Conservation of Date Palm (*Phoenix dactylifera* L.) Genetic Resources

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- Date palm (*Phoenix dactylifera* L.) is an old tree with many distinguished socioeconomic and nutritional values.

- It is considered as one of most ancient fruit trees in the world. It is expected to be originated s in the desert oases of northern Africa and southwest Asia, and it's expected that date palm was domesticated 6,000 years ago in the Mesopotamian Region

- It is a monocotyledon tree and belongs to family Arecaceae, where Phoenix genus consists of 14 species

TABLE 1 | Species of the genus *Phoenix* along with their common local name and geographical distribution. Al-Alawi et al., 2017

Species	Local name	Geographical distribution
Phoenix acaulis	Stemless date palm	Bhutan, Nepal, Northern India
Phoenix andamanensis	Andaman Island date palm	Myanmar
Phoenix atlantica	Cape Verde Island	Cape Verde Islands
Phoenix caespitosa	Date palm	Djibouti, Oman, Saudi Arabia, Somalia, Yemen
Phoenix canariensis	Canary Island date palm	Australia, Bermuda, Canary Islands, Italy, Spain
Phoenix dactylifera L.	Date palm	Arabian Peninsula, Australia, California, China, El Salvador, Fiji, Iran, India, Mauritius, northern and western Africa, Pakistan, Spain
Phoenix loureiroi	Mountain date palm	China, Himalayas, India, Indochina, Philippines
Phoenix paludosa	Mangrove date palm	Andaman, India, Indochina, Sumatra
Phoenix pusilla	Ceylon date palm	India, Sri Lanka
Phoenix reclinata	Senegal date palm	Africa, Arabian Peninsula, Comoros, Madagascar
Phoenix roebelenii	Pygmy date palm	China (Yunnan) to North Indo-China
Phoenix rupicola	Cliff date palm	Andaman Islands, Bhutan, India
Phoenix sylvestris	Indian date palm	Indian Subcontinent, Myanma, southern China
Phoenix theophrasti	Cretan date palm	Greek Islands, Turkey

Moreover, Dates are classified according to their moisture and sugar content in addition to texture into three classes including soft, semidry, and dry cultivars.

Khadrawy (soft)

) Dry dates of (1) 'Pertamoda', (2) 'Malaka

(Abul-Soad et al., 2017).

Medjool (semidry)

Ascel dates (semidry)

- Date palm tree is distinguished by its minimal requirements, although it remains productive for long periods of time.
- All plant parts are included in industrial applications, this in addition to the date fruits which are highly nutritious.

- Moreover, date palms have environmental advantages as they can withstand many adverse environmental conditions, which permit them to be a magical solution for desertification problem .

Table 1: Top ten countries in terms of date production (FAOSTAT 2016)

Country	Production (1000 metric tons)
Egypt	1,694
Iran	1,066
Algeria	1,030
Saudi Arabia	964
United Arab Emirates	672
Iraq	615
Pakistan	495
Sudan	439
Oman	348
Tunisia	241
World total	8,460

Threats facing date palm genetic resources

- Continuous selections by farmers which consequently narrowed down their genetic diversity
- The negative impacts of climate changes, especially the fluctuations in temperatures and water scarcity in addition to high soil salinity.
- The fierce attack of insects and pathogens

- So, genetic diversity of date palm needs to be conserved to maintain cultivars with good quality, and most important to keep the genetic resources safe guard as they are the main requirements for any possible genetic improvement in future.

Conservation of date palm

1. *In situ* conservation

In situ conservation refers to a collection of actions where genetic diversity of selected populations are protected, managed and monitored in their natural habitats

Advantages

- The evolution of the conserved plant species is allowed which would permits creation of new diversity through natural selection

- This criteria has made *in situ* conservation method highly required for conservation of genetic resources as it gives the chance to plants to adapt changes in environment including global warming, fluctuations in rainfall as well as loss of habitat
- Currently, *in situ* conservation is the most common approach used for conservation of date palm genetic diversity as whole plants.

