**Leaf physico-chemical and physiological properties of maize (*Zea mays* L.)
populations from different origins**

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ABSTRACT

In this study we evaluated the leaf surface properties of maize populations native to different water availability environments. Leaf surface topography, wettability and gas exchange performance of five maize populations from the Sahara desert, dry (south) and humid (north-western) areas of Spain were analysed. Differences in wettability, stomatal and trichome densities, surface free energy and solubility parameter values were recorded between populations and leaf sides. Leaves from the humid Spanish population with special regard to the abaxial side, were less wettable and less susceptible to polar interactions. The higher wettability and hydrophilicity of Sahara populations with emphasis on the abaxial leaf surfaces, may favour dew deposition and foliar water absorption, hence improving water use efficiency under extremely dry conditions. Compared to the other Saharan populations, the dwarf one had a higher photosynthesis rate suggesting that dwarfism may be a strategy for improving plant tolerance to arid conditions. The results obtained for different maize populations suggest that leaf surfaces may vary in response to drought, but further studies will be required to examine the potential relationship between leaf surface properties and plant stress tolerance.

Keywords:

contact angles
drought stress
gas exchange rates
plant surfaces
surface free energy
wettability
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Running title: leaf surface properties of five maize populations

1. Introduction

Drought is the main limiting factor for crop production (Boyer, 1982; Farooq et al., 2009) since it may limit plant survival, development and geographic distribution (Grace 1997; Liu et al., 2014). Water shortage is expected to occur more frequently with climate change (Intergovernmental Panel on Climate Change, IPCC, 2013), and this may compromise food production to cover societal demands. Understanding the basis of drought tolerance is of paramount importance for releasing varieties with enhanced drought resistance (Takeda and Matsuoka, 2008), and the identification of the variability of characters related to the response to water stress is necessary as a preliminary step.

Maize originated in a tropical environment in Central America more than 8000 years ago, subsequently spreading throughout America and to the rest of the World after the XV Century (Revilla et al., 2003). The dispersion of maize was possible because of the large adaptive ability of this species; maize is currently cultivated from the Equator to the 50° latitude, from 0 to 4000 m above sea level, and from the rainy tropics to the desert. Although this species was introduced in northern Africa less than 500 years ago, maize grown in the oasis of Sahara desert has produced large morphological and genetic variability (Aci et al., 2013). Due to the extremely arid conditions of Sahara desert, maize populations from temperate areas cannot be successfully cultivated there, while maize populations from the desert are tolerant to such harsh environment.

Plant surfaces are important for protecting organs against multiple biotic and abiotic stress factors including the uncontrolled loss of water (Mamrutha et al., 2010; Wu et al., 2014). On the other hand, plant surface water permeability may be a phenomenon of ecophysiological relevance since it may impede or facilitate the absorption of water in the form of, for example, liquid drops deposited on to the surfaces (Oliveira et al., 2005; Fernández et al., 2014a), fog (Limm and Dawson, 2010; Eller et al., 2013; Berry et al., 2014) or dew (Konrad et al., 2014). The surface of organs with primary growth such as leaves, fruits, stems or flowers, is covered with epidermal cells (including structures such as stomata and/or trichomes), the outermost epidermal cell wall layer being the cuticle (Guzmán et al., 2014). The epicuticular wax layer is the most external part of the cuticle and is in direct contact with the surrounding atmosphere (Mamrutha et al., 2010). Epidermal micro- and nano-surface features (Koch and Barthlott, 2009) together with surface chemical composition (Khayet and Fernández, 2012) will determine the

degree of surface wettability and drop adhesion or repellence (Fernández and Khayet, 2015). While the mechanisms of foliar uptake of water and solutes by plant surfaces are still not fully characterised (Fernández and Eichert, 2009; Fernández and Brown, 2013), several studies analysed the interactions of plant surfaces with water (e.g., Brewer et al. 1991; Hanba et al. 2004; Roth-Nebelsick et al. 2012; Rosado and Holder, 2013; Urrego-Pereira et al., 2013a,b; Wang et al., 2014; Koukos et al., 2015). Recent studies (Fernández et al. 2011, 2014a,b; Fernández and Khayet 2015) measured contact angles of three liquids with different degrees of polarity and apolarity as tool for characterising the surface free energy and solubility parameter of a few organs (mostly leaves) and species.

