Chapter 14
Radiation-Induced Mutations for Date Palm Improvement

S.M. Jain

Abstract Micropropagation technique is used for rapid shoot proliferation of date palm. Somatic embryogenesis is meant for clonal propagation of date palm and genetic gains can be captured through it, which is rather difficult by zygotic embryo due to its heterozygous nature. Genetic variability is highly desirable for the genetic improvement of crops, which can be either spontaneous or induced by mutagen treatments. Mutation-assisted breeding has been quite successful for the production of new mutant cultivars with desirable traits in both seed and vegetative propagated crops (see: http://www-mvd. iaea.org). In the IAEA date palm project, somatic embryogenic cell cultures were irradiated with gamma radiation, and regenerated plants were transferred to the greenhouse and treated with bayoud toxin, isolated from the causal fungus *Fusarium oxysporum* f. sp. *albedinis*. Several putative mutants tolerant to bayoud disease were initially maintained in the greenhouse and later transferred to the field for further evaluation. Over the last 4 years, these plants have not shown any sign of susceptibility to bayoud disease under field conditions, but they have yet to flower. Thus far, our results suggest that the combination of *in vitro* culture and mutagenesis would be an ideal system for date palm improvement. However, molecular tools are needed to characterize mutants for trait specific gene (s) identification and to develop molecular marker assisted selection and breeding programs. Date palm also has great potential to provide renewable energy or *bioenergy* or *green energy* for producing *bioethanol* and blend it with petrol in the transport industry.

Keywords Genetic variability • *In vitro* selection • Mutation • Tissue culture

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14.1 Introduction

The unique characteristics of date palm (*Phoenix dactylifera* L.) can be truly called a *tree of life* and is considered one of the most ancient plant cultivated in Mesopotamia some 4,000 years ago (Dakheel 2003; Omar and Hameed 2006). Few plant species have been so closely connected with the survival and wellbeing of humans living in hot and arid environments. This plant is basically responsible for the human settlements and expansion in hot and barren parts of the world, and forms the most sustainable agro-ecosystems in harsh dry environments. The date palm is distributed throughout the Middle East, North Africa, South Sahel, areas of East and South Africa, and even in certain parts of Europe and USA (Jain 2007). Dakheel (2003) studied the unique date palm production system under harsh climatic conditions, and described as due to:

- High resilience and tolerance to environmental stresses- high temperature and radiation, low soil and atmospheric moisture, extended periods of drought, high salinity levels, and large diurnal and seasonal fluctuations.
- High resource utilization efficiency and limited input requirement.
- High productivity.
- High nutritional value of date fruit.
- Long productive life, as long as 100 years and multiple uses.
- Creates equable microclimate within oasis ecosystems and thus enables agriculture development (Jain 2007).
- Helpful in the conservation of the fragile environment structure and reduce desertification risks.

Date palm belongs to the monocot family Arecaceae and is an arborescent, dioecious tall evergreen and highly heterozygous plant (Jain 2007). Date fruits are a most important source of human nutrition as well as an export item for many date palm-growing countries. The annual world production of dates was just over seven million mt in 2008 (FAOSTAT). The major bulk of date palm production, 75% of the total world production, comes from Egypt, Iran, Saudi Arabia, UAE, Algeria, Pakistan and Iraq. In the Kingdom of Saudi Arabia, over 200 date palm varieties are grown and date fruit production currently is 980,000 mt, which represents about 15% of world date production.

14.2 Date Palm Fruit as a Staple Fruit

The rich date fruit plays an important role in providing nutrition to humans living under the harsh climatic conditions. Date fruits are the rich source of sweeteners, glucose and fructose (Al-Eid 2006). Date syrup analytical studies showed that it is mainly composed of reduced sugars; glucose and fructose as major source of sugar fraction. Al-Ghamdi and Al-Kahtani (1996a, b, c) made detailed analyses of date

palm fruits and described their chemical properties, sugar content and minerals. The sugar content increases as the fruit ripens. The fruit chemical properties and mineral content vary depending on the genotype. However, there was no difference in fruit quality between *in vitro* plants and conventionally grown plants. Furthermore, nutritional analysis of date fruits (Kingdom of Saudi Arabia, Ministry of Agriculture) indicated that they contain:

- High percentage of carbohydrates (total sugars 44–88%).
- Fat (0.2–0.5%).
- Salts and minerals (15 different types).
- Proteins (2.3–5.6%).
- Vitamins and a high percentage of dietary fiber (6.4–11.5%).
- Flesh of dates contains 0.2–0.5% oil.
- Seeds of dates contain 7.7–9.7% oil, which has 14 types of fatty acids.
- Contains elemental fluorine useful for preventing dental decay.
- Contains selenium that prevents cancer and proper function of immune system.

