



Fruit Breeding in the Context of Climate Change Adaptation^(§)

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ABSTRACT

Spain is the world's leading exporter of fresh stone fruits belonging to the genus *Prunus*. This position has been favored by Spain's climate, which enables earlier production than in the rest of Europe, as well as and commercial use of high-quality, certified, virus-resistant new cultivars adapted to climate change the almond, apricot and plum by the development national in the different national breeding programs and particularly conducted at CEBAS-CSIC in Murcia in the case of almond, apricot and plum. In the context of the new climate, this communication aims to address the latest approaches being carried out by the CEBAS-CSIC of Murcia in the development of new *Prunus* varieties in the postgenomic scenario (once the complete sequence of the genomes of the main species is known). Furthermore, massive genome (Genotype by Sequencing, GBS) and transcriptome (RNA-Seq) sequencing is helping to better understand the biological and molecular processes associated with the expression of these traits of agronomic interest. Once these genes are identified, we can use this information to select seedlings that have these characteristics using classical Polymerase Chain Reaction (PCR) or quantitative Retrotranscribed PCR (qRT-PCR) with a potential application in marker-assisted selection. More recently, a new line of research has been initiated aimed at identifying epigenetic marks associated with characters, especially adaptation to the environment, such as flowering time. Epigenetics studies change in phenotype that are inherited, but are not due to alterations in the DNA sequence. This DNA methylation mechanism has been described as a regulator of dormancy breaking in fruit trees or fruit ripening.

Keywords: Stone fruits, *prunus*, breeding, genetics, genomics, transcriptomics, epigenetics

Introduction

Prunus production around the world was around 50 million tons being the peach the most important species, with an annual production of more than 27 Mt in 2023 (<http://faostat.fao.org>, updated June 2 2026). In Spain, *Prunus* production was of 2 Mt ranking this country in the second position behind China, and closely followed by Türkiye and Italy (Figure 1). In addition, Spain is the first exporter of peaches in the world, with more than 50% of the exported production (0.8 Mt) and an added value of this exportation of more than 900 million euros (M€) (www.fepex.es, Asociaciones de Productores Exportadores de Frutas, Hortalizas, Flores y Plantas vivas, FEPEX, updated June 2 2026). This leadership in the production and export of fruits has been marked by the climate compared to the rest of Europe, which allows early production, in addition to the development

and exploitation of new quality, certified, virus-resistant and adapted varieties to the climate change obtained in the different national breeding programs.

Stone fruit (*Prunus*) breeding is an applied science of design where constantly new designs are performed to solve specific problems, such as productivity, resistance to biotic and abiotic stresses, fruit quality, postharvest performance and sensorial attributes (Martínez-Gómez et al., 2003). At this moment, the treating of breeding as science of design supposes to consider this activity closer to software development or the pharmaceutical industry than biology. The requirement of these new designs demands each time more complicated mechanisms to choose parents for crossing and select the new descendants (Martínez-Gómez, 2017). In addition, genetic transformation to develop Genetically Modified Organisms (GMOs),

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including CRISPR (clustered regularly interspaced short palindromic repeats) application, is at this moment a far option from a consumer point of view (Martínez-Gómez, 2021; Martín-Valsaneda et al., 2023).

In this review, we have analyzed the new perspectives on fruit breeding in the context of climate change adaptation to ensure production and quality approaches being carried out by the CEBAS-CSIC of Murcia in Spain.

Development of new *Prunus* varieties

Plant breeding is thus a scientific discipline that contributes to the knowledge necessary to use a certain technology, which in turn applies that knowledge to produce new plant varieties and plant patents. Classical fruit breeding is a long and tedious process involving the generation of large populations through controlled crosses of previously selected progenitors and the final selection of top individuals, the future new varieties. This process can take between 5 years in the case of horticultural crops to 15 years in the case of perennial fruit crops (Figure 2) such as *Prunus* or 25 years in forest species (Martínez-Gómez et al., 2003).

Plant breeding differs from basic sciences in that it seeks not only to understand biological systems but also to modify them to achieve defined objectives. It therefore combines elements of natural science and design science, drawing on observation and experience to develop cultivars that as practical solve problems. When successful, this process transforms scientific knowledge into technological innovation through the breeding of new plant cultivars (Martínez-Gómez, 2020).

