通风对好好吧无性系芽的增殖能力 和抗旱能力的影响作用

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摘 要:该文旨在研究通风在不同好好吧无性系芽增殖阶段对它们增殖体的叶面积、地上部分总面积和央水 能力的作用,筛选抗旱性好的好好吧无性系。研究结果表明,在13个好好吧无性系中,通风使他们的叶面积、 总面积和抗失水力均有增加。其叶面积分别增加了 72.2%到 470.5%,其总面积分别增加了 39.0%到 145.8%,其抗失水力分别增加了(失水分别减少了)22.2%到 138.0%。从中选择了一个叶面积和总面积都增 加最多(分别为 470.5%与 145.8%)和失水减少次多(86.7%)的耐旱好好吧无性系 CQ63,可供沙漠和旱地种植。 关键词:好好吧;增殖;通风;失水

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Effect of ventilation on the proliferation and drought-resistance of jojoba (Simmondsia chinensis) clones

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Abstract: The purpose of this research is to study effect of ventilation on leaf area, shoot area and water loss in different jojoba clones, and select some drought-resistant jojoba clones from those at their proliferation stage. The research results showed that ventilation helped increase their leaf areas from the percentage of 72. 2% to 470. 5%, total area of jojoba propagules from 39% to 145. 8% and their resistance to(decrease in) water loss from 22. 2% to 138. 0% in 13 jojoba clones. One good drought-resistant jojoba clone, CQ63, was selected with the most increase of 470. 5% in leaf area and 145. 8% in total area as well as a greater decrease of 86. 7% (the second-most one among 13 clones) in water loss to be planted in arid desert and land. Key words: jojoba (*Simmondsia chinensis*); proliferation; ventilation; water loss

Jojoba (Simmondsia chinensis L.) is a native oilseed shrub of the Sonoran desert of the southwestern USA and northern Mexico (Vermaut Sabien *et al.*, 1995), an evergreen crop of high economic interest which is grown for the exceptional quality of its seed oil(Amarger and Mercier, 1995). Native jojoba populations have growing temperature ranging from 9 °C to 50 °C (Gentry HS, 1958), being drought-resistant and salt-resistant (Mills and Benzioni, 1977; Yermanos, 1977). The dry

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seeds of jojoba contain about 50% lipids in the form of simple wax esters that have properties similar to those of sperm whale oil, which are used in cosmetics, pharmaceuticals and polish industries (Jacoboni and Standardi, 1987; Benzioni *et al.*, 1992). Therefore, jojoba was introduced to many countries during the late 1970s and early 1980s.

Currently, the main jojoba oil producers are Israel, USA, Argentina, Australia and Mexico. Nowadays, increasing attention has been paid to Jojoba plantation and breeding. New jojoba plantations are grown from vegetatively propagated material and seeds, but in vitro propagation is a good alternative method offering some important advantages such as mass propagation without the limitation by the number of explants and the production of pathogen-free plants and new good-quality breeds or cultivars (Mills et al., 1997). Because tissue culture technique is being widely used for crop improvement and clonal multiplication, the selected jojoba clones are multiplied by micropropagation method(Benzioni et al., 1992; Sardana and Batra, 1998; Farook, 1998; Roussos et al., 1999), and jojoba micropropagation has been described (Mills et al., 1997; Roussos et al., 1999). However, micropropagated jojoba plantlets often lose their ability to control water loss caused by high relative humidity and limited gas exchange(Mills et al., 2001; Ziv, 1991). The improvement of the ability through ventilation(by using specific membranes) at the elongation stage had been reported (Mills et al., 2001). But up to now, the effect of ventilation on jojoba clones at the proliferation stage has not been shown. Therefore, in the present study, effect of ventilation on different jojoba clones is studied and drought-resistant jojoba clones are selected from them.