Dates could be considered as an ideal food which provides a wide range of essential nutrients and potential health benefits.

14.3 Major Date Palm Diseases and Pests

Date palm suffers from several diseases and insect pests leading to severe economic lose to growers. There are about 25 diseases and disorders affecting date palm worldwide. Among them, 14 are caused by fungi (Karempour and Pejman 2007). Date bunch fading disorder (DBF) is the most harmful phenomenon damaging both the quality and quantity of date yield. In Iran, this disorder has caused wilting and drying of bunches and finally severe defoliation of date palms over the last 5 years (Karempour and Pejman 2007). In Egypt, 21 fungal species belonging to 15 genera were isolated from diseased date palm samples collected from different Egyptian localities (El-Deeb et al. 2007).

Bayoud disease is a serious threat to date palm plantations in North African Saharan and sub-Saharan regions, which is caused by a soil-borne fungus *Fusarium oxysporum* f. sp. *albedinis* (FOA) (Fig. 14.1). It was first observed in the Draa Valley, Morocco in 1870, and from there it reached the Algerian Central oasis around 1889, and it rapidly advanced into new areas. The disease has destroyed more than ten million palm trees in Morocco and nearly three million trees in Algeria. The rate of destruction by bayoud disease is thus estimated at 5% per year (Oihabi 2003). Presently, the disease is known to occur in Morocco, Algeria and Mauritania. Tunisia takes very drastic quarantine measures and strict surveillance to prevent the introduction of the disease from Algeria. The most popular commercial date palm cvs. such as Deglet Noor, Medjool and Boo Fagoos are under threat from bayoud disease.

Fig. 14.1 Soil-borne fungus *Fusarium oxysporum* f. sp. *albedinis* (FOA) is the causal agent of bayoud disease

Quenzar et al. (2001) identified two circular plasmid-like DNAs (S and R plasmids) in the mitochondria of date palm. By employing a PCR-based approach, they showed that the presence of R plasmid and absence of S plasmid can be considered as a reliable molecular marker of bayoud disease resistance. In this situation, the presence of S plasmid and the absence of R plasmid are correlated to the susceptibility to bayoud disease. This diagnostic molecular tool could ultimately become a simple, reliable, rapid and efficient approach to identify bayoud resistant and susceptible genotypes from the large pool of date palm lines.

Since 1994, the International Atomic Energy Agency (IAEA) has had a technical cooperation project on the selection of bayoud disease resistant date palm mutants by gamma radiation treatment, working in collaboration with Algeria, Tunisia and Morocco (Jain 2005, 2006), and it continued until 2008. There was a consultant meeting on bayoud disease of date palm, organized by IAEA in March 1996 (RAF/5/035 project). This meeting summarized the status of knowledge on the fungus FOA, its biotypes, its spread in North Africa, disease symptoms and movement of the pathogen in the host, characterization of the fungal toxin, short- and long-term strategies to breed for resistance, induction of variation for resistance in well-established date palm varieties using induced mutations and *in vitro* culture techniques, improvement of screening procedures to select for resistance in breeding programs as well as characterization of variation in the pathogen population and use toxin for *in vitro* selection.

Red palm weevil (RPW) is a major pest in date palm-growing countries in the Near East including the United Arab Emirates, Iran, Egypt and others (Oihabi 2003). It first appeared in the Middle East in 1985 and is of great concern to the date-palm growers in these countries. The control of RPW is mainly done by applying chemical insecticides through direct injection into the trunk of the date palm tree or by fumigation. Pheromone traps are also commonly used to control RPW, but still

require more refinement and effectiveness to control this pest. Baculoviruses could be another way to control RPW, especially genetically engineered ones inserted with a set of genes dealing with neurotoxin, light-emission (firefly gene) and heat tolerance.