In addition, to develop suitable new cultivars, we need to know the relevant variables that influence our knowledge of the possible future in the success of new cultivars. It is thus necessary to examine the internal and external variables influencing the phenomenon in question. Both types of variables must be subjected to scientific evaluation. The first internal variables to consider in any almond breeding program are derived from plant traits that are considered as objectives. The knowledge of these internal variables of genetic type will indicate their suitability with respect to the proposed design objectives. Secondly, internal variables also include the current methodologies available for use in the selection of individuals. These methodologies are closely related to the level of knowledge available at the molecular levels especially in relation to the development and application of new markers for the selection of individuals and will give us an idea of the efficacy and feasibility of new designs or cultivars. Finally, we can include various economic factors within the production framework, i.e., the goals, processes and outcomes. The effectiveness and

viability of the improvement program will depend on these methodological developments dedicated to the evaluation. These two terms (effectiveness and viability) are related in the economic context to the relationship between science and economics. To develop successful cultivars, breeders must identify the internal and external variables that influence the likelihood of achieving the breeding objectives. Internal variables include the genetic traits targeted by the programme and the methods available for selecting parents and progeny (Martínez-Gómez, 2017; 2020).

On the other hand, in plant breeding, it is necessary to consider a number of external variables including biotic and abiotic stress and interactions with the environment over a period of several years. There are many characteristics such as flowering time and floral compatibility which must be evaluated for at least three years, when trees produce the first flowers. Due to drawbacks, the use of molecular marker-assisted selection methods is of particular interest in breeding programs. These drawbacks make the use of methods of assisted selection by molecular markers already discussed. These environmental conditions will determine the feasibility of the objectives and the new variety in a given environment. The adaptation of new designs or new cultivars to specific climatic conditions is fundamentally conditioned by the winter chilling requirements and dormancy and by their adaptation to different soil conditions in the case of rootstocks. Its capacity for production depends largely on this character and on its interaction with the environment considered as an external variable. In addition, the knowledge of these external variables related to possible biotic stresses (pest and disease) will indicate their feasibility with respect to the proposed design objectives (Martínez-Gómez, 2017). The feasibility of developments is another important issue to consider. The technological feasibility will also depend on the existence of germplasm with the desired genes for resistance to different biotic or abiotic stresses (Figure 3).

Finally satisfying consumer demands is also an important external variable. New designs must have the appropriate characteristics to meet such demands. In addition, the availability of different techniques for analysis and selection is another important external variable. In addition, the social acceptance of these new cultivars will depend on criteria of environmental sustainability (Infante et al., 2008). On the other hand, we can include various factors of an economic nature, provided that they are considered within the framework of production: the phase of objectives, processes and results. This affects external economic variables, which are those that affect their market value. Because the

result of a new genetic variety presents durability (a life cycle) and involves a market price, which conditions the creative transformation carried out by technological innovation (García-Gómez et al., 2021).

The degree of knowledge of the variables determines the quality of the prediction in the design and development of new cultivars of *Prunus* in addition to its effectiveness, feasibility and suitability. New designs must meet the appropriate characteristics to meet those requirements. External variables are also the availability of the different analysis and selection contemporary techniques, since they have varied throughout history.

Application of molecular markers in the development and management of new *Prunus* varieties

The management, phenotyping and selection process of progenitors and seedlings are the main factors limiting the generation of new cultivars. In order to improve efficiency and viability of breeding programs in relation to parent and seedling selection, the implementation of molecular tools is an essential requirement, including development of new Marker Assisted Selection (MAS) strategies based on the molecular knowledge of the determination of the phenotypic traits. Molecular effort should be oriented in the analysis of the molecular basis of the target traits and the implementation of molecular tools to be incorporated together with the classical activity of the visual selection of good descendants (Martínez-Gómez et al., 2003). In this context, at this moment in the present Postgenomic era we are facing a new molecular-biological perspective based on new methodologies that are affecting the molecular analysis. This new molecular perspective opens new possibilities to improve the use of molecular tools in new more efficient and viable plant breeding (Martínez-Gómez, 2019).

