalk cavity		95			
Amount of over	colour coverage		96		130	
Over colour of full	y mature fruit skin		78		112	
Global amount o	frusset coverage		96		131	
Global tree foliage	health assessment		88		114	
	Harvesting date		93			
	Ripe eating maturity		94			
Fruit fi	rmness		86		111	
Flesh sv	veetness		89		110	
Flesh	acidity		87		110	
Ratio acidity / sw	eetness of flesh		89		110	
Flesh jı	uiciness		92		110	
Flesh cr	uchiness		83		110	
ntensity flesh overall aroma	Intensity of "musky" aroma		77		110	
	Fruit flesh texture		94			
	Importance of grit cells (around the core)		79			
Overall fruit quality			87		112	
Fruit mean weight (g)			19		11	
uit firmness measured with netrometer 11 mm probe without skin Mean firmness (penetrometer)			38		45	
veetness of flesh measured with fractometer Mean sweetness (refractometer - Brix)			38		46	
ruit eye cavity/stalk basin width mean atio					22	
roma type					3	
Acidity						
	dity		2		4	

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**Table 3** Number of reference cultivars from which data were collected by different partners (2017-2019). BA: Bosnia and Herzegovina; BE: Belgium; CZ: Czech Republic; DE: Germany; EE: Estonia; ES: Spain; FR (France); GB: United Kingdom; LV: Latvia; NO: Norway; PL: Poland; RO: Romania; SI: Slovenia.

Partner	Country	Apple		Pear		
		2017	2018	2017	2018	
1	FR	5	5	8	8	
2	DE	8	10	9	9	
3	LV	11	11 *	1	1	
5	BE	10	10	9	9	
6	EE	4				
7	NO	7				
8	GB	11		12		
9	BA	1	1	2	2	
11	CZ				10	
17	PL	8	10	7	7	
20	ES	2	1	2	2	
22	SI	5	3	4	4	
	RO	8	8	5	5	

<sup>\*</sup> plus 11 from 2019

As a consequence of carrying out the common test, all partners were familiarized with the agreed priority descriptors and could suggest any improvement of them.

Finally, as one of the most important deliverables of this Grant Scheme Activity, the updated and enhanced ECPGR Descriptor lists and methods for characterisation and evaluation of apple and pear genetic resources have been published by the ECPGR Secretariat (Lateur et al. 2022a, Lateur et al. 2022b).

## Harmonize SSR data across genotyped collections of European pear and apple

#### Pear

Data sharing agreements are in place for mutual sharing of the PUNQ code of each accession between INRAE-Angers and the following partners: INRAE and Union Pomologique de France (France), NFC, EMR, FruitID and Hartpury Heritage Trust (UK), ISSA Orchard (Ireland), JKI and Ökowerk collection (Germany), Univ. Copenhagen (Denmark), SLU (Sweden), LUKE (Finland), Institute of Horticulture (Latvia), LAMMC (Lithuania), RBIPH (Czech Republic), Research Institute for Fruit Growing (Romania), Univ. Ljubljana (Slovenia), Univ. Sarajevo (Bosnia and Herzegovina), HCPHS (Croatia), FOAG and Agroscope-Fructus (Switzerland), UNIBO, CREA and Laimburg Research Center (Italy), CITA and Univ. Navarra (Spain), VNIISPK and NCRRIH&V (Russia), Institute of Horticulture (Ukraine), and USDA (USA). Previously published SSR data from the European FruitBreedomics project and the French CorePom project were also considered for sharing, together with samples from the former ECPGR EcoHisPy project.

A new set of 19 SSR was defined for pear, including 11 out of the 12 previously defined ECPGR SSR set supplemented by 8 SSR described in the literature. SSR data were standardized and/or analyzed for 4,912 accessions collected from 26 European collections: more than 1,000 accessions originating from 20 European collections were newly genotyped with the 19 SSR; ~3,900 accessions originating from 6 European collections holding existing SSR data were standardized through the development of dedicated allelic correspondence tables. Following the removal of missing data, a dataset with at least 9 standardized SSR across 4,861 accessions was produced. This is sufficient for the identification of duplication with limited risk of error. Among these 4,861 accessions, 2,176 distinct genotypes were identified. A PUNQ code (Pyrus UNiQue genotype code) was assigned for each unique genotype.