14.4 *In Vitro* Culture of Date Palm

Date palm is well known to propagate both sexually through seeds and vegetatively by offshoots that are produced from axillary buds situated at the base of the trunk during the juvenile life of the palm tree. Seed propagation of date palm is not appropriate for commercial production due to the heterozygous characteristics of seedlings, which is related to the dioecious nature of the date palm, half of the progeny are generally male and do not produce fruits; also, large phenotypic variation can occur in the progeny (Jain 2006). Currently, there is no known method for sex determination of date palm at the early stage of tree development making it rather difficult to discriminate between productive female and non-productive male trees in the nursery before transplanting them to the field. Furthermore, the seed propagation method has another limitation in that the growth and maturation of seedlings is extremely slow, and only begin to fruit after 8–10 years of planting. The ideal way would be to look for molecular markers for sex determination along the lines of work done in papaya (Deputy et al. 2002).

Offshoot production is slow; their numbers are limited, laborious to separate and cannot meet the rapidly growing demand of varieties. Normally offshoot numbers vary from 10 to 30, depending on the genotype, and are produced only within a certain period time in the mother palm's life (Jain 2007). No field-based methods are yet available for increasing the number of offshoots per plant. There are only a few commercial tissue culture laboratories worldwide micropropagating date palm for large-scale plant production (for more information see: Al Kaabi and Zaid 2003).

In vitro culture techniques such as somatic embryogenesis and organogenesis have been effectively used for large-scale plant multiplication of horticultural crops and forest trees (Jain and Gupta 2005; Jain and Haggman 2007; Jain and Ishii 2003). Plant multiplication via organogenesis is routinely followed in commercial laboratories worldwide especially in ornamental plant industries and also to some extent in fruits and cash crops like coffee, sugarcane etc. The cost of plant production is generally high due to labor and electricity, which reduces the profit margin. Many Western companies have started outsourcing plant multiplication facilities to low labor cost countries such as India, China, Brazil, Kenya, Tanzania and others; in fact, most of the date palm micropropagation commercial laboratories are operating in countries with low labor cost. The performance of *in vitro* propagated plantlets seems to be improving in terms of yield and early flowering. Al-Ghamadi and Al-Kahatani (1996a,b,c) made detailed comparative analyses of fruit quality of micropropagated and conventionally-propagated plants and found no major variation in fruit quality and properties. Results clearly indicate that *in vitro*-grown date palm

are quite uniform in terms of fruit quality and physical properties. Smith and Aynsley (1995) reported on field performance of tissue culture-derived date palm clonally produced by somatic embryogenesis. These plants started bearing fruits within 4 years of field planting of small plants with a leaf length of 100 cm and 1.5 cm diameter at the base. Fruit from the tissue culture-derived plants, cv. Barhee, was indistinguishable from the fruits of plants originated from offshoots. These results certainly justify the commercial scale of micropropagation procedures of somatic embryogenesis to provide rapid, cost-effective means of obtaining elite date palm planting material. However, this approach has a major bottleneck in that the plant multiplication rate is highly genotypic dependent, and may require modification of culture medium, depending on the genotype. For more information see Jain (2006) who described the advantages and limitations of date palm micropropagation. Some of the major advantages of micropropagation are year-round availability of plants, quality control, rapid production of plants of elite cultivars and cold storage of elite genetic material.

14.4.1 Techniques for Plant Regeneration

Date palm tissue culture work has revolved around somatic embryogenesis (Al-Khayri 2005; Fki et al. 2003) and organogenesis for plant regeneration (Al-Khayri 2007; Khierallah and Bader 2007). Aaouine (2003) reported plant regeneration from 30 genotypes of date palm via direct shoot organogenesis. Many commercial laboratories in Europe, the Middle East, the United States, North Africa and South Africa are using a combination of somatic embryogenesis and organogenesis. Furthermore, the initiation period for somatic embryogenesis induction is 4–6 months as compared to 8–10 months for organogenesis; the total time from induction phase to plant marketing is 40–44 months via somatic embryogenesis vs. 60 months via organogenesis (Aaouine 2003).