1 Materials and Methods

1.1 Plant material and culture conditions

Jojoba clones used in the present study are mico-propagated and provided by DAVID Mills and ALIZA Benzioni's plant tissue culture laboratory

in Institutes for Applied Research, Ben-Gurion University of the Negev, Israel. They include Clone64, Clone154, Clone Q63, Clone78, Clone85, Clone F, Clone141, Clone96, Clone OMER, Clone Male, Clone K120, Clone32, Clone Kalial1. They were cultured in 5 mL of modified MS medium (Mills et al., 1997) for proliferation in a growth room. After a culture of 7 weeks, they were cut into single nodal segments, which had opposite axillary buds and leaves and contained two parts; one above buds and leaves, about 0.5 mm long; another under them, about 10 mm. Eight nodal segments excised from each clone were cultured in 70 mL of the same medium as above for proliferation in a growth vessel, magenta box (375 mL in volume). Every growth vessel was sealed with one lid which had one 16 mm diameter membrane at the center, corresponding to a membrane area of 207 mm², to create ventilation treatments. In the control treatment, every growth vessel was sealed with one lid without any membrane, and every vessel also contained eight nodal segments from the same clone. All the vessels were sealed with adhesive tape at the lid edge. Nodal segments inside them were cultured for twenty eight days in a growth room at a temperature of 25 ± 1 °C under $45 \sim 55 \ \mu mol \ m^{-2} s^{-1}$ photon flux supplied by cool white fluorescent bulbs for a photoperiod of 16 h light.

1, 2 Growth measurements

After a culture of twenty eight days, the following parameters were determined: shoot area, leaf area and water loss. For area determination, shoots and leaves were photocopied, the image was scanned, and the area was measured with the image analysis software NIH Image. For water loss determination, propagules were taken out of the vessels separated from callus at the base and weighed every ten minute (60 minutes /seven times) in an electric balance under room conditions (usually 23 ± 2 °C and 61 ± 3 % RH). Water loss was calculated by dividing the loss of weight after designated periods by the propagule surface area.

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1.3 Statistics

In each experiment, one vessel containing eight propagules was examined for every treatment. Sixteen propagules (control and treatment) in every clone were sampled for area and water loss. Experiments were repeated 2~3 times. Data were evaluated by analysis of variance (Fisher's Protected LSD; Probability of 5%) using super ANOVA. Figures were prepared with Excel.

2 Results and Discussion

All segments at the multiplication stage were cultured in magenta vessels with 16 mm diameter membrane or closed vessels (no membrane). After twenty eight days, the above thirteen jojoba clones (Fig. 1) were examined for leaf area, shoot area and water loss.

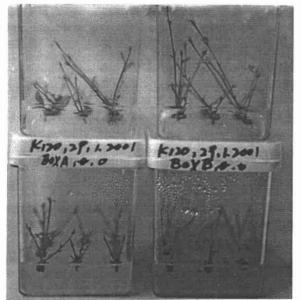


Fig. 1 Jojob clone Kaliall's propagules cultured for 28 days in growth vessels and in growth room Up:every vessel sealed with one lid which had one 16 mm diameter membrane at the center Down:every vessel sealed with one closed lid

2.1 Effect of ventilation on leaf area and total area of propagules in jojoba

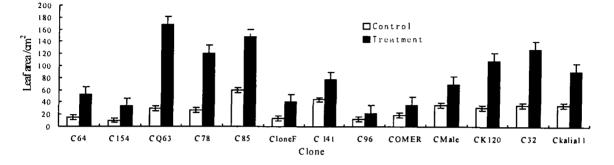
As shown in Fig. 2 and Fig. 3, ventilation had a positive effect on the growth of leaf area and total shoot area of propagules in different jojoba clones. According to the equation; area increase rate = leaf area or total shoot area of tested propagule/leaf area or total shoot area of control propagule. From clone C64 to clone Kalia11:(1)Leaf areas of jojoba propagules from the vessels with 16 mm diameter membrane in turn increased 247. 3%, 227. 5%, 470. 5%, 347. 4%, 148. 8%, 192. 9%, 73. 9%, 72.2%, 92. 1%, 95. 8%, 250%, 265. 7% and 155.4% over than that of those from the closed ones without membrane. Among them, leaf area of clone C63 increased most, by 470. 5%. The next one was clone C78, whose leaf area increased by 347.4%. Leaf area of clone C96 increased least, by 72.2%; (2) Total areas of jojoba propagules from the vessels with 16mm diameter membrane in turn increased 167%, 50%, 145. 8%, 81. 5%, 63. 2%, 80%, 57%, 39%, 40%, 50%, 76.8%, 47.6% and 122% over that of those from the closed ones without membrane. Out of them, total area of clone CQ64 increased most, by 167%. The next was clone CQ63 and clone Kalia11, whose total area increased by 145.8% and 122%, respectively. That in cloneC96 increased least, only by 39%. The increase of leaf area and shoot area caused by ventilation might be due to two aspects; one was that ventilation could decrease the inhibition of such volatiles as ethylene, ammonia, methyljasmonate and so forth on propagules, which could not be accumulated because of ventilation; the other was that ventilation could improve the carbon dioxide supply in growth vessels so that photosynthesis was enhanced and propagules could grow well, Meanwhile, leaf color was light green in closed vessels and darker under ventilation in the above clones like Rose and hosta plantlets (Shallanon and Maziere, 1992; Murphy et al., 1998). Relatively high water content is a characteristic of hyperhydric plants (Ziv, 1991). Reduction of water content by ventilation was also reported for carnation (Majada et al., 1997). In papaya and potato, it was also showed that ventilation made leaf area increased(Lai et al., 1998). However, because of reduced RH, reduction of leaf area in potato was also reported(Kosai et al., 1993). The difference in response to ventilation among different species might