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The 2-3 additional MPs genotyped for the USDA collection allowed an additional comparison of the European collections with this US collection and PUNQ assignment for this collection.

#### **Apple**

For apple, further SSR harmonization was achieved for over 2,266 accessions from Pometum collection (Univ. Copenhagen, Denmark), ISSA Orchard (Ireland) and various sites in Germany (including JKI-Dresden and Ökowerk collections). MUNQ codes were assigned accordingly.

The number of common SSR used in apple between ECPGR and USDA was low (only 5) such that any alignment between these sets should be treated with caution.

#### RECOMMENDATIONS

We would recommend further consideration of potential reference cultivars to be used in countries (preferably regions) where the current set of reference cultivars is not represented.

The alignment of collections through genetic analysis is fundamental to the development of the AEGIS concept. However, we would recommend that the next steps to realizing and identifying the Most Appropriate Accessions (MAAs) for inclusion within AEGIS would ideally require agreement from the expert groups on the issue of 'preferred naming' and synonymy as well as the clarification in many cases of the agreed 'country of historic origin'. Furthermore, it would be important for consideration within the 'duplicate' groups as to whether there might be valuable mutant or 'sported' selections as it is accepted that these would not be distinguished by the current technology. Lastly, for the further improvement of any decentralized collection, it would be important to better understand the phytosanitary status of accessions, specifically with respect to quarantine diseases and/or current regulations regarding the movement of germplasm.

We would recommend the continued use of the standard ECPGR SSR markers for both pear and apple in addition to the continued use of standard reference samples to aid with initial data alignment. Looking forward, work is underway to investigate SNP-based systems that can be aligned with the current dataset through MUNQ and PUNQ coding.

We have recommended that EURISCO continues to develop the capability to host harmonized SSR data and or at least provide the possibility to introduce the MUNQ and PUNQ data linked with unique accession identifiers.

#### **DATA AVAILABILITY**

MUNQ and PUNQ codes have been shared with partners and a combined MUNQ dataset (containing 4,319 accessions from 16 partners in 11 countries [including partners from outside ECPGR]) has been made available through the INRAE data repository <a href="https://doi.org/10.15454/HKGMAS">https://doi.org/10.15454/HKGMAS</a>. This dataset will be regularly updated.

Further individual SSR data will be made available after final agreement of the owners according to their wishes to first publish their own analysis.

#### COMPARISON OF REPORTED VS EXPECTED RESULTS

## Finalize and implement characterization and evaluation descriptors for apple and pear

Frost conditions during the 2017 flowering period made it impossible to collect phenotypic data from fruit in a number of collections. Consequently, the test of methods was delayed to 2018/19.

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The timing of finalization of draft descriptor documents was also delayed, in part due to a range of unexpected workload commitments such as all key partners being involved in the EU H2020 Valdifruit project submission although documents are now complete.

## Harmonize SSR data across genotyped collections of European pear and apple

Prior to the beginning of the project, it was expected that a formal agreement on data sharing would be required. However, from initial discussion with potential data suppliers it became apparent that those who were willing to share data were happy to do so on the basis of an informal agreement established by e-mail; those who wished to retain data until after publication were not persuaded by the offer of an agreement and so it was decided to progress on an informal basis. Nonetheless, a significantly larger number of datasets were amalgamated than were originally envisaged for both pear and apple.

The acquisition of new samples following the budget revision in March 2018 was more time-consuming than expected. The overall range of datasets and data providers was also significantly larger than expected and this made the analysis more complex and time-consuming. For example, a budget revision was agreed in March 2018 on the agreement that at least 288 additional accessions would be genotyped, where in total ~500 new accessions were eventually genotyped. The consequence of this additional work is that a much larger harmonized dataset has been produced than was originally envisaged.