At this moment, more than 650 plant species have been sequenced and their reference genomes are available including the most important crops in the world (<https://www.ncbi.nlm.nih.gov/genome>, accessed 1st July 2025). This whole sequenced work started in 2002 with the development of the reference genome of the rice. In recent years, for instance several important crops were sequenced including hazelnut, mulberry, pistachio, poplar or almond in 2019. The development of complete genomes is making any organism accessible and amenable for many kinds of studies which will allow a precise reference of the molecular results obtained, and the development of high-throughput methods for genomic analysis involving the most abundant genetic variation (Single

Nucleotide Polymorphisms, SNPs) and transcriptomic analysis at Differential Gene Expression (DEG) level in a new postgenomic perspective. At this moment, the new Big Data Biology harboring the development of DNA (and RNA as cDNA) high-throughput sequencing technologies together with bioinformatics analysis as well as the creation of databases with billions of data has made possible to access genetics knowledge at the level of each nucleotide. This new methodological perspective where millions of sequences are available in one single experiment with the detailed information of complete genomes (defined as the DNA organized into separate chromosomes inside the nucleus of a cell) and transcriptomes (described as the complete list of all types of RNA molecules). Several authors have even characterized this data-intensive biology as a new kind of science, a science of information management, different from traditional biology. Big data science is now being introduced in the development of molecular markers to assist breeding selection methodologies (Martínez-Gómez, 2019).

On the other hand, these molecular tools are also particularly useful when the evaluation of the complex traits such as biotic and abiotic stress and fruit quality including nutraceutical values. These complex traits are being recently introduced in the stone fruit breeding programs. Nutraceutical properties will add an important value to the new releases for industrial purposes. In these species nutraceutical value studies are focused on fiber contents; phenolic contents; carotenoid (correlated with vitamin A) and vitamin C and E contents; and mineral contents including magnesium, potassium, selenium and zinc (García-Gómez et al., 2021).

This new postgenomic perspective integrating available reference genomes and new sequencing and bioinformatic methodologies will allow the implementation of new Marker Assisted Selection (MAS) to accelerate breeding process. The application of these molecular tools will increase the viability and efficiency in the development of the new planned design. In this context, high-throughput sequencing technologies resulted in a great advance in the development and application of MAS strategies. Additionally, from a global point of view, the new post-genomics era is characterized by a change in perspective on trait expression derived from the Encyclopedia of DNA Elements (ENCODE) project, performed on humans, in which the study of RNA rather than DNA was determined as the linchpin of trait expression processes (Martínez-Gómez 2019). In fact, the genomes of eukaryotic organisms are almost entirely transcribed, giving birth to an enormous number of non-protein-

coding RNAs. In this new context, a recent concept, pervasive (interleaved) transcription, is being applied to the whole transcription process. These authors described pervasive transcription as the transcription of the interspersed genes that are embedded within the normal coding genes. The entire stretch of the genome (in other times called “junk DNA”) is transcribed, whether it is coding for a particular protein or not. Therefore, not all coding sequences lie juxtaposed, and they may also overlap one another (Martínez-Gómez et al., 2012).

New integrated classical and molecular perspective in *Prunus* breeding

Plant productivity for feeding an increasing human population and the study of plant pests and diseases and their management in an unpredictable context of global climatic changes related to different abiotic stress are particularly relevant aspects as well as genetic plant breeding from the point of view of the quality of fruits and vegetables. Therefore, classical breeding must be combined with MAS using the diverse molecular tools available in the postgenomic era in each time more complicated scenario.

The generation of large genetic and phenotypic information obtained in a massive way (through high-throughput sequencing and high throughput phenotyping approaches) and the creation of databases with billions of these data, result in what is called in research “Big Data”, which also require the development of new mathematical algorithms (such as machine learning models) and a large set of bioinformatic tools. In addition, from a global perspective, the center of gravity of the molecular processes is focused on the expression of genes and the way in which such expression is regulated. These features have turned researcher’s attention towards the study of gene regulatory networks, and in particular to the role of RNA and other non-derived DNA biomolecules (Martínez-Gómez, 2022). These new approaches will be of great help in the new challenges of the plant breeding.

This situation does not mean that MAS will replace conventional breeding; other ways is a necessary complement. The practical learning that conventional breeding gives is unparalleled with the molecular support. In this context, there has been a significant shift of conventional breeders to molecular breeding aspects. However, in this enjoyed strategy (conventional and molecular) the target must be well defined in a complete integrated work plan. While the ability of breeders to generate large new breeding populations is almost unlimited, the evaluation and selection of these promising seedlings is the main

limiting factors due to the cost and time-consuming. In this context, genomic studies at the DNA level are especially useful for the development of MAS strategies. In addition, proteomic (proteins and enzymes), transcriptomic (RNA) and epigenetic (DNA Methylation and histone modifications) studies are being applied to breeding programs. Finally, these strategies at genomic, epigenetic, transcriptomic and proteomic level should be all integrated for a better understanding of the molecular mechanisms involved in the most important plant breeding aspects, which will facilitate the development and optimization of molecular markers to apply in the field exploitation of the new varieties, offering and integrating complete technological offers increasing feasibility, effectiveness and success of the breeding programs (Figure 4).

