Stomatal Characteristics of Jojoba, *Simmondsia chinensis* (Link) Schneider

D. Glat, A. K. Dobrenz and D. Palzkill
Department of Plant Science, University of Arizona

Introduction

Jojoba (*Simmondsia chinensis* (Link) Schneider) is an evergreen desert shrub native to the Sonoran Desert of the Southwestern United States and northwest Mexico. Its seeds contain a liquid wax which is unique in the plant kingdom and can replace whale sperm oil in many industrial processes. Presently, jojoba oil is being utilized by the cosmetic industry in hair oil, shampoo, soap, and other products. It can also be used in lubricants for high speed, high temperature machines, in polishing waxes, plasticizers, and in other industrial products.

Jojoba is a true xerophyte, which can stand temperatures as high as 115°-120°F and very low levels of water in soil without signs of major injury (Al-Ani, 1972). Being a drought resistant species which produces a valuable product jojoba can possibly become an important crop for arid and semiarid regions of the world where very few crops can be grown profitably.

The most stressful factor limiting plant growth and development in the desert is the shortage of water. Thus a major dilemma that desert adapted species have to face is how to reduce the amount of water vapor lost from the leaves (transpiration) to a minimum while still maintaining some CO_2 uptake (photosynthesis) required for the initial synthesis of all carbon compounds. The vast majority (95%) of the CO_2 and water vapor exchange that occurs between plants and the atmosphere is through small orifices present on leaf surfaces called stomata (stoma, singular). Stomata are openings that appear on the leaf epidermis between two specialized cells called guard cells, when these are filled with water. Thus, stomata opening and closing are primarily dependent on the flow of water to guard cells and on the general water status of the plants. In the absence of water stress, however, stomata follow a diurnal pattern of opening during the day and closing at night. An interesting exception is the behavior of some succulent desert species, which present the cransulacean acid metabolism pathway of photosynthesis. Those plants, contrarily of all others keep stomata closed during the day to avoid any water loss, and open them at night to uptake CO_2.

Desert-adapted species present several other morphological and physiological features that enable them to cope with the adverse environmental conditions of their natural habitat. Some of these features are small leaf size, leaf hairs, thick leaf cuticle, long roots, photosynthesizing stems, drought deciduous leaves, and short life cycle. Some stomatal characteristics are also important to the adaptations of plants to the desert. Most plants have their stomata level or slightly above the leaf surface while some desert adapted species present their stomata recessed, or sunken below the leaf surface. Also, stomata of desert plants are usually smaller than those of most mesophytic plants (Zelich, 1963).

Other stomatal characteristics which influence the water budget of plants, and which vary considerably between species, are stomata distribution over leaf surfaces, and stomata density; for example, some dicotyledon trees have stomata only on the lower surface, while water lily and other aquatic plants have stomata on the upper surface exclusively (Northern, 1968).

Stomata densities between species range from less than 20 to more than 1000 stomata per mm^2 of leaf surface (Meidner, 1968). Stomata density also varies considerably between individual plants of the same species, among leaves of the same plant, and even among different parts of the same leaf surface (Cole, 1970). Other factors being constant, the diffusion of CO_2 in, and water vapor out of the leaves are directly related to stomata density and distribution (Bidwell, 1979). The movement of these gases in and out of leaves is a function of the difference in their concentrations between the atmosphere and the interior of leaves. The water vapor gradient present between these sites is much steeper than that of CO_2; also, the diffusion coefficient for water is higher than for CO_2. Thus a reduction in stomata conductance caused by a decrease in stomata density, would decrease water loss proportionally more than it would decrease CO_2 uptake. This could definitely assist on the water-saving adaptations of drought-tolerant species.

In this present study we decided to investigate the differences in stomata density among individual plants of jojoba and among jojoba clones—groups of genetically identical plants which were produced from a single plant by vegetative propagation. Differences between leaf surface were also studied. Electron microscopy photographies were used to characterize the stomata distribution pattern, shape, size, and any unusual features associated with stomata of jojoba.

Materials and Methods

For the clonal study, 10-15 cm long terminal stem cuttings were taken in 1979 from jojoba plants growing in the wild, and rooted using a mist propagation system. After rooting they were potted in 1 liter black polyethylene containers using a media of loam, pinebark, perlite and vermiculite in the rate of 4:2:1:1. The rooted cuttings were grown for six months in a double-polyethylene inflated greenhouse in which temperatures ranged from 22 to 30°C. At the end of this growing period, at least 10 new leaves (5 nodes) had formed on each cutting.

