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Leaf anatomical adaptation of some spontaneous species of the Sahara (El Oued region, Algeria)

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Abstract

This study aimed to show the correlation between anatomical and physiological response and the adaptation in the event of drought and salinity of six Saharan spontaneous species in the region of El Oued (Algeria) these plants are Zygophyllum *album L, Limoniastrum guyonianum, Sueda fructicosa, Anabasis articulate, Thymelea microphylla* and *Halocnemumstrobilaceum*. The anatomical structure of this species was studied using fresh materials (roots, stems and leaves) by free-handing transverse directions. The sections were stained by double staining with methyl green and Congo red. The epidermis of the studied plants represents three types of stomata that are the paratetracytic, the anomocyticand the Paracytic type.The results showed that the plants studied adapted with the stress to underline the different strategies by the emergence of distinct anatomical structures which appear by the presence of a thick cuticle and a layer of trichomes, the abundance of the palissadic parenchymes and aquifers, the stomata are placed in the crypts. Two species contain calcium oxalate crystals *L. guyonianum* and *A. articulata*.

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Introduction

The Sahara is characterized by climatic conditions very constraining to the spontaneous survival of living beings. Although the very strict and very challenging environmental conditions, there are still geomorphological areas offering more or less favorable conditions for the survival and proliferation of a spontaneous Saharan flora characteristic and adapted to the harsh climatic conditions of this desert environment (Ozenda, 1983).Spontaneous plants are plant species that grow naturally in the wild, without the intervention of humans (Ozenda, 1977; Benkhetou, 2010; Benchelah *et al.,* 2011). In Algeria, the pedoclimatic characteristics favor in many regions processes of salinization of the lands very constraining for the farmer (Kessira, 2002; Ziani, 2003). Despite the hostility of the Saharan conditions, an impressive plant cover still remains thanks to morphological, physiological and anatomical adaptation mechanisms. Indeed, the spontaneous perennial plants use very specific anatomical mechanisms to stabilize their lifestyle in the Saharan region. To fight against the lack of water, the plants develop more adaptive strategies that vary according to the species and environmental conditions (Turner, 1986).The main characteristic of halophytes is to have a living material that can function actively in the presence of high salt concentrations, which is the essential aspect of their salt resistance (Hopkins, 2003).

This living matter is essentially represented by soluble carbohydrates, mainly glucose, fructose and sucrose, also alcohols and organic acids, which seem to play a very important role in the maintenance of a turgor pressure which is at the basis of various processes controlling plant life (Hasegawa *et al.,* 2000; Rodriguez-Rosales *et al.,*1999). Generally, the adaptations of the desert plants relate to the reduction of the leaf surface, the decrease of the speed of evaporation and the constitution of reserve of water inside the tissues (Ozenda, 1977). The present work aims to give an idea of the physiological and morphoanatomical adaptation mechanisms of perennial spontaneous plants.

Material and methods

Presentation of the area studied

The El Hamraia area is located northeast of the province of El Oued (desert area). Geographically, it lies between the two lines of longitude 6.23 05 78 4 east and latitude 34, 1109518. The Hamraia area has many types of soils, gypsum, salt, sand and clay, containing natural and agricultural resources from the saline wealth at Chott Melghir.

The coldest months are December and January (10-11°C). The warmest months are July and October (31-34°C), the wettest month is January, about 22 mm, and the driest month is July (about 0.03 mm).

Plant materials

Six plants growing naturally in the El Oued region in the arid desert and near the Chott were selected; these plants are Zygophyllum *album L, Limoniastrum guyonianum, Sueda fructicosa, Anabasis articulate, Thymelea microphylla* and *Halocnemumstrobilaceum*.

Anatomical study

The fresh plants were gently collected from natural areas; roots were separated and fixed in alcohol 70% before cutting with razor blade. For the anatomical description the material was sectioned free-hand, in transverse directions. Sections were emptied in bleach (sodium hypochlorite 12 °Cl) for 20min and then rinsed with distilled water and then treated with 1% acetic acid for 1mn. The sections were stained by double staining with methyl green for 5min and Congo red for 20mn in agreement with usual techniques in plant anatomy (Langeron., 1949).