Murashige and Skoog (1962) formulated the most commonly-used culture medium for both somatic embryogenesis and organogenesis of date palm, which also is modified depending on the genotype or cultivar (Jain 2006). Young Maktoom cv. offshoots from 2 to 3 year old date palms were used for direct shoot induction after they were sterilized with commercial bleach and rinsed with sterile distilled water (Khierallah and Bader 2007).

The somatic embryogenesis approach for date palm plant regeneration seems to be more effective for clonal propagation. Fki et al. (2003) improved somatic embryogenesis protocol of date palm cv. Deglet Noor for large-scale clonal propagation. Initially, embryogenic callus cultures were initiated from both leaf and inflorescence explants on MS (Murashige and Skoog) medium containing 0.5 and 10 mg/L 2, 4-D (Figs. 14.2 and 14.3). These cultures were used to develop highly proliferating cell suspension cultures in the liquid medium supplemented with 1 mg/L 2, 4-D (Figs. 14.4–14.6). Somatic embryos were initiated from actively growing cell suspension (Fig.14.7), and finally somatic embryos were germinated (Figs. 14.8 and 14.9) and the whole plantlets regenerated (Figs. 14.10 and 14.11).

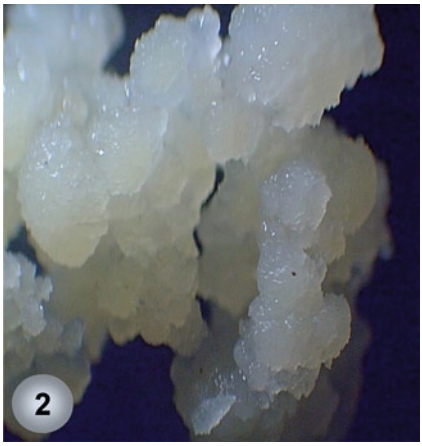

Fig. 14.2 Initiation of somatic embryogenic callus from off shoot of date palm cv. Deglet Noor