Cellulose acetate was used to take leaf impressions for stomata counting. Leaves were dipped in distilled water and blotted dry, then cellulose acetate was spread on the central portion of the leaf to one side of the midrib. After drying and hardening for 3 to 5 min, the acetate was peeled from the leaf using tweezers, placed on a glass slide and covered with a cover slip. Seven microscopic fields (0.135 mm², 450X) were counted on each impression, and the mean of these counts was used for statistical analysis. Counts were made on upper and lower surfaces of fully expanded leaves (3rd node from apex) of 5 plants for each of 5 clones.

Three month old jojoba seedlings grown in the same soil media described above were also used to evaluate differences in stomata density between leaf surfaces, and individual plants. Photomicrographs from upper and lower surface of a 4th node leaf of a seedling were taken with scanning electron microscopy (E.T.E.C. Auto Scan).

Results and Discussion

Jojoba stomata, as observed on S.E.M., present characteristics that favor water conservation. Guard cells at both surfaces are sunken below the epidermal layer (Figure 1). A protective collar of cuticular material surrounding the stomata is also evident on Figure 1. This protective collar may be what was referred to as a cuticular dome by Yermanos in an earlier report (Yermanos, 1967). Figure 1b and 1c are from the lower and upper surface of a fully expanded leaf, respectively. The white thread-like substance observed on Figure 1b is an intricate wax deposition which seems to be absent on the upper surface (Figure 1C). This may be another adaptation of jojoba to drought conditions. It is known that when a plant is subjected to water stress, the stomata on upper surfaces tend to close first, with those on the lower surface remaining opened longer (Meidner, 1968). Thus, this wax deposition on the lower leaf surface could have a "counteracting" effect, to diminish water loss from transpiration through the lower surface when water stress is developing. This wax deposition could also serve as a mechanism to increase energy dissipation from leaf surface (cooling off effect), what would be specially desirable in a hot, arid environment.

Table 1. Stomatal density of upper and lower leaf surfaces of leaves from the third node from the apex of 5 jojoba clones.

Clone	Upper	Lower	Mean
		Stomata mm²	
A	115[z]	131	123a
B	98	117	108ab
C	95	123	109ab
D	84	126	105 b
E	82	97	90 b
Mean	95[a]	119[b]	107

[z]Each value is the mean of 5 replicates. Mean separation within columns by Student-newman Keul's Test, 1% level.

Stomata resistance, which is an important factor controlling gas exchange, is a combination of the stomatal pore area and stomata density. From the pictures obtained, we estimate the jojoba stoma area to be around 60-90 u² (9-10 u long and 2-3 u wide) depending on the degree of opening. Assuming mean stomata density to be 107 stomata/mm² of leaf surface, we calculate total pore area of jojoba leaves to be around .8% of total leaf area. This is close to values reported for other crops—around 1% —(Zelitch, 1963); therefore, total pore area doesn't seem to be a major factor influencing the drought tolerance of jojoba.

Significant differences were found in stomata density between 5 different clones from Arizona's native stand (Table 1). There was a 30% difference in stomata density between the highest and lowest clonal means. The magnitude of this difference could have important effects on gas exchange of these clones.

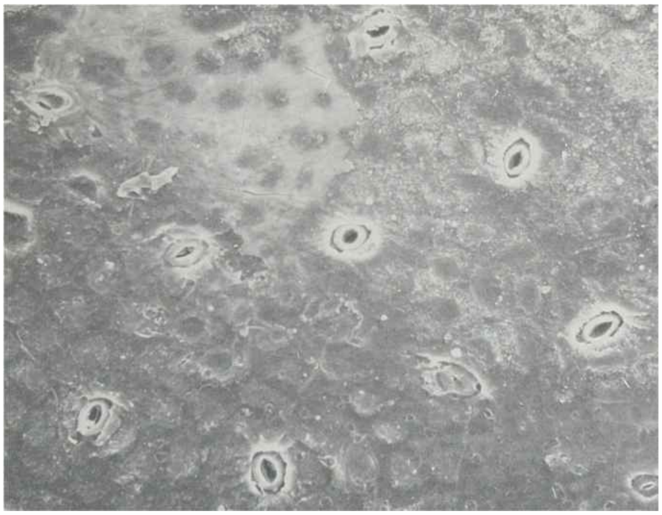
The average stomata density for all leaf surface measured on the clonal study was 107 stomata/mm². This value is not much different than other major crops. It is lower than the average of several alfalfa varieties—around 200 stomata/mm² (Cole and Dobrenz, 1970). It is much higher than that of tomatoes—12-13 stomata/mm² (Northern, 1968) and similar to corn—100-200 stomata/mm² (Zelitch, 1963).

A wide range in stomata density was found between 10 jojoba seedlings (Table 2). The highest individual stomata density mean is 70% higher than that of the lowest. Since these plants were grown in very similar environmental conditions, we believe that these differences are caused mainly by genetic factors.