Results

Zygophyllum album

After microscopic observation of the epidermis of the *Z. album* leaf, the stomatal type of this plant is anomocytic type. The cross-section of the *Z. album* leaf from the outside to the inside shows that the epidermis is a tissue formed by a single cellular base, covered by thick cuticle. The mesophyll is non-homogeneous, it includes (Fig.1). a parenchyma palissadic located under the epidermis, it consists of a layer of elongated cells rich in chloroplasts and an aquifer parenchyma of large cells;the cerebro-vascular vessels arranged on a single cycle, consisting of the secondary veins; and the cerebro-vascular vessels arranged in the center, constitutingthe main vein.

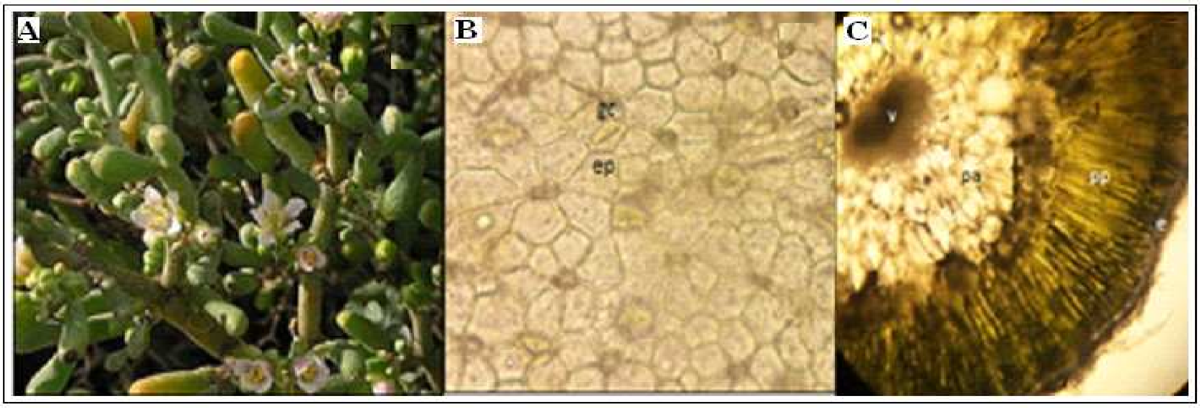

Fig. 1. Leaf of *Z.album* (A),Stomata of the epidermis leaf (B), guards cells (gc), epidermal cells peristomatic (ep). Cross-section of the leaf (C), Cuticle (c), parenchyma palissadic (pp), parenchyma aquifer (pa), vein (v).

Sueda fructicosa

For the epidermis of *S. fruticosa* leaf, there are two stomatologic types, the paratetracytic type and the paracytic type (Fig. 2). The anatomical observation at the *S.fructicosa* leaves shows that the epidermis is a tissue consisting of a single cellular base in oval form. The parenchyma assimilator is organized into two layers of cells, an outer layer of elongated cells and an inner layer of cubic cells; it is located directly under the epidermal base. The aquifer parenchyma is composed of very large cells.

The Crib-vascular vessels in the middle include xylem and phloem superposes.