Under humid climates or in sprinkler irrigated areas, water drop repellence of, for example, rainfall or irrigation water, may favour plant water use efficiency, principally by increasing the amount of water that goes into the soil (Holder, 2012; Rosado and Holder, 2013). For species with wettable leaves, water deposited on to the surfaces may decrease photosynthetic rates (Hanba et al., 2004). Working with sprinkler irrigated maize, Urrego-Pereira et al. (2013a) found that photosynthetic rates decreased during the irrigation period. This was associated with its high leaf wettability which ultimately led to a reduction of maize yield (Cavero et al., 2008; Urrego-Pereira et al., 2013b). Consequently, maize cultivars with less wettable leaves may be cultivated for increasing water use efficiency under humid climates and sprinkler-irrigated areas. Under dry conditions (arid and semiarid climates), dew deposited on to plant surfaces can be a substantial water source for plants. The amount of dew water in arid and semiarid climates may range between 10 to 30 mm yr⁻¹ (Duvdevani, 1964; Zangvil, 1996; Malek et al., 1999; Zhang et al., 2015). Moreover, dew may increase plant water use efficiency in the early morning (Ben-Asher et al., 2010). Several studies showed that dew should be taken into account when calculating crop irrigation requirements in arid and semi-arid climates (Moratíel et al., 2013, 2016; Yasutake et al., 2015). Dew formation on to aerial plant organs depends on the wettability of the surfaces (Agam and Berliner, 2006; Fernández et al., 2014a). For maize, values of dew have been reported to range from 0.01 to 0.6 mm per night (Atzema et al., 1990; Kabela et al., 2009; Yasutake et al., 2015). Thus, under dry conditions maize cultivars with increased leaf wettability may be advantageous for improving plant water economy. The potential adaxial and abaxial surface variations between leaves from various cultivars of a crop developed under different eco-physiological conditions have never been analysed so far, either alone or

in relation to water drop interactions. In this preliminary study, we examined for the first time the wettability and physico-chemical features of abaxial and adaxial maize leaf surfaces of cultivars from different climates, ranging from the humid temperate northwest of Spain to three different Sahara desert areas.

2. Materials and methods

2.1. Plant material

Three maize open-pollinated populations representing the geographic diversity of the Saharan desert were included in this characterization, namely PI542687 from the south, PI527470 from the west, and the dwarf population PI527464 from the north of the Algerian Sahara (Table 1). These three populations have similar plant size and growth cycle under the conditions of Pontevedra (northwest of Spain). Two Spanish populations were used for comparison: Cee from Corunna (a province in the northwest of Spain with humid climate), and EPS14(FR)C3 which is an improved synthetic population made from the combination of four populations coming from the dry regions of eastern Spain. The five populations were sown in Pontevedra (northwest of Spain) the 13th of May, 2015. Pontevedra has a mild humid climate with an average annual rainfall of 1600 mm with a mild summer drought.

2.2. Leaf surface traits and physico-chemical properties

For assessing the relationship between leaf surface features and water interactions, several aspects have been measured using maize populations originated in areas of differential aridity (Table 1). For the development of trials, the leaf below the main ear of ten plants per population was cut after female flowering (mid July of 2015). The topography of gold sputtered adaxial (upper) and abaxial (lower) leaf surfaces of the different maize populations was observed by scanning electron microscopy (SEM; Hitachi S-3400 N, Tokyo, Japan). The average number of trichomes and stomata were assessed by image analysis of adaxial and abaxial SEM micrographs (ImageJ 1.45s, W.R., National Institutes of Health, Bethesda, Maryland, USA). For characterizing the physico-chemical properties of adaxial and abaxial maize leaves, advancing contact angles of drops of double-distilled water, glycerol and diiodomethane (both 99% purity, Sigma-Aldrich) were measured at 20 °C with a Drop Shape Analysis System (DSA 100, Krüss, Germany). Approximately 2 µl drops of each liquid were deposited on the adaxial and abaxial surface of 10 maize leaves (40 repetitions per leaf surface) with a manual dosing system holding a 1 ml syringe with a 0.5 mm diameter needle. Side view