Disadvantages

- Genetic material of date palm conserved *in situ* is exposed to natural disasters such as, high temperatures, floods or water scarcity and attack of pests and pathogens.
- High requirements of cost and labor
- Above all these limitations, the loss ability of date palm trees to give off shoots after 15-20 years of culturing which would result in genetic erosion.

2 *Ex situ* conservation

Plant genetic resources are maintained in gene banks or botanical gardens where they are allowed to grow safely under suitable conditions

Advantages

- Ease of identification of the conserved genetic diversity due to systemic documentation,
- High security level as plants are strictly monitored and so loss of plant material rarely happens .

Disadvantages

- Huge number of samples need to be collected to conserve genetic diversity, which recommends large land space and trained labor requirements which implies very high cost.

- Plant material under ex situ conservation is largely exposed to diseases and environmental disasters, wars and changes in policies which makes maintenance of genetic material very difficult and costly.

3. *In vitro* conservation

- *In vitro* conservation is a technique where plant material is conserved under aseptic and pathogen free environment.

Advantages

- Plants conserved *in vitro* are allowed to grow under minimal space, labor and cost requirements.
- *In vitro* conservation might be the only hope to conserve the germplasm of some plant species when seed storage is not feasible.

- Plant germplasm can be conserved in *vitro* for short to mid- term periods of time (slow growth conservation) by either modifying the culture media (adding osmotic agents) or incubation under low temperature.
- Germplasm can be stored for long term durations using cryopreservation technique, where plant material is stored under ultra- low temperature (196 ˚C).

- As for date palm germplasm, *in vitro* conservation has been an excellent choice for conservation due to the dioecious and heterozygous nature of this tree that mostly propagated through offshoots which made it difficult to maintain its genetic fidelity or stored and handled conventionally

Techniques of *in vitro* conservation

3.1 *Slow growth conservation:*

Slow growth technique involves altering growth speed of the conserved plant material by either:

- Changing the constituents of maintenance culture media **(chemical approach)** :

growth speed is slowed down by adding osmotic agents such as, sugars and salts to the growth media, or by reducing optimal carbohydrates level or by adding abscisic acid or combinations of these techniques

Slow growth conservation

- Keeping cultures under sub-optimal growth room conditions **(physical approach)**: growing the conserved plant material under either low temperatures or light intensities or by changing state of gaseous environment.

- The brightest achievements of slow growth conservation are that:
  - conserved plant germplasm is maintained disease-free throughout storage period.
  - able to retain its normal plant multiplication rate after storage.
  - readily available to users all year
  - Its genetically stable.

Major constraints

- losses due to contamination
- High labor requirements
- Destruction of the conserved genetic material due disasters (fire, earth quakes… etc.)

3.2. Cryopreservation

- Cryopreservation involves storage of genetic material for long periods of time in liquid nitrogen where storage temperature of -196°C

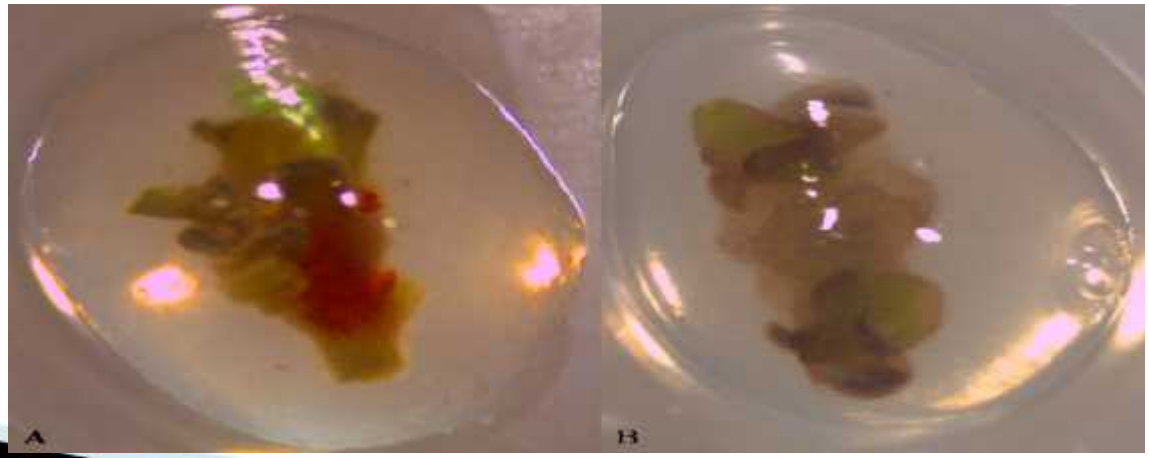
Cryopreservation

- Cell division and metabolic activities of plant cell stop which allows storage of plant genetic material for long periods without threats of deterioration or loss of genetic stability.

