



PROTOCOL FOR THE PRODUCTION OF GENETIC QUALITY SPECIMENS OF PHOENIX SPECIES



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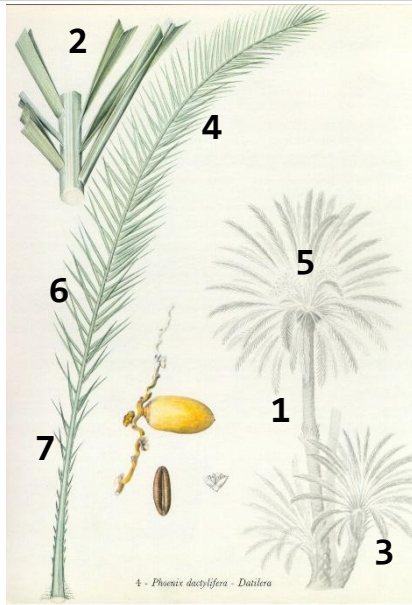
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This protocol has been elaborated in the framework of the European project LIFE-PHOENIX [<https://lifephoenix.gesplan.es/es>] called Restoration and improvement of the priority habitat 9370* "Palm groves of Phoenix".

September 2024



Identification of the specimen by morphological characters

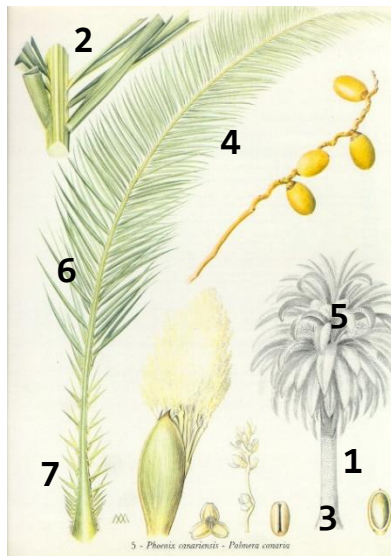


Phoenix dactylifera

The existence of hybrid specimens between *Phoenix canariensis* and *P. dactylifera* in the natural populations of the Canary Islands makes it necessary to reliably identify the species to which the individual from which the seeds are to be obtained belongs.

Morphologically, it can be differentiated according to different characteristics such as the shape of the leaf crown, the size of the leaves, the pinnae and thorns, the presence of suckers at the base of the trunk or the colour of the leaves.

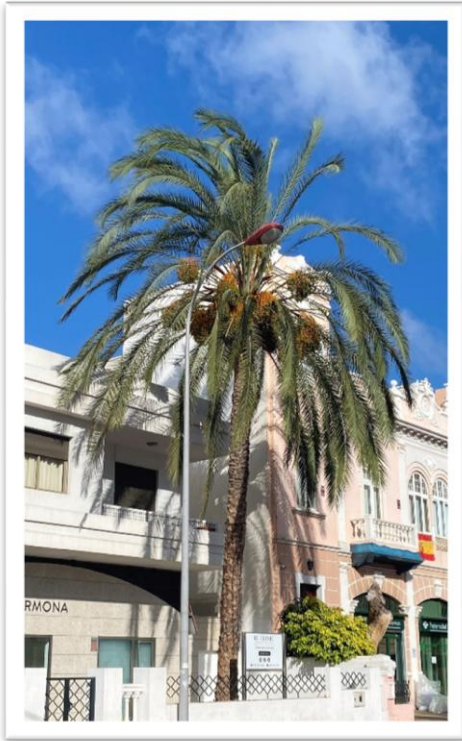
The safest and most reliable way is through molecular markers.



