Microscopic Studies on Epidermal Cells and Stomatal Behavior of Some Globular Cacti (Mammillaria spp.)

¹Vinita Janu and ²R.K. Raghuvanshi ¹Centre for Rural Development and Technology, IIT Delhi, New Delhi, India ²Department of Botany, University of Rajashan, Jaipur-302004, India

Abstract: Background: The objective of this work was to study of epidermal cells and stomatal behavior (during day and night) of *Mammillaria*. The genus *Mammillaria* is most popular and morphologically variable genus in the Cactaceae. Genus distributed in all over the world, plants are specially cultivated in gardens as ornamental spp. and xerophyteic in nature called globular cacti. Results and Conclusion: Many epidermal character like number of epidermal cells (per microfield), size of epidermal cells, number of stomata (per microfield), size of stomata (during day and night), number of crystals and stomatal index were studied in five species of *Mammillaria* i.e., *Mammillaria baumii*, *M. comptotricha*, *M. rekoi*, *M. prolifera* and *M. magnimamma*. All selected species has very interesting stomatal morphology and differ from each other in many characters.

Key words: Epidermal cells, globular, Mammillaria, stomata, Cacti

INTRODUCTION

The genus Mammillaria is one of the largest in the cactus family (Cactaceae), with currently 171 known species and varieties recognized. Cacti are most popular species from arid and semi-arid regions (Pereira et al., 2009). America is the center of origin of cacti plants (Avina-Padilla et al., 2009) most of the Mammillarias are native to Mexico, but some come from the southwest USA, the Caribbean, Colombia, Venezuela, Guatemala and Honduras (Bergaoui et al., 2007). Members of the genus Mammillaria are low-growing, globular (Mihalte et al., 2010) cacti with distinctly tuberculate stem morphology. Plants may either be solitary or form massive mounds. These traits are shared with other members of the Mammilloid clade (Butterworth et al., 2002). The leaves were also consumed as vegetables by some people. Some species of cactus have anti-cancer, anti-tumor, anti-rheumatic, anti-ulcer and anti-inflammatory properties (Sri Nurestri et al., 2008). They are also used as remedy for the relief of gastric pain and for revitalizing the body (Goh, 2000).

Most plants open their stomata during the day in order to allow carbon dioxide to diffuse into the leaf and with water to function immediately as a substrate for photosynthesis. Inevitably, this also allows water saturated air surrounding the photosynthic tissue to diffuse outwards; at night, when photosynthesis ceases, the stomata close in order to stop the loss of water (Gibson and Nobel, 1986a, b; Mauseth, 1984; Sajeva and

Mauseth, 1991). CAM plants have a special mechanism that allow them to take in carbon dioxide at night and hold it in organic acids; this causes the pH to fall at night (hence the name acid metabolism) (Goldstein et al., 1991). During the daytime, the carbon dioxide is released from the organic acids and feeds into photosynthesis; stomata remain closed. Opening the stomata at night, rather than during the day results in much less loss of water because of lower air and tissue temperature. It has been calculated that cacti lose 1/6000th as much water as a mesophyte of the same surface area, by such strategies (Benson, 1982). Cacti commonly lose 60% of their water before significant physiological stress sets in. Most mesophytes, in contrast will wilt and die with a 20% loss of their water (Benson, 1982). This results in high WUE (Water Use Efficiency). WUE refers to the amount of water lost by transpiration relative to the amount of carbon dioxide taken up and fixed into organic compounds by photosynthesis (Sajeva and Mauseth, 1991).

Present study deals with microscopic study of epidermal cells, size of cactus stomata (during day and night) and stomatal Index.

MATERIALS AND METHODS

For the epidermal studies of *Mammillaria*, the fresh tubercles were used. Some portions of fresh tubercles were peeled off. The peel was removed with the help of a pair of forceps. Later on, they were stained with dilute safranin and mounted in dilute glycerine. The data on

number of epidermal cells, number of stomata per microfield and stomatal index were calculated at X150 magnification. The size of epidermal cells and stomata was determined using ocular micrometer and stage. The data on size of epidermal cells and stomata were carried out at X675 magnification in the compound microscope. The measurable data were presented as Arithmetic Mean (X) and Standard Error (SE).

Stomatal index: The stomatal index was calculated by the following formula:

$$I = \left(\frac{S}{E+S}\right) \times 100$$

Where:

I = Stomatal index

S = No. of stomata per unit area

E = No. of epidermal cells per unit area

RESULTS

Many epidermal character like number of epidermal cells (per microfield), size of epidermal cells, number of stomata (per microfield), size of stomata (during day and night), number of crystals and Stomatal Index were studied in five species of *Mammillaria* i.e., *Mammillaria baumii*, *M. comptotricha*, *M. rekoi*, *M. prolifera* and *M. magnimamma* (Table 1).