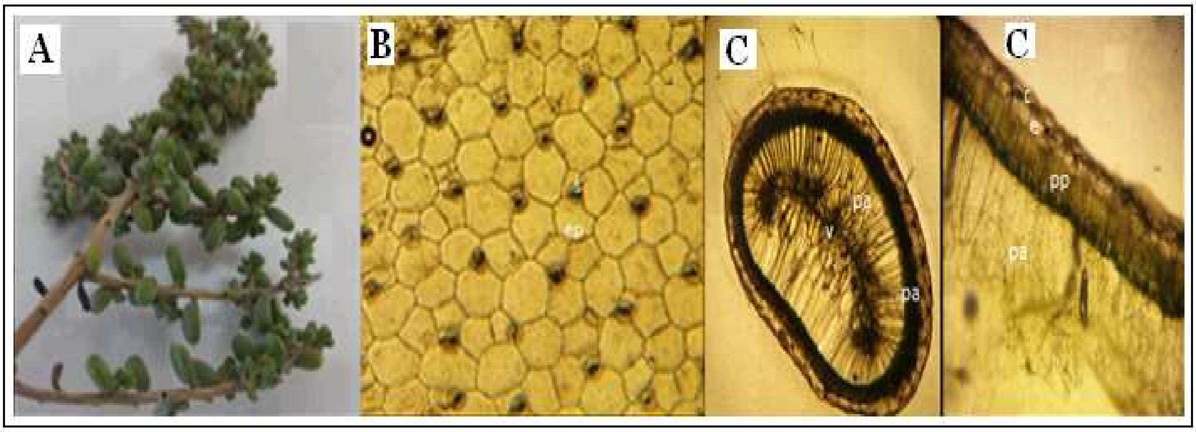

Fig. 2. Leaf of *S. fructicosa*(A), Stomata of the epidermis (B), Stomata (s) , epidermal cells peristomatic (ep), Cross-section of *the S. fructicosa*leaf(C),Cuticle (c), epidermis (e), parenchyma palissadic (pp), parenchyma aquifer (pa), vein (v).

Anabasis articulate

Only one stomatal type was observed on the epidermis of the leaves of the plant of *A.articulata*, the paratetracytic type (Fig. 3). The anatomical observation shows that the epidermis is formed by a single cellular base, with a thick wall and polygonal shape, it covered with a thick cuticle.

Under the epidermis there is a layer of hypodermic cells containing calcium oxalate crystals.

The mesophyll, differentiated into parenchyma assimilator is organized into two layers of cells, an outer layer of elongated cells and an inner layer of cubic cells;it is separated from the epidermis by a hypodermis, also a wide-cell aquifer parenchyma.

Limoniastrum guyonianun

One stomatal type was found on the epidermis of the leaves of the *L.guyonianum* plant, which is the Anomocytic type (Fig. 4).

Thymelea microphylla

A single stomatal type on the epidermis of *Thymelaea microphylla* leaves is the anomocytic type. The trichomes are located throughout extension, which are non-glandular trichome, multicellular, with elongated cells (Fig. 5). The epidermis is a continuous sitting of polygonal cells that covers the leaves; these cells are thickened by the cuticle. The mesophyll is a simple tissue composed by a single class of cells (homogeneous), which is the palisade parenchyma.

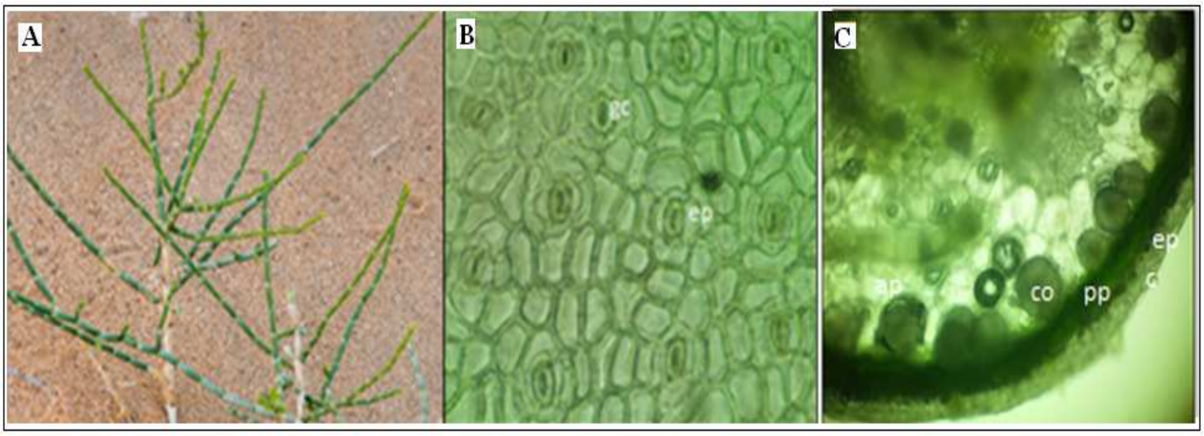

Fig. 3. Leaf *of A.articulata*(A), Stomata of the epidermis (B), guards cells (gc), epidermal cells peristomatic (ep), Saline glands of the leaf of *A.articulata*, cuticle (c), epidermal cells (ep), palissadic parenchyma (pp), Aquifer parenchyma (ap), calcium oxalate crystals (co).