Fig. 14.3 Further development of somatic embryos

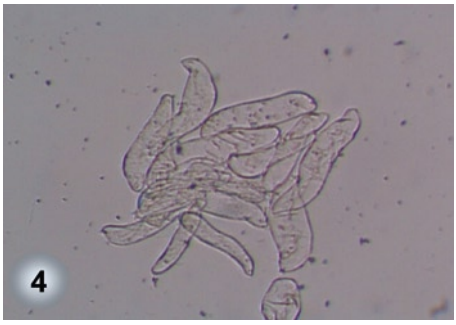

Fig. 14.4 Rapid growing somatic embryogenic cell suspension cultures

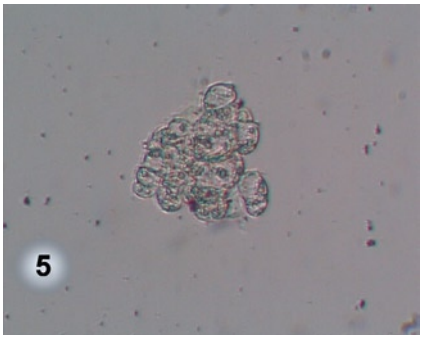

Fig. 14.5 Rapid growing somatic embryogenic cell suspension cultures

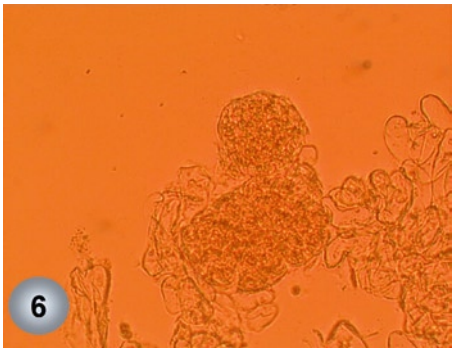

Fig. 14.6 Rapid growing somatic embryogenic cell suspension cultures

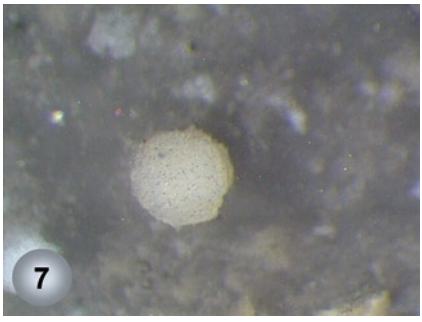

Fig. 14.7 Developing somatic embryo of date palm

Fig. 14.8 Germinating date palm somatic embryos

Fig. 14.9 Somatic seedlings of date palm with well-developed shoots and roots

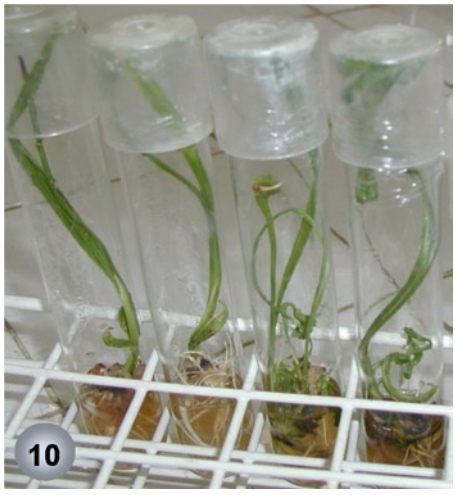

Fig. 14.10 Well-developed somatic seedlings with shoots and roots

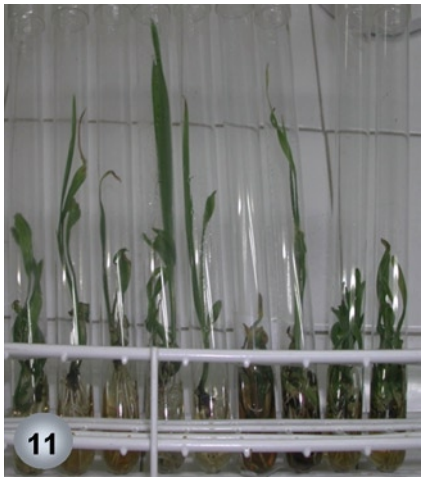

Fig. 14.11 Well-developed somatic seedlings with shoots and roots

The overall production of somatic embryos can reach 10,000 units per liter per month. The partial desiccation of the mature somatic embryos significantly improves the somatic embryo germination rate from 25% to 80%. Cutting back of cotyledon leaf was stimulatory to the germination rate. Furthermore, flow cytometry analysis showed no variation in ploidy level of somatic seedlings. Several research groups have modified the culture medium composition by adding vitamins, adenine sulfate, thiamine, glycine, glutamine, myo-inositol and activated charcoal (Al-Khayri 2005). The role of vitamins in date palm tissue culture is not known. For more details see Al-Khayri (2005) and Jain (2006).

14.5 Mutation Induction

The exploitation of genetic variability is essential for the development of new cultivars. Genetic variability can be induced by chemical and physical mutagens, T-DNA insertional mutagenesis, and tissue culture-derived variation or somaclonal variation. The most common physical mutagen used is gamma radiation. In this chapter, we will deal only with physical mutagens. Induced mutations are random changes in the nuclear DNA or cytoplasmic organ, resulting in chromosomal or genomic mutations that enable plant breeders to select useful mutants such as disease resistant, high yield etc. First of all, gamma irradiation breaks DNA into small fragments and secondly DNA starts a repair mechanism. During this second step, new variations develop or mutations occur. In date palm, there is hardly any work done on mutation induction, except that of FAO/IAEA Coordinated Research Project on development of bayoud disease resistant date palm mutant cultivars in North Africa (Jain 2005, 2006). Mutation induction in date palm is feasible now due to a reliable plant regeneration system via somatic embryogenesis and organogenesis. The somatic embryogenesis system is the more preferable approach due to single cell origin of somatic embryos which prevents or reduces the occurrence of chimeras. Moreover, mutant somatic embryos are germinated into direct plantlets in a single step, avoiding the laborious rooting step. The irradiation of multicellular structures, e.g. seed, meristem tissue or offshoots, may result in chimeras in regenerated plants, and that would require a lot of extra work to dissociate chimeras by plant multiplication up to M1V4 generation (Jain 2007).