Epidermal characters

Shape of epidermal cells: In *M. baumii*, *M. rekoi*, *M. prolifera* and *M. magnimamma*, the epidermal cells were sinuous (Fig. 1a, c, d, e). However, in *M. comptotricha* it was straight walled (Fig. 1b). Mucilage substance has been observed in epidermal cells of *Mammillaria* species (Fig. 1). In *Mammillaria* species crystals were totally absent.

Table 1: Comparative study of epidermal and stomatal characters of Mammillaria (spp.)

Epidermal character	M. baumii ±SE	M comptotricha ±SE	M. rekoi ±SE	M prolifera ±SE	M magnimamma ±SE
No. of epidermal cells*	152±12.385	148±9.127	182±6.794	154±2.136	195±11.804
Size of epidermal cells (μm)	82.42×56.77	150.48×119.01	146.72×114.91	120.38×88.92	111.83×89.60
	±6.053×3.496	$\pm 10.898 \times 5.923$	±4.678×4.647	±5.553×4.191	±5.928×5.711
No. of stomata*	23±1.140	15±0.849	23±0.532	13±0.584	24±0.529
Size of stomata (µm) during day	35.57×20.18	38.99×26.68	37.96×27.70	51.98×34.2	39.33×25.31
	$\pm 0.530 \times 1.021$	$\pm 1.104 \times 1.060$	$\pm 0.583 \times 0.758$	$\pm 0.810 \times 0.684$	$\pm 0.541 \times 0.530$
Size of stomata (µm) during night	34.27×29.07	35.23×29.41	36.94×32.83	48.91×37.96	37.96×32.49
	$\pm 0.865 \times 0.872$	$\pm 0.693 \times 0.866$	±0.649×0.530	$\pm 0.693 \times 0.583$	$\pm 0.899 \times 0.726$
Stomatal index	13.142	9.202	11.219	7.784	10.958
Crystals	Absent	Absent	Absent	Absent	Absent

^{*}Per micr- field

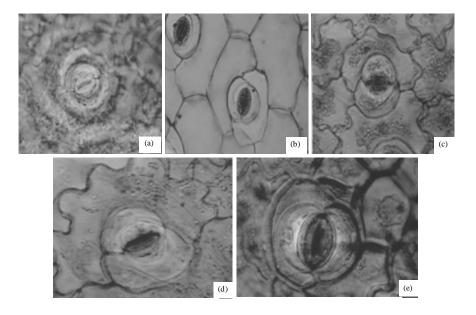


Fig. 1: Study of stomata in (a) Mammillaria baumii, (b) M. comptotricha, (c) M. rekoi, (d) M. prolifera and (e) M. magnimamma

Number of epidermal cells: In *M. baumii*, number of epidermal cells/microfield was observed as 152, in *M. comptotricha*, 148 in *M. rekoi* as 182 in *M. prolifera* and *M. magnimamma*, number of epidermal cells/microfield was observed as 154 and 195, respectively.

Size of epidermal cells: Length and breadth of epidermal cells in *M. baumii*, were 82.42 and 56.77 µm respectively. There were some differences in the size of epidermal cells in *M. comptotricha* and *M. rekoi*. However, in *M. prolifera* and *M. magnimamma* the size of epidermal cells was almost similar.

Stomata: Stomata in *Mammillaria* species open during night.

Number of stomata: Number of stomata / microfield was in *M. baumii* and *M. comptotricha*, was 23 and 15, in *M. rekoi*, *M. prolifera* and *M. magnimamma*, number of stomata was observed as 23, 13 and 24, respectively.

Size of stomata: During the day, size (L X B) of stomata in *M. baumii*, *M. comptotricha*, was 35.57×20.18 μm and 38.99×26.68 μm, However, there was some variation in size of stomata during night in above said five species of *Mammillaria* (Table 1, Fig. 1a-e).

Stomatal index: However, there were little variations in stomatal index in above said five species of *Mammillaria* (Table 1).

Crystals: Crystals were absent in above said five species of *Mammillaria*.

DISCUSSION

Detailed epidermal studies were carried out in *M. baumii*, *M. comptotricha*, *M. rekoi*, *M. prolifera* and *M. magnimamma*. Many epidermal characters like number of epidermal cells, size of epidermal cells, nature of epidermal cells, number of stomata, stomatal index and size of stomata were studied in these species of cacti. Stomata in these cacti were nocturnal in nature. Each stomata has a pair of kidney shape guard cells. They contain single nucleus and many chloroplasts.

In *M. magnimamma*, the number of epidermal cells per micro field was found as 195±11.804. However in other four species investigated, its number was reduced. In *M. baumii*, the epidermal cells were small sized whereas in *M. comptrotricha* it was of large size. In *Mammillaria* species, crystals were totally absent in their epidermal cells. In *Mammillaria* species, number of stomata per

microfield and stomatal index differed invariably. The size of stomata during both day and night did not show many variations in *Mammillaria* species investigated except *M. prolifera* where they were of large size.