Halocnemum strobilaceum

There are two different stomatal types on the epidermis of the leaves of the plant *H. strobilaceum*, these are: The anomocytic type and the paratetracytic type.

The epidermis is a tissue formed by a single cell base with polygonal shape, which consists of a thick wall covered with a very thick cuticle and stomata (Fig. 6). The non-homogeneous mesophyll comprises a palisadic parenchyma lying on the shape of several layers of cells, consisting of elongate, parallelepipedic cells, tightly packed together and forming continuous beds that do not have intercellular spaces. The aquifer parenchyma consists of large, long cells.

Discussion

Types of stomata

The epidermis of the studied plants *Z. album* (Zygophyllaceae), *S. fructicosa* (Amaranthaceae), *H. strobilaceum* (Amaranthaceae), *L. guyonianum* (Plombaginaceae), *A. articulata* (Amaranthaceae) and *T. microphylla* (Thymeleaceae), represent three types of stomata that are the paratetracytic, the anomocyticand the Paracytic type.

Theanomocytic type appeared in the plants Z.album, *H. strobilaceum*, *L. guyonianum* and *T. microphylla*, for the paratetracytic is *H. strobilaceum*, *S. fructicosa*, and *A. articulata*, finally the only plant that has the paracytic is *S. fructicosa*. According to Kadi-bennane *et al.* (2005) the increase of the paracytic-type frequency depends on the increase of drought, so we dress up that plants that possess paracytic type are the most suitable for drought and salinity. The stomata in all the plants studied are shown to be driven inwards; this causes them to locate deeper inside the organ and is no longer directly exposed to the sun (Hopkins, 2003). According to Boullard, (1997) and Heller *et al.*, (1998) some xerophytic plants adapt to drought, by the deposition of their stomata in crypts, which minimizes the loss of water.

Trichomes

A presence of trichomes on the leaf surface of *T.microphylla* to reduce water loss and maintain leaf moisture, these results are reinforced by many authors who have reported that desert shrub leaves are generally characterized by small, well-developed epidermal hairs covering the leaf surface (Liu *et al.*, 1987). The xerophyte leaves are usually small (Maximov, 1929). This serves not only to minimize the surface on which perspiration can occur but also minimizes heat build-up in the tissues (Smith, 1978).

Dense hairs and spines covering the aerial parts of many desert plants perform a similar function (Juniper and Jeffree, 1983).

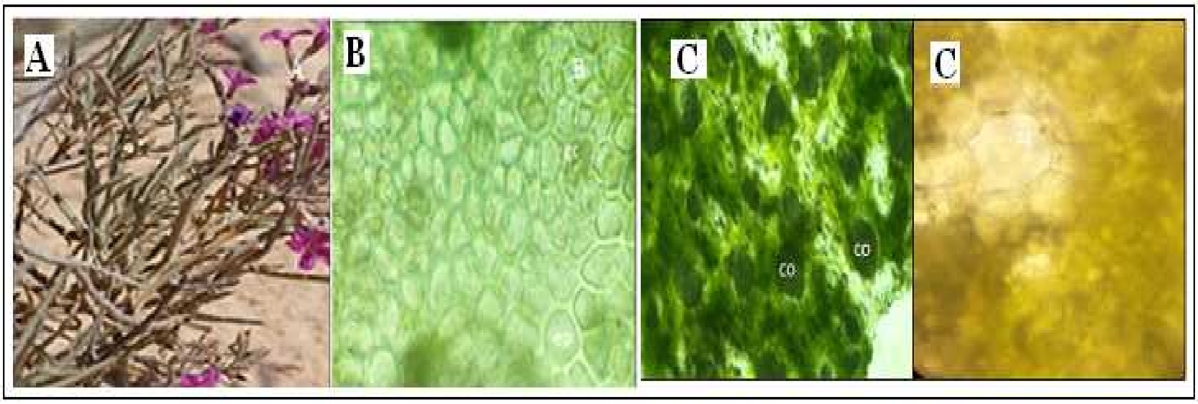

Fig. 4. Leaf of *L.guyonianun* (A), Stomata of the epidermis (B), guards cells (gc), epidermal cells peristomatic (ep), Saline glands of the leaf of *L.guyonianun*(C), calcium oxalate crystals (co) Saline glands (sg).