14.5.1 Determination of Radiosensitive Dose

Plants differ in radio sensitivity and that is why it is important to make a radiosensitive curve to determine LD_{50} dose for mutation induction. High radiation doses are detrimental to the plant genome and cause heavy damage to DNA. This leads to large number of mutations, which are mostly undesirable; it is cumbersome to identify useful mutants, and the handling of the mutant population becomes more difficult.

Fig. 14.12 Rapid growing somatic embryo cell suspension ideal for radiation treatment

In some crop plants, a low radiation dose promotes shoot growth, e.g. in citrus 30 Gy dose stimulates shoot growth (Jain personal communication) and 10 Gy maintains somatic embryogenesis nature of date palm up to 3 years (Drira, personal communication). In date palm 20–30 Gy was used for mutation induction depending on the genotype used. For Deglet Noor date palm cv., LD_{50} of somatic embryogenic cell suspension cultures was 20 Gy and used for mutation induction. Actively growing cell suspension in the growth phase (Fig. 14.12) was transferred onto filter paper in the Petri dish. Cell clumps were uniformly spread on the filter paper and the dishes sealed with Para film. Cells were irradiated with different gamma radiation doses and they were transferred for overnight to fresh solid culture medium for recovery from the radiation treatment. The irradiated cells were transferred into liquid medium, and distributed in 50 mL flasks containing 30 mL liquid medium. The cell viability test was made with FDA (fluorescein diacetate) staining to determine the cell survival rate after the different radiation dose treatment. The number of surviving cells was calculated on the basis of cells per mL per radiation dose. After this step, in each flask, 100,000 cells per mL were added as starting material and the cell growth was determined after 1 week. The number of cells per mL per radiation dose treatment was counted with a haemocytometer. These results established the radiosensitive curve and determined LD_{50} radiation dose for mutation induction.

14.5.2 Mutant Isolation

Mutant isolation can be done in two ways, either in a single step or stepwise selection. In the first approach, irradiated cells are put under very high selection pressure for the isolation of mutant cell clumps/lines. The initial selection pressure should be

as high as high LD_{75}. Isolated mutant cells are removed and transferred onto fresh culture medium with reduced selection pressure allowing them to recover from the initial selection pressure for about 1 week. The selected lines are put for shoot and root differentiation. Before selected mutant lines are put for shoot differentiation, they should be grown for two generations devoid of selection pressure and then returned to the selection pressure. This step is done to make sure that the selected mutant lines are stable and due to genetic changes rather than epigenetic changes. In the second approach, the selection pressure is reduced stepwise, from high to low concentration. All other steps are more or less similar to the first approach.

With *in vitro* selection of mutants, normally the type of selection pressure varies, e.g. salt concentration, fungal toxin, polyethyl glycol (PEG), herbicide etc. For appropriate selection pressure, it is desirable to determine LD_{50} dose.

A third option is to select mutants at the whole plantlet level, e.g. by spraying herbicide or withholding water for drought-tolerance selection, fungal toxin spraying or injection. In date palm, bayoud disease resistant mutant plants were selected in the greenhouse by treating them with isolated toxin from *Fusarium oxysporum* f. sp. *albedinis* fungus, the causal agent (Jain 2006). These plants have been in the field for the last 4 years and so far are doing just fine.

14.6 Date Palm as a Bioenergy Source

Today the whole of humanity is very much dependent on fossil energy for routine daily life. The surge in the industrialization in the developing world has enhanced fossil energy consumption. Emerging economies like China and India with huge populations are consuming more fossil fuel energy than ever before due to improvement in the socio-economic status of consumers. China is buying 40% more fossil fuel from the international market followed by India 12%. Consequently, the price of fossil fuel is skyrocketing, almost exceeding USD 70.00 per barrel on the international market and this is gradually creating serious problems in the economies of the world. As fossil fuel consumption rises, another point of concern is the gradual depletion of fossil fuels that could last for only 40–50 years more. The rise in fossil fuel energy consumption in the transportation sector, together with other gaseous pollutants, is adversely influencing world climatic conditions. For example, global warming is finally accepted as a cause of concern by the international scientific community and the United Nations. The rise in global temperature even by 1°C will have a negative impact on the earth including reduction in agriculture production, appearance of new pests and diseases, disappearance of some old pests and diseases, water shortage, increase in carbon dioxide and ozone layer depletion. An increase in the ozone layer hole would increase ultra violet-B radiation on the earth that would increase skin cancer incidents and decrease the photosynthesis rate resulting in poor performance of plant productivity.