Cryopreservation techniques

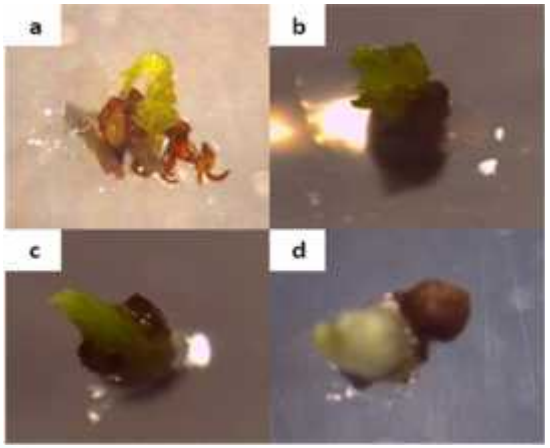
- **Encapsulation-dehydration:**

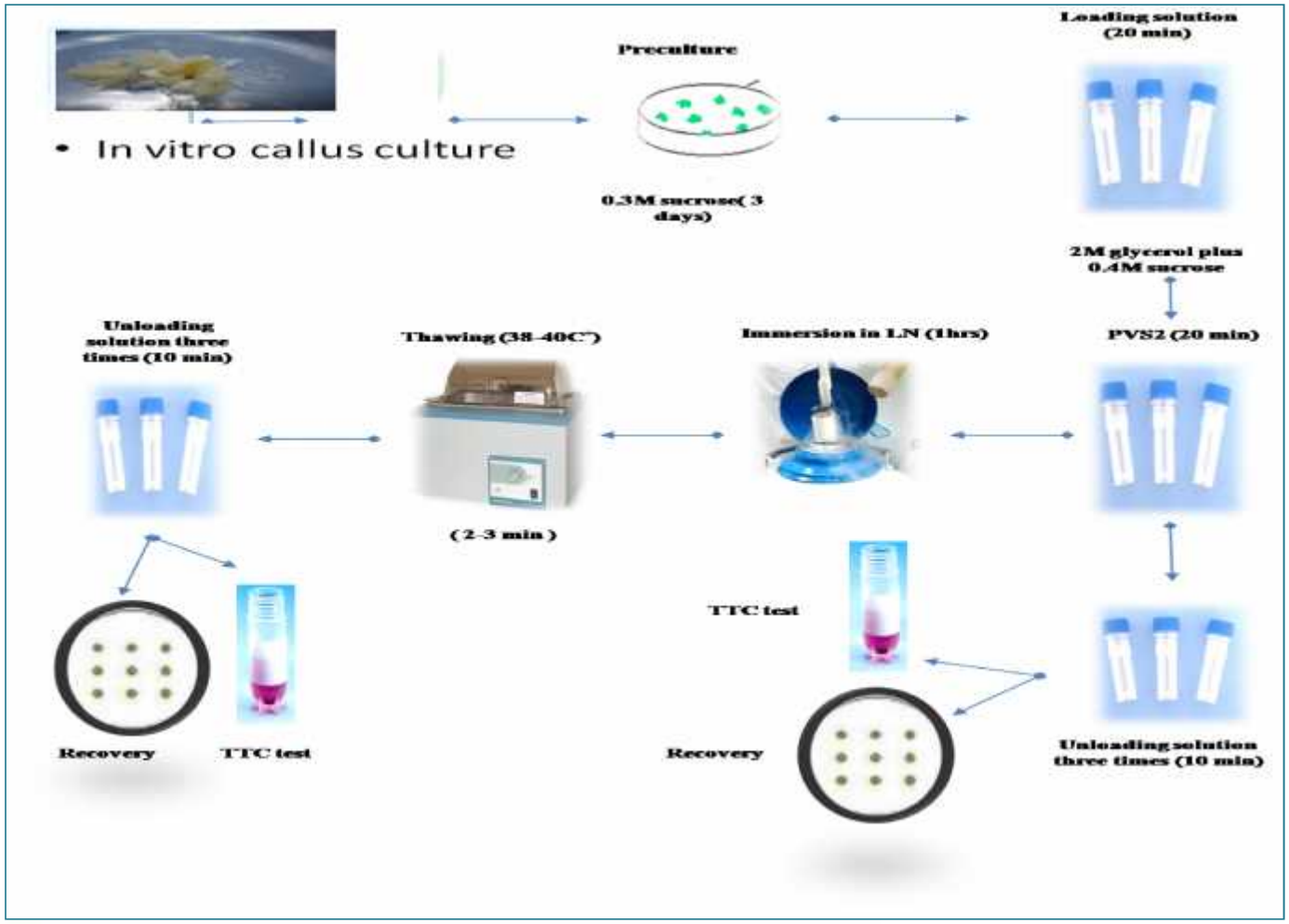
Explants are coated within alginate beads before being cultured in a medium with high levels of sucrose of sucrose, then allowed to dehydrate to minimize moisture content before dipping in liquid nitrogen .