The glands

Only two species contain calcium oxalate crystals *L. guyonianum* and *A. articulata*, which have the same type of crystals but have a different distribution in *L. guyonianum* crystals were located on the epidermis whereas in *A. articulata* they were localized both around the vascular tissue, in the parenchyma palissadic and the spongy tissue of the mesophyll.

Houari *et al.* (2013) conclude that the cells of the hypoderm of *A. articulata* appear rich in calcium oxalate crystals in the form of twins, which may explain the high calcium content in Saharan plants, which give them great rigidity.The glands are composed of a set of epidermal cells of theplexuses; those who capture salt from the mesophyll cells below them and secrete to the surface of the leaf where a salt layer of the crystals are formed.

The process of salt excretion by the saline gland has yet to be elucidated by some researchers; however, one of the conditions is the availability of the energy that is needed for ionic pumping. In halophytes this energy is provided by the active respiration of the glandular cells (Marcum *et al.*, 1992).

Many plants in desert environments produce different shapes and volumes of crystals in plant tissues. Calcium oxalates are among the most abundant crystals present in different tissue plants and are found in the form of crystalline deposits (Arnott and Pautar, 1970).

According to Thomson (1975), some anatomical modifications appear in different organs. Halophytes have specialized structures, the salt glands, located at the epidermal cells of the leaves and stems and whose function is to excrete salt when the mineral tissue load is excessive such as Na^{+}, Ca^{++}, Mg^{++}, $NO3^{+}$and Cl^{-}.The formation of calcium oxalate is considered a high capacity mechanism for Ca regulation in many plants.

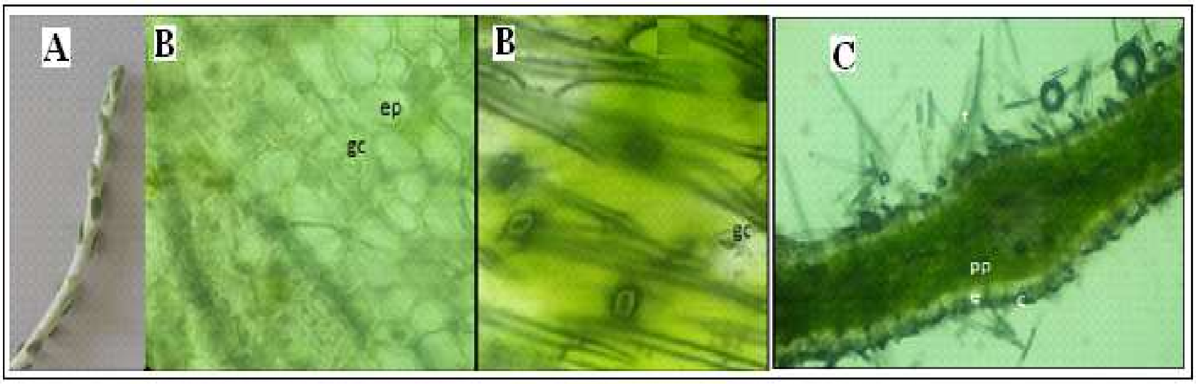

Fig. 5. Leaf of T. *microphylla* (A), Stomata of the epidermis (B), guards cells (gc), epidermal cells peristomatic (ep), Cross section of *T. microphylla* leaf (C), trichome (t), cuticle(c), epidermis(e), palisade parenchyma(pp).

Many halophytic plants have epidermal glands on their leaves and stems that secrete salt (Metcalfe and Chalk., 1950), these glands have been considered effective devices for the secretion of excess salt that accumulates within tissues (Haberlandt, 1914).

The presence of cuticle

The plants studied contain a cuticle of different thickness, the thin cuticle is observed in the leaf of *Z.album* and *A.articulata*, while the other plants have a thick cuticle.

The increase in cuticle thickness decreases transpiration (Fisher and Turner, 1978; Henchi, 1987), but is not the only factor regulating cuticular permeability.

The role of the cuticle is therefore to reduce the evaporation of water directly from the outer surfaces of the leaf epidermis and to protect the underlying epidermal cells and mesophyll cells against desiccation that could be lethal (Hopkins, 2003).