images of the drops were captured at a rate of 10 frames s⁻¹. Contact angles were automatically calculated by fitting the captured drop shape to the one calculated from the Young–Laplace equation. For adaxial and abaxial leaf surfaces the total surface free energy (γ), its components (i.e., the Lifshitz-van der Waals (γ_s^{LW}) and acid-base (γ_s^{AB} ; γ^+ and γ^-)), and surface polarity were calculated considering the surface tension components of water ($\gamma_l = 72.80 \text{ mJ m}^{-2}$, $\gamma_l^{LW} = 21.80 \text{ mJ m}^{-2}$, $\gamma_l^+ = \gamma_l^- = 25.50 \text{ mJ m}^{-2}$), glycerol ($\gamma_l = 63.70 \text{ mJ m}^{-2}$, $\gamma_l^{LW} = 33.63 \text{ mJ m}^{-2}$, $\gamma_l^+ = 8.41 \text{ mJ m}^{-2}$, $\gamma_l^- = 31.16 \text{ mJ m}^{-2}$) and diiodomethane ($\gamma_l = \gamma_l^{LW} = 50.80 \text{ mJ m}^{-2}$, $\gamma_l^+ = 0.56 \text{ mJ m}^{-2}$, $\gamma_l^- = 0 \text{ mJ m}^{-2}$) (Fernández and Khayet, 2015).

2.3. Leaf photosynthetic traits

The net photosynthetic, stomatal conductance, and transpiration rates of the leaf below the uppermost ear of ten plants per population were determined with a portable photosynthesis system Li-6400XT (Li-Cor Inc., Lincoln, NE, USA). The relative leaf chlorophyll content (SPAD) was assessed with a hand-held CCM-200 Chlorophyll Content Meter (Opti-Sciences, Tyngsboro, Massachusetts, USA). The quantum efficiency of photosystem II (Φ_{PSII}) was recorded using an OS-30p Chlorophyll Fluorometer (Opti-Sciences, Tyngsboro, Massachusetts, USA) in the leaf below the main ear of ten plants per population.

3. Results and discussion

3.1. Leaf surface traits and physico-chemical properties

Stomata were present on both sides, but trichomes were restricted to the upper leaf side (Fig. 1, Table 2). The density of trichomes on upper leaf sides slightly varied between populations being between 0.4 to 1.5 long trichomes per mm⁻² and 10 to 24 short trichomes per mm⁻². Irrespective of the population origin, higher stomatal densities were always recorded for the lower leaf side, while the widest range of stomatal variations was also recorded in relation to the abaxial leaf surface. Upper leaf stomatal densities ranged between 58 and 75 stomata per mm⁻². The NW population, tolerant to more humid conditions, had the lowest adaxial and abaxial stomatal densities. The highest leaf stomatal densities were observed on South (S) and North (N) Sahara populations, followed by Dry (D) Spain. The stomatal frequencies determined are within the values reported for other maize cultivars (e.g., Driscoll et al., 2008; Zhao et al., 2015). Some studies analysed the variation of leaf stomatal densities in relation to drought stress, and found increased stomatal densities with water shortage in maize (Zhao et al., 2015),

wheat (*Triticum aestivum* L.) (Quarrie and Jones, 1977), black cottonwood (*Populus trichocarpa* Torr. & Gray) (Dunlap and Stettler, 2001), olive (*Olea europea* L.) (Bosabalidis and Kofidis, 2002), or eggplant (*Solanum melongena* L.) (Fu et al., 2013). Stomatal densities decreased in ginger (*Zingiber officinale* Roscoe) (Xu et al., 2003) and *Leymus chinensis* (Trin.) Tzvelev under water deficit (Xu and Zhou, 2008). The possibility of manipulating stomatal densities of crops as tool for reducing stomatal numbers and increasing drought tolerance has been suggested by Hepworth et al. (2015). On the other hand, several authors associated the occurrence of increased leaf stomatal densities with xeromorphic plants (e.g., Esau, 1965; Fahn, 1982). The different maize populations evaluated did not suffer from water restrictions in the experimental plot. On the contrary, it is likely that the Sahara desert populations were not able to perform optimally due to the high air relative humidity and high soil moisture prevailing in the experimental site for most of the growing season. Furthermore, previous reports have shown that maize plants may not thrive when grown in environments with mean temperatures below the ones found in their area of origin (Sánchez et al., 2014).