- **Vitrification technique:**

The plant genetic material is loaded with high levels of a cryoprotectant to decrease moisture content and to maximize cell osmolality which would help the cells to withstand exposure to liquid nitrogen.

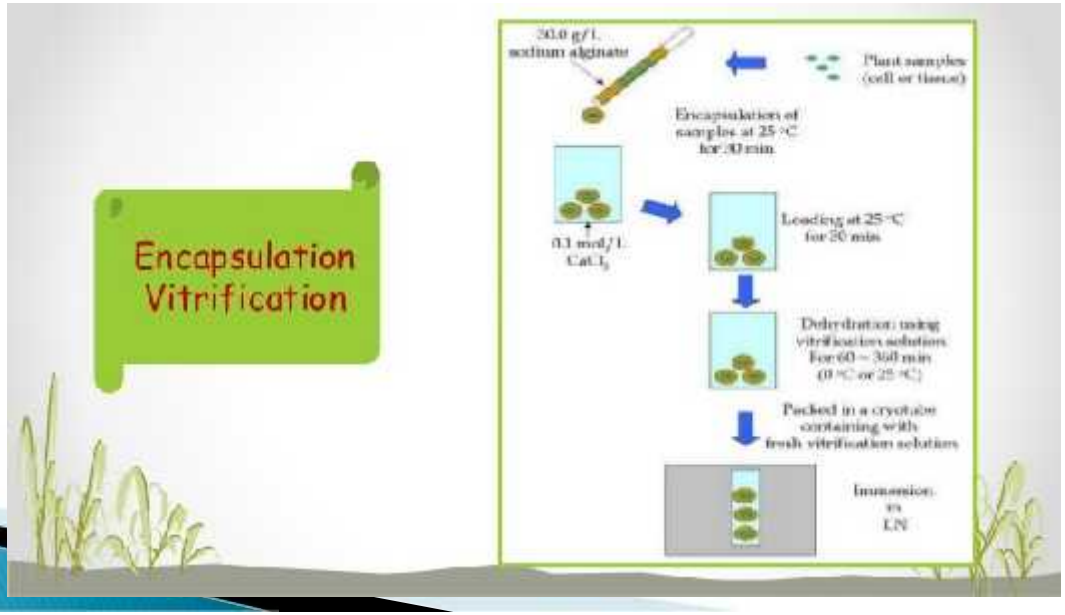




Vitrification protocol

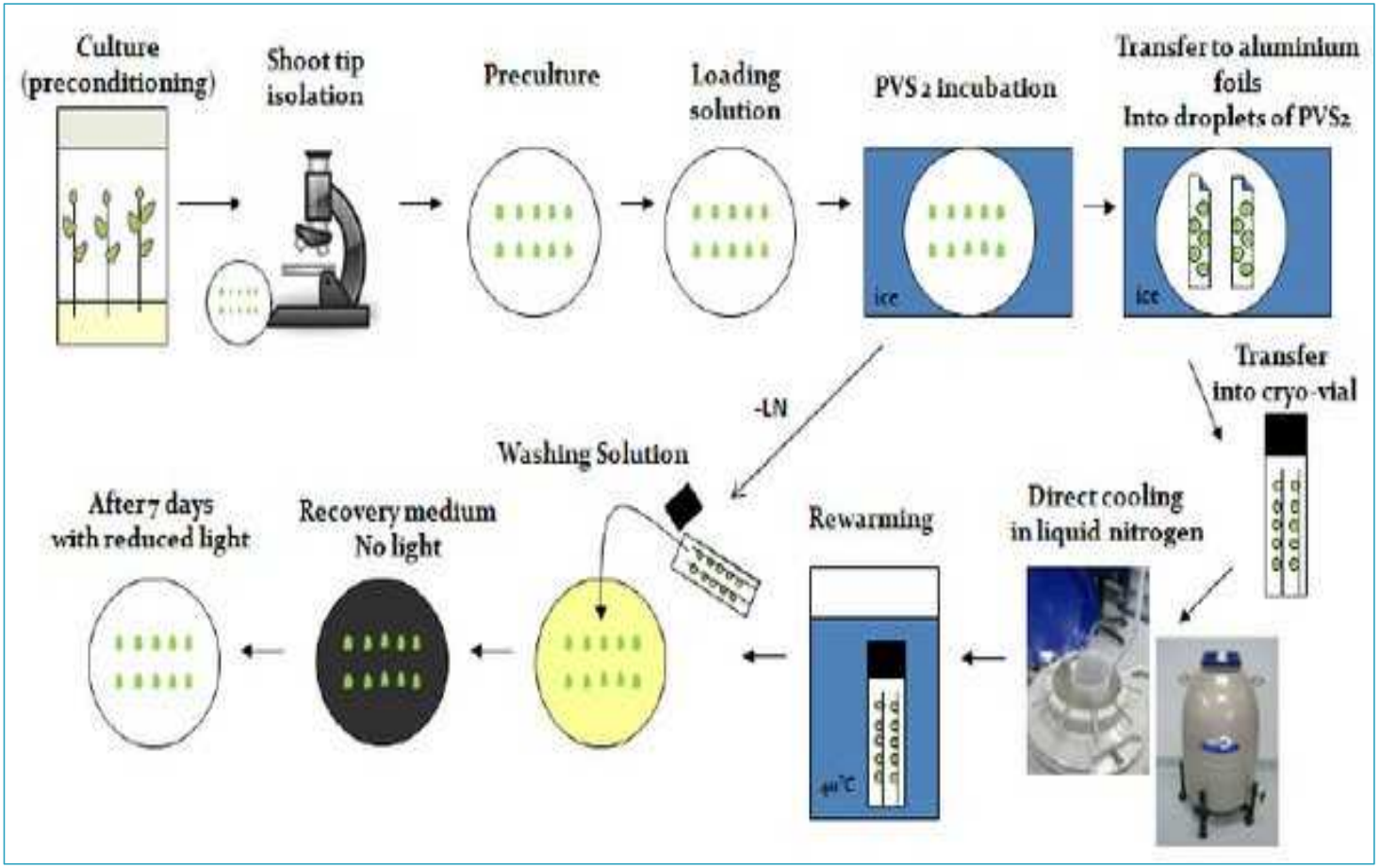
- **Encapsulation-vitrification :**

Encapsulation of explants in beads of sodium alginate then loading them with vitrification solutions before cryogenic exposure