Significant differences in stomata density were also found between lower and upper leaf surfaces among the plants of the 5 clones. On the average, the lower surface had 119 stomata/mm² while the upper surface had 95 stomata/mm². When 3 month old jojoba seedlings were evaluated for stomata density on both leaf surface, we also found the lower surface to present significantly more stomata (Table 2). Several other crops have been reported to present more stomata on the lower leaf surface also; among them we could cite, alfalfa, beans, corn, tomatoes, and pumpkins (Northern, 1968; Cole and Dobrenz, 1970).

Conclusions

Jojoba is dioecious (male and female flowers in separated plants), and usually pollinated by wind. Therefore, populations in the wild display great genetic variability. Experiments at the University of Arizona indicate that jojoba can



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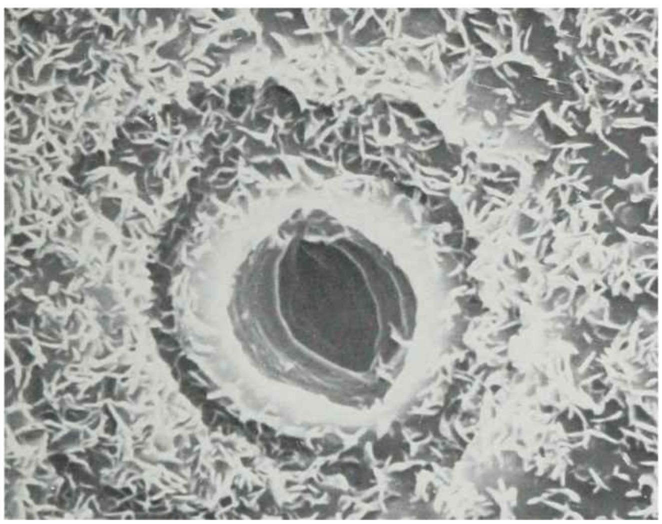

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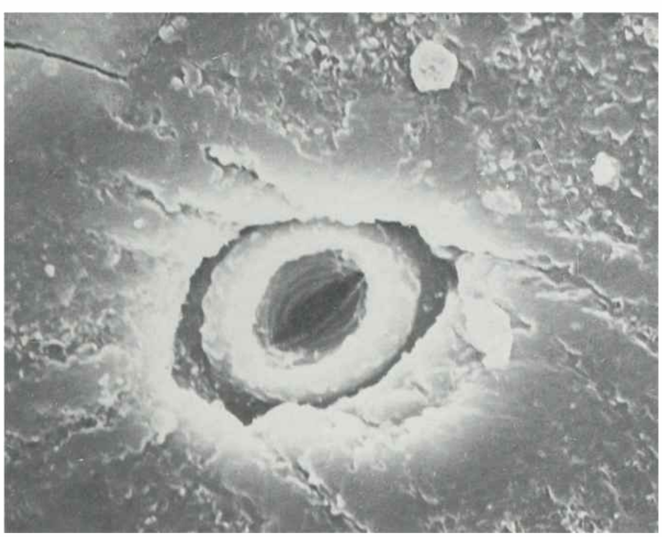

C 0 10.0 u

Figure 1. *Electron micrographs from Jojoba stomata on the upper (1a and 1c) and lower surface (1b), of a fully expanded leaf.*

Table 2. Stomatal density of upper and lower leaf surfaces of 10 jojoba seedlings.

Seedling	Upper Surface	Lower Surface	Mean
		Stomata mm[2]	
1	67[y]	84	75
2	63	82	72
3	84	99	91
4	72	85	78
5	101	110	106
6	87	87	87
7	83	92	87
8	91	102	96
9	88	91	89
10	60	69	62
Mean	79	89	94

[y]Mean value in the mean of 21 separate counts on each of 3 leaves.
[x]Means of upper surfaces was significantly less than that of adaxial surfaces of the 5% level by the Student Newman Keul Test.

be successfully propagated by cuttings, which allows for the development of clones. Desirable characteristics can thus be perpetuated by vegetative propagation of plants presenting such characteristics. This procedure is especially important for jojoba because taking 3–4 years to produce seeds, it could take many years before actually improved jojoba varieties could be developed by modern breeding techniques. Results of this study show that there are significant differences in stomata density between jojoba clones and individual seedlings. Other researchers have suggested that stomata density would be an important criterion for producing high-yielding wheats (Teare, 1971) or to influence yields of triticale under certain environmental conditions (Sapra, 1975). Further studies are necessary to determine if a decrease in stomata density would indeed increase yields or water use efficiency of jojoba. If either of those factors can be determined experimentally then selection of jojoba clones with low stomata density could result in plants still better adapted to low-water agriculture.

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