Harmonized individual SSR data have not yet been included in EURISCO, although this was reliant upon developments within EURISCO. Members of the project have contributed to discussions within ECPGR around the potential to develop such capability in EURISCO and data could be made readily available for those partners agreeing to immediately share their individual SSR data. However, it has also become apparent that the scope of collections for which data have been aligned is now wider than that of EURISCO and so a more complete dataset will likely need to be managed elsewhere.

Progress needs to be made on a scientific paper resulting from the pear diversity analysis, although at the time of submission, this has not yet been completed. A poster was presented at the XV EUCARPIA Fruit Breeding & Genetics Symposium in Praha, Czech Republic, June 2019. Other publications and output about the project and more especially about the process of MUNQ and PUNQ code assignment are mentioned hereunder.

#### **OUTPUTS**

LATEUR, M., M. ORDIDGE, M. HÖFFER & C-E. DUREL [2017]. Common protocols and tools for characterisation and evaluation of *Malus/Pyrus* genetic resources – an ECPGR project. Lecture presented during the annual and 25th EUFRIN Board meeting – 15th of November, Brussels.

LATEUR, M. [2018]. Strategies for safe management and active uses of Fruit Genetic Resources: from case studies to a European vision. Invited lecturer at Wissenschaftliches Kolloquiumanlässlich der Verabschiedung vonFrau Dir. und Prof.Prof. Dr. Magda-Viola Hanke in den Ruhestand, Julius Kühn-Institut in Dresden-Pillnitz.

LATEUR, M., ORDIDGE M. [2019]. Integrating vegetatively propagated crops into AEGIS, using *Malus* as an example. In: Assessing current practices and procedures to strengthen AEGIS, the initiative for A European Genebank Integrated System, Report of a Workshop. Eds. J.M.M. Engels, L. Maggioni and E. Lipman, pp. 12-14. Bioversity International, Rome.

**LATEUR, M., RONDIA, A., ORDIDGE, M.** [2019]. 'Fruit tree genetic resources: from diversity safeguarding to diversity of uses - a complementary goal'. Invited speaker to the XV EUCARPIA Fruit Breeding & Genetics symposium, Praha.

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LATEUR, M., RONDIA, A., ORDIDGE M. [2019]. Fruit tree genetic resources: from diversity safeguarding to diversity of uses - a complementary goal. Invited lecture at XV EUCARPIA Fruit Breeding & Genetics Symposium, Praha, 3–7 June 2019.

DENANCÉ C., ORDIDGE M., LATEUR M., DUREL C-E. [2019]. Molecular characterization of a very large set of European pear accessions with SSR markers. Poster at XV EUCARPIA Fruit Breeding & Genetics Symposium, Praha,3–7 June 2019. (ANNEX 3)

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**DENANCÉ, C.**, H. MURANTY, AND **C. E. DUREL**. "MUNQ—Malus UNiQue genotype code for grouping apple accessions corresponding to a unique genotypic profile." Portail Data INRAE 1 (2020). WINFIELD, M., BURRIDGE, A., ORDIDGE, M. et al. [2020]. Development of a minimal KASP marker

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**LATEUR, M., GIOVANNINI, D., ORDIDGE, M.** [2021]. 'Fruit Tree Genetic Resources: from maintaining to sharing material through Europe – issues with quarantine and 'regulated non-quarantine pests'. Webinar jointly elaborated by members of the ECPGR Working Groups of Malus/Pyrus and Prunus presented during the GENRes Bridge Seminar 'Workshop on phytosanitary barriers for genetic resources', 24 February 2021, online MS Teams

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LATEUR M, SZALATNAY D, HÖFER M, BERGAMASCHI M, GUYADER A, HJALMARSSON I, MILITARU M, MIRANDA JIMENEZ C, OSTERC G, RONDIA A, SOTIROPOULOS T, ZELJKOVIĆ M K, ORDIDGE M. [2022]. ECPGR Characterization and Evaluation Descriptors for Pear Genetic Resources (*Pyrus communis*). European Cooperative Programme for Plant Genetic Resources, Rome,

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#### ANNEX I

Poster presented at XV EUCARPIA Fruit Breeding and Genetic Symposium, PraHA, 3-7 June 2019

## Molecular characterization of an extensive set of European pear accessions with SSR markers

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One of the major objectives of the ECPGR-funded project "PomeFruit\_C&E" [1] is to promote the molecular characterization of European pear collections and proceed to the standardization of the SSR data across collections. Such standardization is required to compare the pear accessions at the European scale and beyond.