Phoenix canariensis

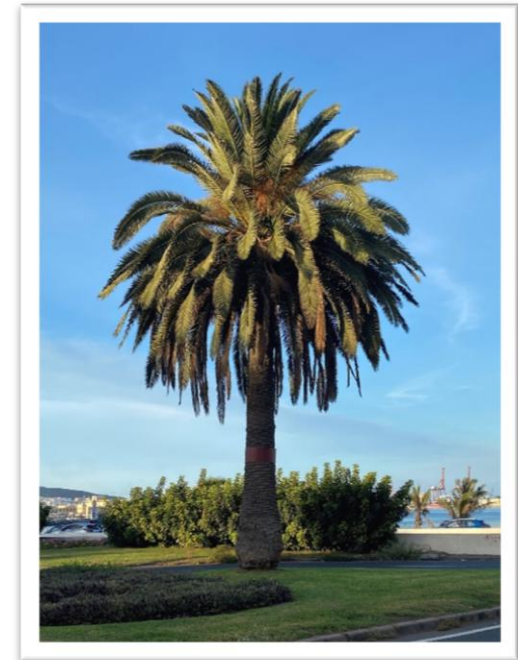
Morphological identification of *Phoenix canariensis*

Phoenix dactylifera



Attributes	<i>Phoenix canariensis</i>	<i>P. dactylifera</i>
Stem	Completely straight. Columnar appearance	Usually, crooked. Not excessively thick
Presence of offshoots	Absence	Occasional presence
Size of leaves	Long, broad leaves with elongated, dense leaflets	Slightly shorter leaves, leaflets not very dense
Colour of the leaves	Dark green	Glaucus
Spines	Long and thick, sometimes yellowish	Short and thin
Shape of the crown	Spherical	Open leaf crown

Phoenix canariensis



Choice of specimens and seeds



Choose females at least 70 m apart from each other to minimise kinship between individuals.

It has been shown that individuals from the same population differ genetically with distance. The greater the distance, the more likely they are to be genetically different.

Choice of specimens and seeds



Collect seeds from different clusters, to maximise the genetic diversity captured.

Although it has been shown that the seeds in a single cluster have been generated from fertilisation by more than one male, genetic variability is optimised by collecting fruit from different clusters of the same female.

Fruit harvesting season



The fruit must always be harvested from the palm tree using disinfected tools.

Although there are palm fruits all year round, due to the overlapping of generations, the months of June to October are usually more favourable for harvesting them.

An analysis of the phenological evolution of the specimens is recommended in order to optimise the collection.

Fruit stage for optimal harvesting



The fruit must be harvested when they are close to maturity. The chambers should be orange in colour.



Always pick the fruits of the palm tree, NEVER pick them from seeds on the ground, as their origin is unknown and they are usually affected and attacked by insect larvae and could be unviable.

Fruit harvesting stage



Avoid picking very green (unripe) or very ripe (dark brown) fruit



Assignment of registration (accession)



*A traceability procedure must be maintained.
Seeds of specimens should initially be stored
in individualised bags or containers
appropriately marked until the time of
cleaning.*

*This should be assigned a registration number
called Accession.*

Removal of spoiled fruit



The harvested fruits are dispensed in aluminium trays with their corresponding accession and the seeds damaged or affected by weevils or insects are removed.

Fruit processing



The fruit must be hydrated. Soak the fruit in water at room temperature for about 48 hours. This will allow and help to remove the outer layers (epicarp and mesocarp) of the fruit.

If collection missions are prolonged in time, it is necessary to pre-wash the fruits in the field and to extract and dry the seeds to reduce the volume and weight during transport, and to remove contaminants and bring the moisture content to a safe level.

Germplasm processing



*All traces of pulp must be removed from the fruit.
Shelling*

*It can be done mechanically or manually
depending on the number of seeds.
It is advisable to do this manually with the help of
tweezers, as, although it is more laborious, it
ensures that no seeds are lost.*



*Clean seeds without epicarp and mesocarp
improve sample purity, facilitate the removal of
damaged and immature seeds, optimise storage
space and reduce costs.*

Seed disinfection



It is essential to disinfect the seeds of all types of bacteria, fungi and other possible pathogens.

Soak the cleaned seeds for two minutes in hydrogen peroxide and stir gently in an electric shaker.



Seed drying



Leave the seeds in a container open to the air to remove excess moisture.

Leave it in the air at room temperature for 24-48 h.



Long-term *ex situ* conservation



Leave for about 4 months in a chamber with controlled ventilation and humidity and in a desiccator.

Finally, place in airtight containers at 4°C with silica gel separated from the seeds by cotton wool.



Co-financed by the European Union. However, the views and opinions expressed are solely those of the author(s) and do not necessarily reflect those of the European Union or CINEA. Neither the European Union nor CINEA can be held responsible for them.

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