- **Droplet vitrification:**

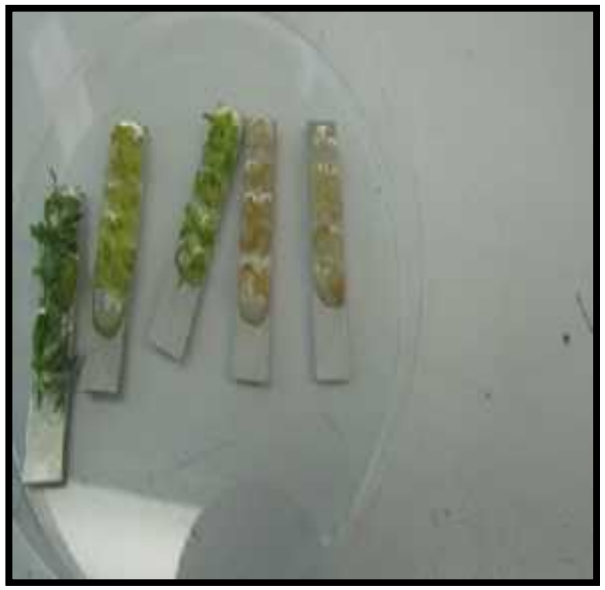
The explants are plated on a piece of aluminum foil loaded with a media supplemented with a cryoprotectant before dipping in liquid nitrogen



Cryopreservation procedure for the droplet vitrification method.

➢ **V- cryoplate:**

Plant material is placed on aluminum cryo-plates consisting of 10 wells containing alginate gel and osmoprotected with a loading solution then dehydrated with a proper plant vitrification solution (PVS) before cryopreservation



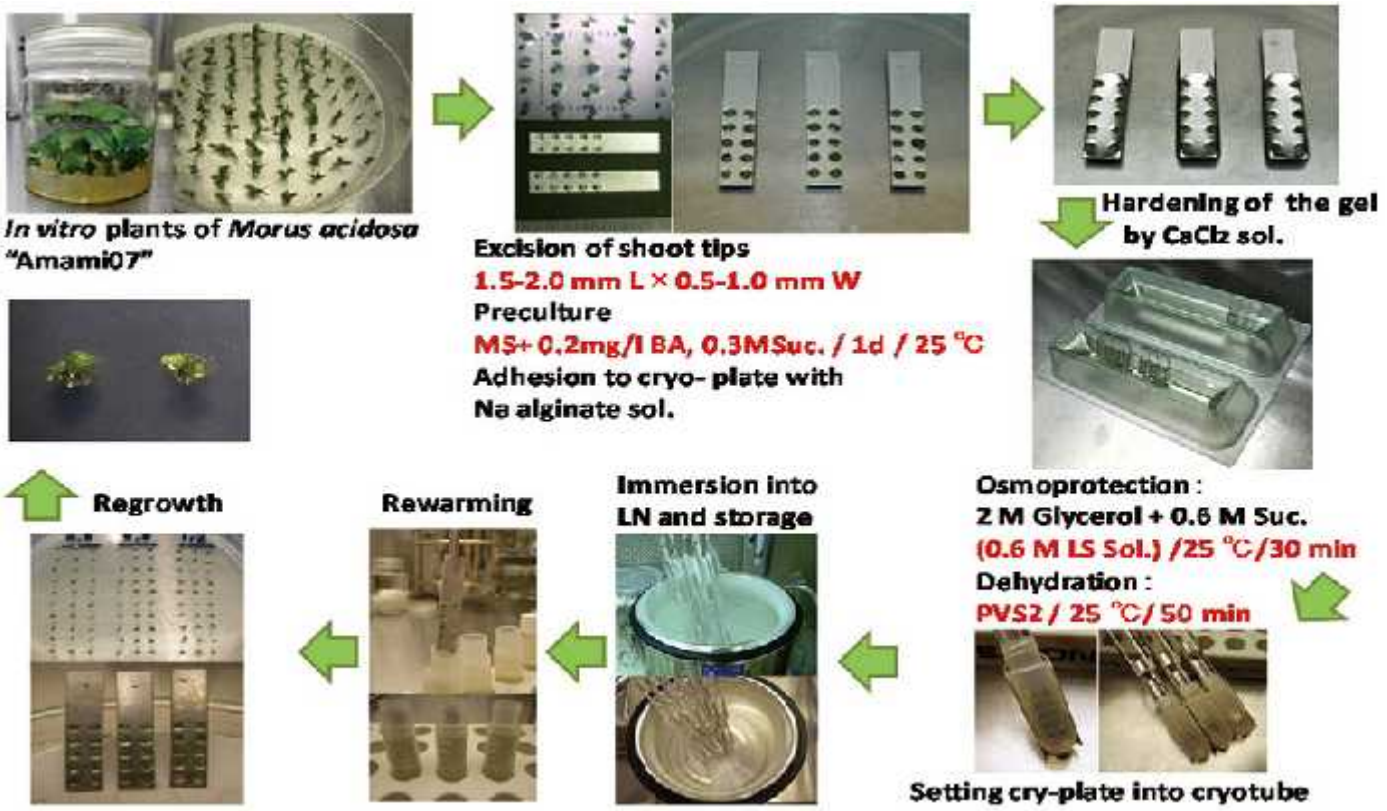
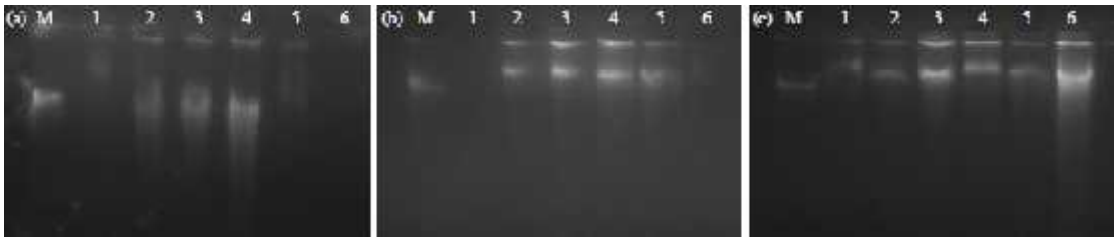