In this new context DNA and RNA markers and epigenetic marks could be applied in the development (breeding) and exploitation (cultivation) of the newly developed *Prunus* varieties in Marker Assisted Selection (MAS), biological validation of DNA markers and monitoring flowering and ripening process (Figure 4).

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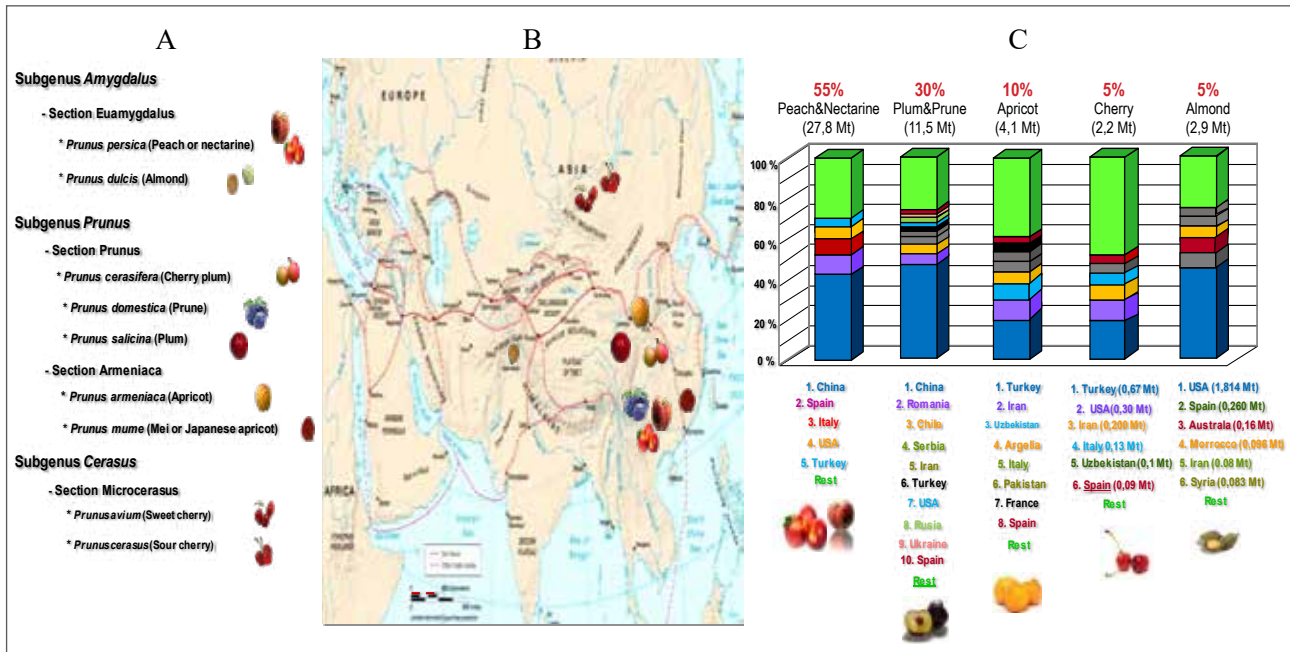


Figure 1. Botany of main cultivated *Prunus* species (A). Origin and dispersion of *Prunus* species through the silk road (B). Main *Prunus* production countries in the world (C).