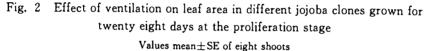
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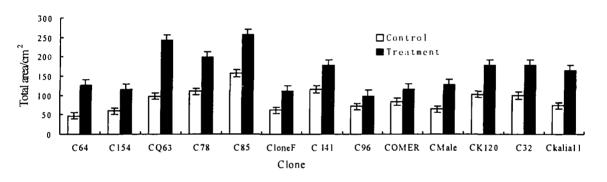
be due to the differences in ventilation rate and media or plant genotypes(Mills *et al.*, 2001).

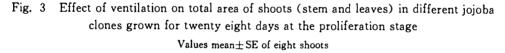
2. 2 Effect of ventilation on water loss of jojoba propagules in thirteen different clones

As shown in Fig. 4, all the propagules in different jojoba clones from the vessels with 16mm diameter membrane lost water at slower rate. This demonstrated that ventilation had a negative effect on water loss of the propagules in different jojoba clones. According to the equation, water loss rate = water loss weight of control propagule/water loss weight of tested propagule. From clone C64 to



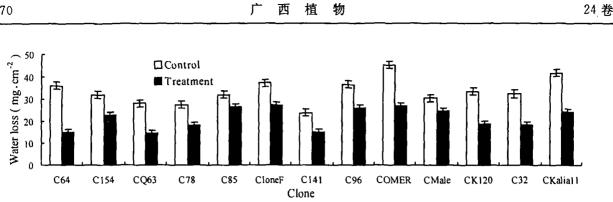


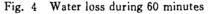




clone Kalial1, during sixty minutes, their water loss in turn decreased 138%, 41%, 86. 7%, 52%, 22. 2%, 37. 6%, 58. 3%, 47. 2%, 70. 3%, 23. 5%, 76. 9%, 24. 3%, 80% and 70% over than that of those from the closed ones without membrane. Among them, the clone C64 decreased most, by 138%. The next one was clone CQ63, whose water loss decreased by 86. 7%. That in cloneF decreased least, only by 22. 2%. Therefore, the response of different clones to ventilation was rather different. The higher its water loss rate was, the less a clone lost water. Such resistance can result from closure of stomata, which was caused by the decrease of their density when humidity declined in the culture tubes (Ghashyghaie *et al.*, 1992) or a decrease in cuticular transpiration due to increased deposition of epicuticular wax on the leaf surface or both(Ziv, 1991; Ziv, 1986; Majada *et al.*, 1998). The improvement of stomatal function in response to ventilation is due to a decrease in K^+ concentration in guard cells (Majada *et al.*, 1998).

In conclusion, all the thirteen jojoba clones examined had increase in leaf area, total area(leaf and stem area) and water loss under ventilation, so ventilation will help jojoba clones grow and resist water loss. It is possible that drought-resistant jojoba clones can be selected through ventilation. Here one good drought-resistant jojoba clone, CQ63, was





Effect of ventilation at the proliferation stage on subsequent water loss of shoots in clones C64-Ckalial1, Shoots were removed from vessels, kept under room conditions and weighed periodically. Values mean±SE of eight shoots per treatment and per clone.

selected with a larger increase of both 470, 5% in leaf area and 145.8% in total area as well as a greater decrease of 86.7% in water loss.