Renewable energy is the alternate source to the fossil fuel energy, which can be termed as *green energy* or *bioenergy* or *plant-based energy*. Biofuel is gradually replacing petroleum in the transport sector, which is a blend of petrol and bioethanol

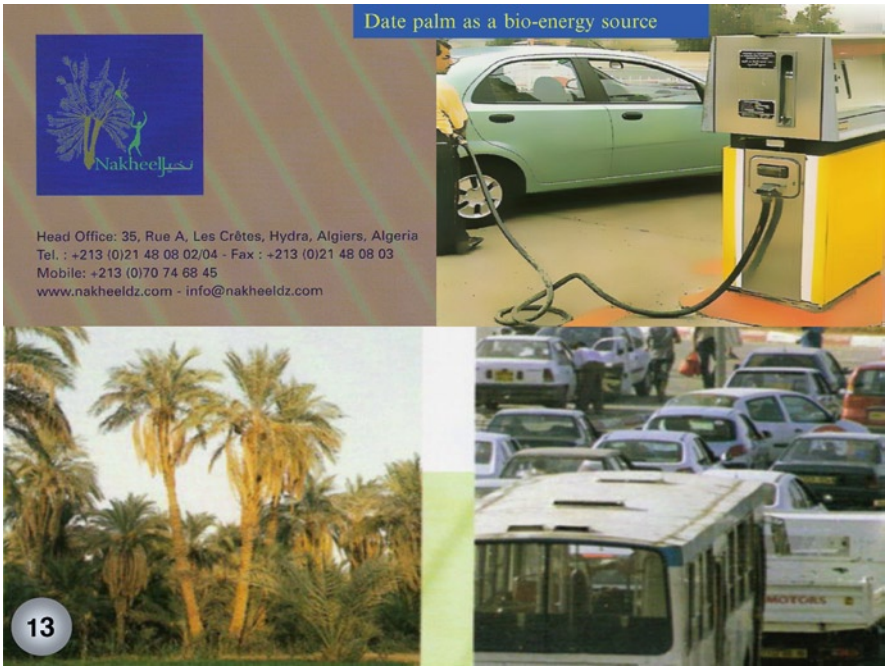

Fig. 14.13 Nakheel, an Algerian biotechnology company, is the first company in the Arab world to promote date palm as a renewable energy crop to produce bioethanol in transport

or biodiesel and fossil diesel. Brazil is the world leader in exploiting biofuel in the transport industry, especially bioethanol produced from sugarcane. The Europeans are leaders in commercial production of biodiesel, mainly from vegetable oil, e.g. *Brassica.* However, the production of biodiesel is comparatively small as compared to bioethanol production in Brazil and USA. Date palm could become a major source of producing bioethanol, since its fruits have a high percentage of carbohydrates (total sugars 44–88%). Millions of date palm trees are grown in the Middle East, North Africa, and South Asia, and they provide food and nutrition to millions of people, and could also become a major source of bioenergy. In Algeria alone, the estimated number date palm trees is over ten million, and production increased from 302,993 mt in 1997 to 526,921 mt in 2007. This increased production provides extra income to date palm growers if price levels are maintained.

The over production of date palm fruits, however, could lead to price reduction and loss of farm income. These problems can be overcome by using date palm for bioethanol production. In Algeria, Nakheel, a biotechnology company, is the first enterprise in the Arab world to promote renewable energy to use in transport. Date palm is the main source for producing bioethanol, which can easily be blended with petrol (Fig. 14.13). In the future, there is a potential to increase date palm-growing areas when the demand is high without compromising dates as a food and nutrition source. Moreover, date palm micropropagation is well established for clonal propagation and the supply of planting material year around.