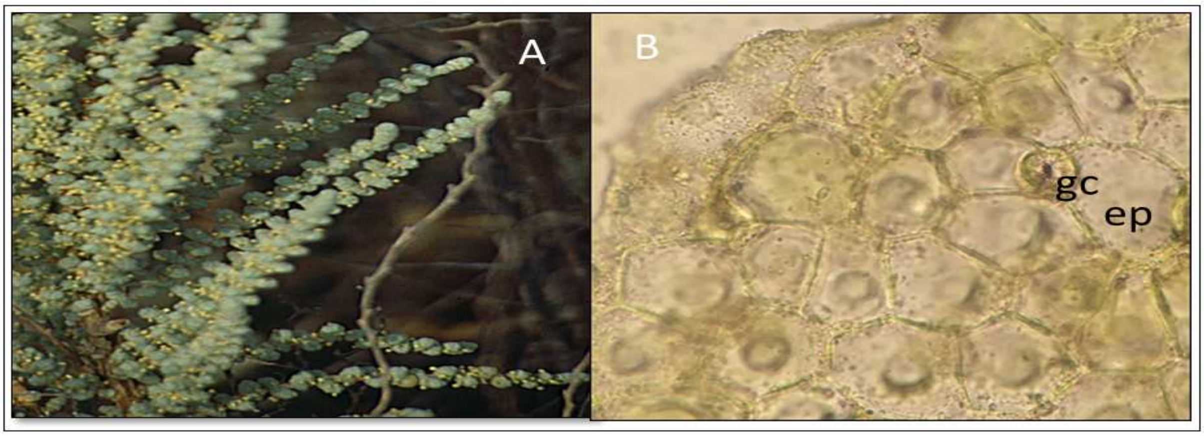

Fig. 6. *H.strobilaceum*(A), Stomata of the epidermis (B), guards cells (gc), epidermal cells peristomatic (ep).

The main physiological roles of the cuticle are related to its hydrophobic nature; it maintains a water-poor area on the surface of the plant which protects the plant from pathogens, on the other hand it limits water losses of the plant.

The formation of a thick cuticle is often associated with the reduction in leaf area (Denden *et al*., 2005); corresponds to the results that plants *H. strobilaceum* and *T. microphylla* have small leaves, at the same time are covered by a thick cuticle.

The presence of the epidermis and the hypodermis

All plants studied contain epidermis covered with *aerial* parts with a single base, except the species *H. strobilaceum* which is in a phase of secondary thickening. The hypodermis found in the species *A. articulata* and *L. guyonianum* removes assimilative parenchyma from the surface of leaves and stems, which accentuates adaptation to a warm and dry environment. According to Lysthed, (1977).

The translocation of water from the inner cell layers to the epidermis is thereby prevented, thus reducing water loss.

The presence of palisade parenchyma

It is observed that the parenchyma assimilator in the plants studied is variable, It can consist of several bases of similar cells such as *Z. album*, *L. guyonianum*, *H. strobilaceum* and *T. microphylla*plants, or only two seated an outer cell elongated and an internal to cubic cells such as *A. articulata* and *S. fructicosa* plants. According to Oguchi *et al.*

(2005), increasing the mesophyll thickness improves photosynthetic capacity if accompanied by an increase in the number of chloroplasts exposed near the surface facing intercellular spacing.

The presence of aquifer parenchyma

The plants that have an aquiferous parenchyma are *Z.album* (Zygophyllaceae), *A. articulata* (Amaranthaceae), *H. strobilaceum* (Amaranthaceae) and *S. fructicosa* (Amaranthaceae), for the other species studied are devoid of aquifer parenchyma.

According to Lemee (1978), Binet and Brunel (1968), the succulents avoid dehydration by the reserve of water in the aquiferous tissues which is associated with a good peripheral protection, these reserves are localized either in the stems or leaves.

Large water storage cells with a sinuous cell wall form the water storage tissue (spongy tissue) in the leaf; it is a common figure of typical xeromorphic leaves.

It improves plant water retention, water absorption and preserves the moist micro environment for surrounding photosynthetic cells (Anwar, 2006).

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