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References:

- Amarger V, Mercier L. 1995. Molecular analysis of RAPD DNA based marker: Their potential use for the detection of genetic variability in jojoba (Simmondsia chinensis L. Schneider) [J]. Biochimie, 77: 931-936.
- Benzioni A, Nerd A, Rosengartner Y, et al. 1992. The effect of Nacl salinity on growth and development of jojoba clones. I. Young plants[J]. J Plant Physiol, 139, 731-736.
- Farook. 1998. Standard in vitro experimental protocol for high frequency mass micropropagation of jojoba[J]. Advancesin-Plant-Sciences, 12: 361-366.
- Gentry HS. 1958. The natural history of jojoba (Simmondsia chinensis) and its cultural aspects[J]. Econ Bot, 12: 261-265.
- Ghashyghaie J, Brenckmann F, Saugier B. 1992. Water relations and growth of rose plants cultured in vitro under various relative humidity[]]. Plant Cell Tiss Org Cult, 30: 51-57.
- Jacoboni A, Standardi A. 1987. Tissue culture of jojoba (Simmondsia chinensis Link)[J]. Acta Hortic, 212: 557-560.
- Kosai T, Tanaka M, Jeong BR, et al. 1993. Effect of relative humidity in the culture vessel on the growth and shoot elongation of potato plantlets in vitro[J]. J Jap Soc Hort Sci, 62: 413-417.
- Lai CC, Yu TA, Yeh SD, et al. 1998. Enhancement of in vitro growth of papaya multishoots by aeration[]]. Plant Cell Tiss Org Cul, 53: 221-225.
- Murphy KP, Santamaria JM, Davies WJ, et al. 1998. Ventifation of culture vessel. Increased growth in vitro and sur-

vival ex vitro of delphinium[]]. J Hort Sci Biotechnol, 73: 725-729.

- Mills D, Friedman R, Benzioni A. 2001. Response of jojoba shoots to ventilation in vitro[]]. Israel Journal of Plant Sciences, 49: 197-202.
- Majada JP, Fal MA, Sanchez-Tames R. 1997. The effect of ventilation rate on proliferation and hyperhydricity of Dianthus caryophyllus L. []]. In Vitro Cell Dev Biol, 33; 62-69.
- Majada JP, Centeno ML, Feito I, et al. 1998. Stomatal and cuticular traits on carnation tissue culture under different ventilation conditions[J]. Plant Growth Reg, 25: 113-121.
- Mills D, Benzioni A. 1977. The effect of Nacl salinity on growth and development of jojoba clones: Nodal Segments grown in vitro[]]. J Plant Physiol, 139: 737-741.
- Mills D, Wenkart S, Benzioni A. 1997. Micropropagation of Simmondsia chinensis (Jojoba). Bajaji YPS Biotechnology in Agriculture and Forestry 40[M]. Heidelberg, Berlin: Springer-Verlag, 371-393.
- Sardana Jyoti, Batra Amla. 1998. In vitro regeneration of jojoba (Simmondsia chinensis); A plant of high potential[]]. Advances in Plant Sciences, 11, 143-146.
- Roussos PA, Tolia Marioli A, Pontikis CA, et al. 1999. Rapid multiplication of jojoba seedlings by in vitro culture[J]. Plant Cell Tissue and Organ Culture, 57: 133-137.
- Shallanon H, Maziere Y. 1992. Influence of growth room and vesselhumidity on the in vitro development of rose plants []]. Plant Cell Tiss Org Cult, 30: 121-125.
- Vermaut Sabien, Busselen Paul, Spencer Stuart. 1995. Gallbladder contractions in chickens, guinea pigs and mice following Treatment with Simmondsis or Cholecystokinin[J]. Belgian Journal of Zoology, 125(1): 251-259.
- Yermanos DM. 1977. Jojoba-genetically controlled botanical traits[]]. J Am Oil Chem Soc, 54: 545-548.
- Ziv M. 1986. Plant tissue culture and its agricultural applications[M]. London: Butterworths, 187-196.
- Ziv M. 1991. Micropropagation[M]. Dordrecht: Kluwer Academic Publishers, 45-69.