14.7 Conclusion and Prospective

Date palm is a life-line of people living in Saharan and sub-Saharan regions and also an important source of income in Near Eastern countries. Most of the date palm trees are very old, as much as 70–100 years and probably are becoming more vulnerable to various diseases and pests. One of the reasons could be due to global warming or global climatic change. An increase in global temperature would bring new pests and disease and eliminate some existing types. Since date palm has a long life cycle, it could become more vulnerable to global warming, and that is why it is highly desirable to pay more attention to the genetic improvement of date palm varieties that could withstand natural calamities without compromising yield and quality. The use of chemical insecticide and pesticides is very common to control diseases and pests of date palm. These practices could become a serious hazard to human health and that may also curtail the export market. Innovative techniques need to be applied to the control of disease and pests, and that is where genetic modifications of organisms would be highly effective. Genetic engineering of baculoviruses may be of great help in controlling the RPW by inserting a set of genes including neurotoxin (a gene from the scorpion or snake), light-emitting (fire-fly) and heat tolerance (bacterial gene). The engineered baculoviruses would multiply inside the insects and kill them instantly. One could monitor the rate of viral multiplication inside the insect by light meter. Insertion of *Bt* gene in date palm won't be the proper approach due to long life cycle of date palm and it would be rather difficult to predict the behavior of transgenes in the long run. Moreover, food safety regulations do not permit insertion of the *Bt* gene in food crops.

The progress of *in vitro* culture techniques has enabled date palm micropropagation to become a routine technique for large-scale plant production in many countries. The influence of genotype has handicapped micropropagation of different commercially valuable date palm varieties. This area needs serious attention by modifying, through more empirical work, the culture medium well suited for several date palm cultivars. Now the question arises of how well the molecular approach would assist plant tissue culturists to modify the culture medium and growing conditions or the selection of appropriate explants or pre-conditioning of explants. To answer these questions, plenty of work is foreseen; in other words this area of research is *virgin.*

The date palm shoot multiplication rate could be improved by using a liquid culture system or *bioreactor.* Few groups have started working on liquid culture for *in vitro* propagation of date palm. The RITA bioreactor, based on temporary immersion system, should be tried in date palm shoot multiplication and somatic embryo production. Micropropagation via organogenesis or direct shoot formation is extensive and labor-intensive. Somatic embryogenesis may reduce labor cost and also assist in developing automated somatic embryo production. However, genetic fidelity of micropropagated plants should be maintained with minimal somaclonal variation, otherwise there will be severe economic loses to growers. Molecular marker analysis would be an ideal approach to identify genetic variability at the early stage of plant development. It would be difficult to identify point mutations or any genetic change at the early stage of plant development because it may not express change

phenotypically or may express it at the latter stage of plant development. This scenario occurred in African oil palm tissue culture-derived plants in Malaysia and the oil palm industry lost millions of US dollars.

Haploid production in date palm has not yet been accomplished. Inflorescence culture will be one way to induce haploid somatic embryo production. Fki et al. (2003) induced callus from immature inflorescences of date palm cv. Deglet Noor, and the calli originated from the proliferation of floral primordia showed embryogenic potential. The capacity of the inflorescence to form callus was much higher than cultured leaves. They did not determine the ploidy level of callus and regenerated plants from inflorescence-derived callus. In the future, the success of this type of work could revolutionize date palm genetic improvement programs as well as molecular genetics for useful gene identification.

Somatic embryogenic cell suspension is an excellent system for mutation induction and isolates useful mutants of date palm. Direct mutant somatic embryos can be produced and germinated into mutant somatic seedlings. These mutant seedlings can further be micropropagated for large-scale production. The utmost care should be taken when handling somatic embryogenic cultures; failure to do so significantly increases the chances of getting somaclonal variation. This approach is an excellent example of combining mutagenesis and biotechnology for date palm improvement. Transgenic date palms have a long way to go before consumers accept them and consequently export markets initially may also be lost. Therefore, the transgenic approach to modify date palm should be followed with a great caution, even though it has a great potential to overcome several of its problems.

There is a complete lack of date palm molecular biological research to address several issues facing date palm genetic improvement. Molecular marker-assisted selection and breeding need serious attention to identify trait-specific genes from natural or induced genetic variability. Functional genomic date palm breeding could probably become a reality in the future that will speed up genetic improvement of date palm.

Date palm has a great potential as a bioenergy crop, since date fruits have high sugar content and would be ideal for bioethanol production. Molecular genetics will be of great help in gene identification for sugar production and related chemicals. This really needs utmost attention.

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