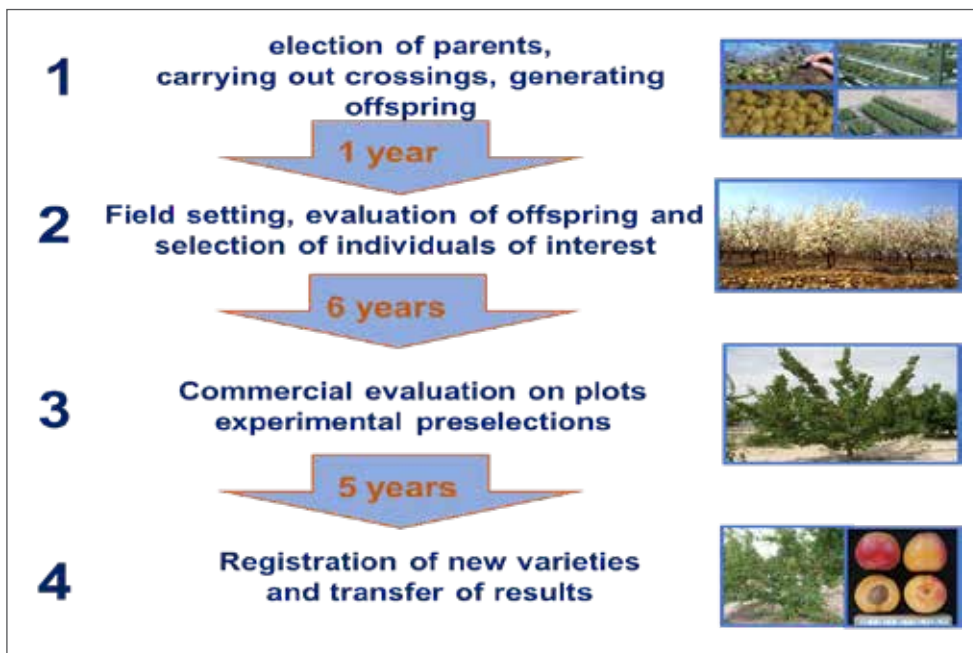


Figure 2. Representation of the most important steps in a *Prunus* breeding program.

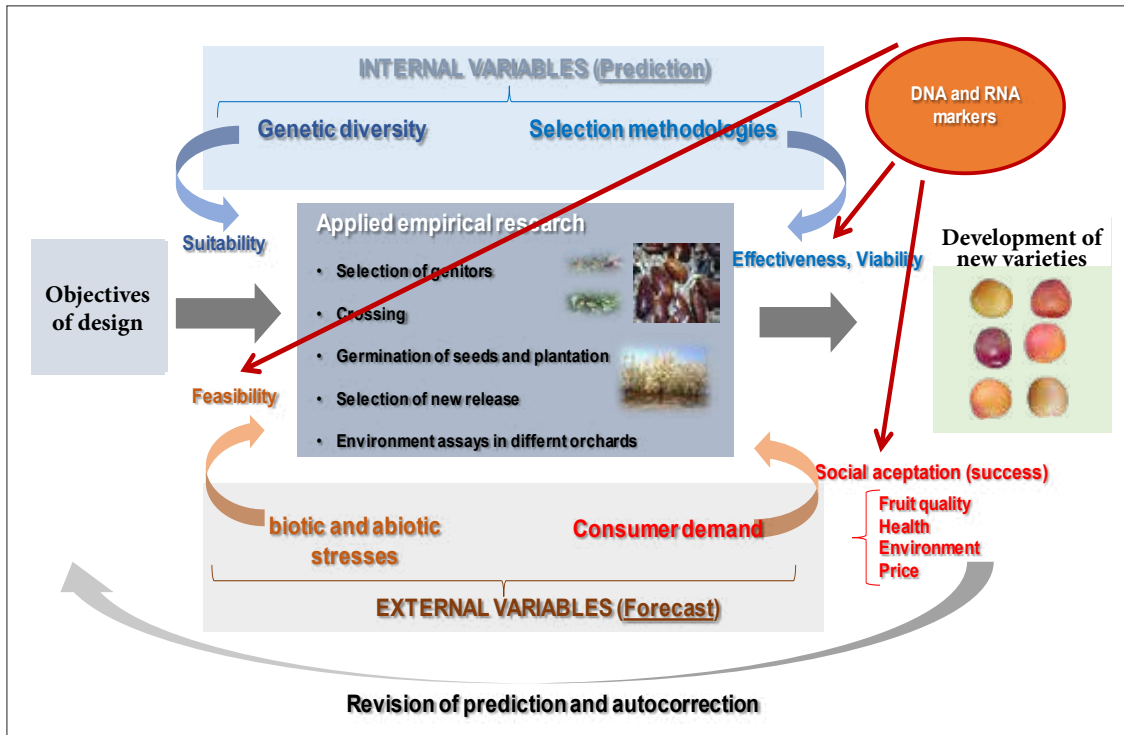


Figure 3. Schematic representation of *Prunus* breeding process as a science of design.

	DNA markers	RNA markers	Epigenetics marks
Breeding (development)	➤ MAS ➤ Self-compatibility ➤ Sharka resistance ➤ Bitterness ➤ Fruit colour	➤ Biological validation ➤ Flowering ➤ Fruit quality ➤ PPV resistance	➤ MAS ➤ Flowering time ➤ Fruit quality
Production exploitation	➤ Identification ➤ Varietal protection ➤ Traciability and quality	➤ Monitoring ➤ Flowering ➤ Ripening	

Figure 4. Application of molecular markers in the development and exploitation of new *Prunus* varieties.

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