In *M. baumii*, *M. rekoi*, *M. prolifera* and *M. magnimamma*, the epidermal cells were sinous. However in *M. comptrotricha*, it was straight-walled. In some epidermal cells of *Mammillaria* species, mucilage substance has been observed. Where as the size of stomata was concerned there were some variations within species of *Mammillaria*.

According to Ting (1985) and Nobel (1988) cacti have Nocturnal stomatal opening.

Cacti in general, have fewer stomata per unit area than do most other plants. During severe water stress stomata of cacti may remain closed throughout both day and night. Stomata closure eliminates not only water loss but also CO₂ exchange. The plants will increase acidity at night due to respiration. CO₂ is first fixed as malate, then decarboxylated and moved into the chlorenchyma to make glucans. Furthermore, there is still a reduction in acidity during the day due to assimilation of CO₂. This pattern of no net CO₂ uptake but continued day-night oscillations in acidity has been termed idling (Gibson and Nobel, 1986a,b; Mauseth, 1984).

CONCLUSIONS

Many epidermal characters like number of epidermal cells, stomatal index, size of stomata and occurrence of crystals were studied in these species. Stomata in those cacti are nocturnal in nature. The size of stomata during day and night showed some variations *Mammillaria*. The size of epidermal cells varied invariably in these species of cacti. Crystals were absent in all five species of *Mammillaria*.

In *Mammillaria* species, number of stomata per microfield and stomatal index differed invariably. The size of stomata during both day and night did not show much variation in *Mammillaria* species investigated except *M. prolifera* where they were of large size. In some epidermal cells of *Mammillaria* species, mucilage substance has been observed. Epidermal cells were usually sinuous in all four species investigated except *M. comptrotricha*, where it was straight-walled. Whereas the size of stomata was concerned there were some variations within species of *Mammillaria*.

ACKNOWLEDGMENT

Department of Science and Technology (DST) New Delhi, India.

REFERENCES

- Avina-Padilla, K., F. Parra-Cota, J.C. Ochoa-Sanchez, C. Perales-Segovia and J.P. Martinez-Soriano, 2009. Phytoplasmas associated to diseases of ornamental cacti in Mexico. J. Biol. Sci., 9: 268-271.
- Benson, L., 1982. The Cacti of the United States and Canada. Stanford University Press, Stanford, California.
- Bergaoui, A., N. Boughalleb, H. Ben Jannet, F. Harzallah, M. El-Mahjoub and Z. Mighri, 2007. Chemical composition and antifungal activity of volatiles from three *Opuntia* species growing in Tunisia. Pak. J. Biol. Sci., 10: 2485-2489.
- Butterworth, C.A., J.H. Cota and R.S. Wallace, 2002. Molecular systematics of tribe cacteae (Cactaceae: Cactoideae): A phylogeny based on rpl16 intron sequence variation. Syst. Bot., 27: 257-270.
- Gibson, A.C. and P.S. Nobel, 1986a. Gas Exchange and Crassulacean Acid Metabolism. In: The Cactus Primer. Harvard University Press, Cambridge MA.
- Gibson, A.C. and P.S. Nobel, 1986b. The Cactus Primer. Harvard University Press, Cambridge.
- Goh, K.L., 2000. Malaysian Herbaceous Plants Millennium. 1st Edn., Advanco Press, Malaysia.
- Goldstein, G., K.E. Ortega, N. Nerd and P.S. Nobel, 1991. Diel pattern of water potential components for crassulacean acid metabolism plant *Opuntia ficusindica* when well-watered or droughted. Plant Physiol. Rockville, Md.: Am. Soc. Plant Physiol. Bull., 95: 274-280.

- Mauseth, J.D., 1984. Introduction to cactus anatomy Part 7. Epidermis. Cact. Succ. J., 56: 33-37.
- Mihalte, L., G. Feszt, A. Baciu and A. Vilcan, 2010. Phylogenetic distances among several genotypes of Rebutia, Mediolobivia and Sulcorebutia (Cactaceae). Int. J. Bot., 6: 266-272.
- Nobel, P.S., 1988. Environmental Biology of Agaves and Cacti. Cambridge University Press, New York.
- Pereira, C.A.P., M.J.M. Alvarez, C.A.M. Martinez and D.M.G. Pantaleon, 2009. Obtention of enzymatically hydrolyzed product from cactus (*Opuntia boldinghii* Britton and Rose) cladodes whole flour. Pak. J. Nutr., 8: 459-468.
- Sajeva, M. and J.D. Mauseth, 1991. Leaf like structure in the photosynthetic succulent stems of cacti. Ann. Bot., 68: 405-411.
- Sri Nurestri, A.M., A.W. Norhanom, Y. Hashim, K.S. Sim, S.L. Hong, G.S. Lee and N.S.A.R. Syarifah, 2008. Cytotoxic Activity of *Pereskia bleo* (Cactaceae) against selected human cell lines. Int. J. Cancer Res., 4: 20-27.
- Ting, L.P., 1985. Crassulacean acid metabolism. Ann. Rev. Plant Physiol., 36: 395-622.