#### **Material & Methods**

- ➤ 19 SSR (11 SSR of ECPGR set [2] + 8 additional SSR)
- ➤ 2,094 French accessions initially genotyped at INRA (blue dot)
- 499 accessions originating from 19 European countries newly genotyped (green dot)
- Development of dedicated allelic correspondence tables for 2,374 accessions originating from 7 European countries previously genotyped (red dot) [3 - 7]

MultiPlex	Marker	LG	Dye	Size Range	ECPGR set
	CH0197a	10	NED	170 - 251	X
GP_MP01	CHOING	17	NED	77 - 143	
	CH02b10	2	VIC	116 - 166	- 8
	CH05806	5	PET	160 - 195	
	EMPc11	11	FAM	117 - 179	×
	CH01/03b	9	FAM	139 - 274	
	CH01h10	5	VIC	84 - 160	
GP_MP02	CH02c11	10	NED	200 - 260	
	CH05c06	16	NED	86 - 137	2.
	GD147	13	PET	118 - 176	×
GP_MP03	CH-Vft	1	VIC	133 - 179	
	C1901d08	15	VIC	195 - 311	x
	CH03d12	- 6	NED	86 - 167	x
	EMPc117	7	PET	72 - 152	x
	GD142	9	FAM	122 - 213	
GP_MP04	CH01409	12	NED	121 - 179	x
	CH03g07	3	VIC	201 - 294	X
	CH04c07	14	VIC	84 - 162	
	C3104e03	5	FAM	18D - 242	×

Table 1. List of the 19 SSR markers used for fingerprinting with linkage group location, size range and multiplex assignment. PCR conditions are available upon request

#### **Results & Discussion**

- ➤ 4,912 accessions with at least 9 standardized SSR
- ➤ 2,206 unique genotypes (i.e., distinct SSR profiles)
- PUNQ code (Pyrus UNiQue genotype code) assigned for each genotype in order to better characterize any new accession according to this standardized dataset.



Country	Institut Acronym	Nb acc.	Country	Institut Acronym	Nb acc.
AL	ABG	10	GR.	NAGREF	10
	UNSA	34	HB.	HCPHS	10
BA	UNIBL-GRI	17	HU	NARIC	8
BE.	CRA-W	152	IE	155A.	25
BY	BELSAD	9	п	UNIBO	56
CH	Fructus	179	т.	CREA-OFA	24
CZ	RSIPH	20	LT	LAMMC	20
DE	JKI	122	LV	LLU	60
DK	UCPH	28	NO	NNU	8
***	UPNA.	156	PL.	InHort	10
ES	CITA	8	RO	TCDP	32
FI	LUKE	29	RU	NCRRIHV	48
TD	INRA	1059	800	VNIISPK	32
FR.	UPF	1035	SE	UNI-LJ	9
GB	UoR	549	SK	VURV	6
	FruitID	743	SW	SLU	136
	HHLBT	312	UA	IH NAAS	11

<u>Table 2.</u> List of the collections with numbers of accessions genotyped

#### Next steps

 Analysis of genetic structure and relationship inferences

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XV EUCARPIA Fruit Breeding and Genetics Symposium, Prague, 3-7 June 2019









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## **ANNEX II**

ECPGR Characterization & Evaluation Descriptors for Apple & Pear Genetic Resources, published in 2022.



#### The Descriptors are available at:

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Pear - <a href="https://www.ecpgr.cgiar.org/resources/ecpgr-publications/publication/ecpgr-characterization-and-evaluation-descriptors-for-pear-genetic-resources-2022">https://www.ecpgr.cgiar.org/resources/ecpgr-publications/publication/ecpgr-characterization-and-evaluation-descriptors-for-pear-genetic-resources-2022</a>