In Table 3, the contact angles (θ) of water, glycerol and diiodomethane with adaxial and abaxial leaf surfaces of the 5 different populations are shown. All the surfaces were wettable ($\theta < 90^\circ$) for water, glycerol and diiodomethane. The chiefly apolar diiodomethane led to low angles (from 43 to 58°) when in contact with all the surfaces examined. Water θ values were also low and varied between 36 and 75°, with higher angles determined for glycerol (70 to 84°). With the exception of N Sahara cultivar leaves, all lower leaf sides had higher θ values (i.e., are less wettable) with water drops than adaxial leaf sides. All surfaces had a high adhesion for the drops of all liquids (data not shown).

The total surface free energy (γ_s) and surface free energy components of adaxial and abaxial maize leaf surfaces are shown in Table 4. In general, the total surface free energy values obtained are above, and in some instances much higher, than any of the plant materials analysed following the Lifshitz-van der Waals-acid-base method (Fernández et al., 2011, 2014a,b; Khayet and Fernández, 2012; Fernández and Khayet, 2015). While the dispersive (apolar) component (γ_s^{LW}) values of abaxial and adaxial maize leaf surfaces are within the range estimated for adaxial Chilean myrtle (*Luma apiculata* (DC.) Burret) and *Ficus elastica* Roxb. ex Hornem leaf surfaces (Fernández and Khayet, 2015) or the abaxial surface of wheat (Fernández et al., 2014b), the non-

dispersive, acid-base components (γ_s^{AB}) calculated are higher than in previously examined plant surfaces, largely due to the contribution of the electron donor (γ_s^-) component. Both the adaxial and abaxial leaf surfaces of S and N Sahara maize populations had similarly high γ_s values because of the high acid-base components estimated. Dry and NW Spain populations had a higher γ_s on the upper versus the lower leaf side. The lowest γ_s values of all of the maize leaf surfaces examined were estimated for the abaxial leaf side of the NW Spain population.

The solubility parameter (δ) values of maize leaf surfaces are also shown in Table 4. This is a thermodynamic parameter indicating the potential apolar, non-dispersive (polar and other kind of forces) and hydrogen-bonding interaction between molecules. When calculated via measurement of contact angles (Khayet and Fernández, 2012), δ provides insight into the combined effect of surface chemical composition and roughness (Fernández and Khayet, 2015). Solubility parameter results ranged between 24 and 50 $\text{MJ}^{1/2} \text{m}^{-3/2}$, values which are higher than those found in the plant materials analysed so far (Fernández et al., 2011, 214a,b; Khayet and Fernández, 2012; Fernández and Khayet, 2015). Working with model chemical compounds, Khayet and Fernández (2012) calculated values between 16 to 17 $\text{MJ}^{1/2} \text{m}^{-3/2}$ for several epicuticular waxes, and 32 $\text{MJ}^{1/2} \text{m}^{-3/2}$ for polymerised D-glucose and D-galacturonic acid. A plant surface with a low degree roughness should provide δ values somehow within the range calculated for model molecular wax structures (Khayet and Fernández, 2012; Fernández and Khayet, 2015). While the presence of trichomes and/or pubescence may yield plant surfaces extremely unwettable and water repellent as reported for the abaxial side of wheat leaf (Fernández et al., 2014b) or peach (*Prunus persica* (L.) Batsch) fruit (Fernández et al., 2011), the role of the trichomes present in the abaxial leaf side of the maize leaves evaluated concerning water interactions, cannot be clearly ascertained despite they seem to be hydrophilic. All of the upper and lower maize leaf surfaces analysed are wettable and have a high degree of drop adherence. In general, the abaxial leaf side where stomatal densities are higher and trichomes are absent, was less wettable with the exception of the N Sahara dwarf population which had a higher rate of wettability (lower θ) on the lower leaf side.