Fig. Procedures for cryopreservation of in vitro plants using V-cryo-plate method

4. DNA banking:

- DNA banking is a new technique where DNA is extracted from plant material at any growth

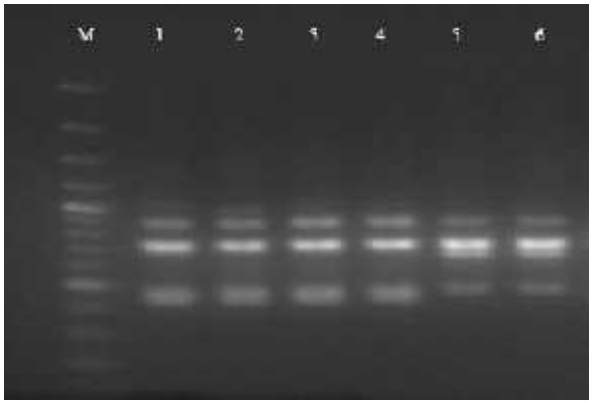


Advantages

- This new emerging technology has been applied in many genetic diversity programs for efficient identification, characterization and documentation of plant genetic diversity
- It allows assessing variation in plant diversity at DNA level and excludes any environmental interference

- DNA wasn’t extracted only for characterization purposes, but also it was stored in a form of DNA bank for long term conservation of plant germplasm.

- Simple and wide applicable



- Disadvantage:
  - Inability to the regenerate a whole plants was the main disadvantage of this method.
  - As for date palm, unfortunately few sequences of DNA are currently conserved in Gene banks

- The great socioeconomic and nutritional value of date palm all over the world in addition to the serious threats that jeopardize its genetic diversity; all imply that DNA banking is necessary for conservation date palm germplasm.

Recommendations

- Great efforts have been made globally to conserve the valuable genetic diversity of date palm.

- However, most of world production of dates and date palm trees are in the Arab world.

- Only few efforts have been made in several Arab countries for characterization, propagation and conservation of some date palm local cultivars

- More efforts are still needed in date palm research in Arab world especially in aspects of conservation.

- Unfortunately, no mutual projects between the Arabic counters for date palm conservation have seen light yet.

- So, there is a great responsibility upon Arab countries to start their own mutual strategy for date palm conservation using all the complementary approaches of conservation to keep this natural fortune of date palm germplasm safeguard for the whole humanity and future generations.

THANKS FOR LISTENING