Wettability of maize leaves has been reported to increase from the juvenile to the adult stage (Salamini, 1963; Bianchi et al., 1985), a phenomenon that has been related to changes in the amount and kind of cuticular waxes (Bianchi et al., 1985; Blaker and Greyson, 1988; Moose and Sisco, 1994). In this study we examined fully-expanded,

healthy leaves of a similar developmental stage and position in the plant for establishing comparisons between different populations. Maize leaf surfaces are covered with waxes whose composition may vary with leaf age and position within the plant (Blaker et al., 1989). The kind of waxes determined in maize leaves are largely long-chain (i.e., with significant apolar components; Khayet and Fernández, 2012) alcohols, esters and acids (Bianchi and Avato, 1984; Ristic and Jenks, 2002). They hence contain functional groups having oxygen and this will contribute to the high γ_s^- values obtained (Khayet and Fernández, 2012). However, the generally high δ , γ_s^- and γ_s^{AB} values recorded for the leaves of this species suggest that such surfaces are prone to non-dispersive interactions (e.g., polar), and that this may be, for instance, due to heterogeneity concerning the development and deposition of waxes and cuticle material over the epidermal cell wall of maize leaves, including guard cells, trichomes remaining epidermal cells. Differences on the chemical composition of abaxial and adaxial leaf epicuticular waxes may arise between populations, but this aspect shall be studied in future investigations.

Foliar water and solute absorption may occur via the cuticle, cuticular irregularities, and epidermal structures such as stomata or trichomes (Fernández and Eichert, 2009; Fernández and Brown, 2013). Leaves from the Spanish populations examined were less wettable on the lower leaf side, where stomatal densities are higher compared to the upper leaf side. This was more marked on the Galician population which may be adapted to the prevailing high moisture conditions in NW Spain. Thus, under frequent rainfall events such as in the environment of origin of Galician cultivars, the occurrence of less wettable leaf surfaces could be a favourable character for limiting potential interferences with gas exchange mechanisms (Hanba et al., 2004). However, under extremely dry conditions, such as the environment of origin of Sahara populations, a higher wettability of the leaf surface will facilitate dew deposition and foliar water uptake to occur, which will result in more water available for the crop.

3.2. *Photosynthetic traits*

Gas exchange and chlorophyll fluorescence parameters have been measured as shown in Table 5. No clear differences can be seen among populations, probably due to the limited drought stress conditions of the experiment. Indeed, the performance of genotypes coming from different areas of the world depends on the adaptation to their native environment. The chlorophyll fluorescence values recorded were within a similar

range for all the cultivars. Despite chlorophyll fluorescence is often used for estimating plant performance under stress (Baker, 2008), its physiological significance may be limited under normal non-stress conditions. Some variability was observed concerning leaf chlorophyll contents, the highest SPAD values being recorded for S and N Sahara cultivars which are tolerant to high light irradiance conditions in Algeria. Plants grown under such conditions generally have low chlorophyll contents, since a reduced number of chlorophyll molecules may be enough for optimal light harvesting, also limiting the risk of photo-damage caused by an excessive light energy absorption which cannot be metabolically used. However, under lower light conditions, higher chlorophyll contents would be required for adequate light capture efficiency, avoiding potential constraints for photosynthesis. This phenomenon has been previously reported in other plant species (e.g. Kitao et al., 2000). Compared to the rest of maize populations analysed, Galician ones had the highest conductance and transpiration values but only medium photosynthetic rates. These results suggest that some degree of plant stress could only be noticed in the NW Spanish population which is adapted to a more humid environment. Photosynthetic parameters are associated with the response to drought which may reduce CO₂ availability due to stomatal closure, limiting photosynthetic efficiency and causing secondary photo-inhibition (Flexas and Medrano, 2002; Chaves and Oliveira, 2004). The dwarf population from the north of Sahara desert had higher leaf chlorophyll contents and also increased photosynthetic, stomatal conductance and transpiration rates compared to the other populations from the desert. This suggests that plant dwarfing is perhaps a mechanism of tolerance to arid conditions. Nonetheless, measurement of gas exchange rates under arid conditions will be necessary for characterising the response mechanisms of these populations and their acclimation capacity.

4. Conclusion

In this preliminary study we characterised for the first time the leaf surface properties of five maize populations from markedly different ecophysiological origins. Differences on the degree of wettability, stomatal and trichomes densities, surface free energy and its components, and solubility parameter, were found between and within leaf sides of plants from the different populations analysed. The leaves from the NW Spanish population which is adapted to the high water availability conditions of Galicia, were less wettable and more apolar (especially the abaxial leaf side), hence having a lower

risk of gas exchange limitations due to the deposition of water. However, the increased wettability of the leaves of cultivars native from the Sahara region may favour dew deposition and foliar water absorption to occur, which may improve plant water use efficiency under extremely dry conditions. It is concluded that further studies will be required to determine the potential relationship between surface properties and drought tolerance in maize.

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Figures

Fig. 1. Adaxial (A, C, E, G, I) and abaxial (B, D, F, H, J) leaf surface SEM micrographs (x50) of the maize populations from: South Sahara (A, B), West Sahara (C, D), North Sahara (E, F), Dry regions of Spain (G, H) and North-west Spain (I, J)

Tables

Table 1

Open-pollinated maize populations used in the study. Average mean temperatures (minimal (Min) and maximal (Max) and precipitation rates in their areas of origin are provided

Population	Origin and nomenclature	Yearly mean temperatures (Min-Max; °C)	Precipitation (mm)
PI542687	South of Sahara (S Sahara)	24.3 (16.0 – 32.5)	43
PI527470	West of Sahara (W Sahara)	23.2 (14.3 – 32.1)	27
PI527464	North of Sahara (dwarf plants; N Sahara)	21.4 (14.1 – 28.6)	64
EPS14(FR)C3	Dry regions of Spain (D Spain)	15.0 (9.5 – 20.4)	317
Cee	Northwestern Spain (NW Spain)	14.4 (11.4 – 17.4)	1008

Table 2

Stomatal and trichome densities on adaxial and abaxial leaf surfaces from 5 different maize populations. Stomata are present on both leaf sides while trichomes were only observed on adaxial leaf surfaces. Data are means $\pm$ standard deviations (SD).

Population	Stomatal densities (N° stomata mm ⁻²)		Long* trichome densities (N° mm ⁻²)	Short** trichome densities (N° mm ⁻²)
	Adaxial	Abaxial	Adaxial	Adaxial
S Sahara	74.5 $\pm$ 9.0	117.8 $\pm$ 14.9	1.5 $\pm$ 0.2	14.2 $\pm$ 3.0
W Sahara	61.8 $\pm$ 4.4	77.4 $\pm$ 11.0	0.4 $\pm$ 0.4	10.4 $\pm$ 3.9
N Sahara	70.4 $\pm$ 9.2	123.1 $\pm$ 14.0	1.0 $\pm$ 0.6	20.7 $\pm$ 5.3
D Spain	70.2 $\pm$ 7.6	96.2 $\pm$ 6.5	0.7 $\pm$ 0.7	23.8 $\pm$ 4.8
NW Spain	57.8 $\pm$ 3.8	85.8 $\pm$ 4.6	0.5 $\pm$ 0.7	16.5 $\pm$ 5.1

*Trichomes longer than approximately 400 μ m

**Trichomes shorter than approximately 100 μ m

Table 3

Contact angles of water (θ_w), glycerol (θ_g) and diiodomethane (θ_d) with adaxial and abaxial leaf surfaces of 5 different maize populations. Data are means $\pm$ SD (n= 40). Within columns, values marked with different letters are significantly different according to Duncan's Multiple Range Test ($P \leq 0.05$)

Population	Leaf side	θ_w (°)	θ_g (°)	θ_d (°)
S Sahara	Adaxial	44.29 $\pm$ 8.56 c	69.42 $\pm$ 6.10 b	46.21 $\pm$ 5.37 c
	Abaxial	48.25 $\pm$ 11.62 c	74.58 $\pm$ 5.92 c,d	45.84 $\pm$ 7.22 c
W Sahara	Adaxial	47.72 $\pm$ 8.83 b	68.64 $\pm$ 8.39 b	43.16 $\pm$ 5.57 d
	Abaxial	53.23 $\pm$ 13.56 c	74.72 $\pm$ 5.32 d	51.56 $\pm$ 5.34 b
N Sahara	Adaxial	49.48 $\pm$ 10.82 b	78.34 $\pm$ 5.96 a	53.68 $\pm$ 6.63 a
	Abaxial	44.29 $\pm$ 8.56 d	78.21 $\pm$ 6.03 b,c	53.83 $\pm$ 4.82 b
D Spain	Adaxial	53.22 $\pm$ 11.45 b	76.54 $\pm$ 5.91 a	47.81 $\pm$ 5.47 c
	Abaxial	63.22 $\pm$ 12.60 b	80.37 $\pm$ 5.51 b	52.77 $\pm$ 5.87 b
NW Spain	Adaxial	63.04 $\pm$ 12.23 a	82.09 $\pm$ 4.93 a	50.59 $\pm$ 5.64 b
	Abaxial	75.35 $\pm$ 12.54 a	84.19 $\pm$ 4.49 a	57.85 $\pm$ 6.30 a

529 **Table 4**

530 Total surface free energy (γ_s) with the corresponding Lifshitz van der Waals component (γ_s^{LW} ; apolar) and acid-base components (γ_s^{AB} ; γ_s^+ and
 531 γ_s^-), and solubility parameter (δ) of adaxial and abaxial leaf surfaces of 5 different maize populations.

Population	Leaf side	γ_s^{LW} (mJ m ⁻²)	γ_s^- (mJ m ⁻²)	γ_s^+ (mJ m ⁻²)	γ_s^{AB} (mJ m ⁻²)	γ_s (mJ m ⁻²)	δ (MJ ^{1/2} m ^{-3/2})
S Sahara	Adaxial	23.88	118.45	9.12	65.73	89.81	36.14
	Abaxial	23.80	123.69	13.14	80.62	104.42	40.53
W Sahara	Adaxial	26.03	102.17	7.66	55.95	81.98	33.81
	Abaxial	22.26	102.03	8.65	59.40	81.66	33.71
N Sahara	Adaxial	19.92	133.07	14.22	86.99	106.91	41.25
	Abaxial	19.13	151.62	15.92	98.27	117.40	44.25
D Spain	Adaxial	23.57	110.29	12.01	72.80	96.36	38.16
	Abaxial	22.76	81.62	8.94	54.03	76.80	32.19
NW Spain	Adaxial	23.37	89.17	11.81	64.91	88.29	35.74
	Abaxial	22.48	46.88	4.84	30.11	52.59	24.23

Table 5

Leaf chlorophyll fluorescence, SPAD units, net photosynthetic rate (A), stomatal conductance (G) and transpiration rate (E) of 5 different maize populations. Data are means $\pm$ SD.

Population	ΦPSII^a	SPAD	A ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	G ($\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$)	E ($\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$)
S Sahara	0.768 $\pm$ 0.005	58.0 $\pm$ 2.7	20.338 $\pm$ 0.753	0.0381 $\pm$ 0.0031	0.0014 $\pm$ 0.00008
W Sahara	0.760 $\pm$ 0.004	50.6 $\pm$ 3.7	21.976 $\pm$ 0.832	0.0360 $\pm$ 0.0020	0.0012 $\pm$ 0.00006
N Sahara	0.765 $\pm$ 0.006	61.3 $\pm$ 3.3	26.368 $\pm$ 1.123	0.0391 $\pm$ 0.0024	0.0015 $\pm$ 0.00010
D Spain	0.773 $\pm$ 0.005	47.5 $\pm$ 4.0	27.672 $\pm$ 1.244	0.0356 $\pm$ 0.0022	0.0014 $\pm$ 0.00011
NW Spain	0.766 $\pm$ 0.007	51.2 $\pm$ 2.7	25.258 $\pm$ 1.254	0.0474 $\pm$ 0.0046	0.0016 $\pm$ 0.00012