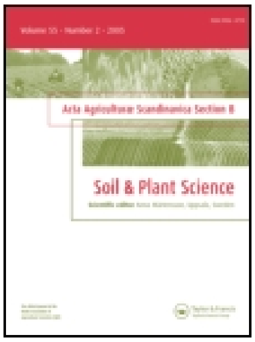

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ORIGINAL ARTICLE

Kaolin influences tomato response to salinity: physiological aspects

Francesca Boari[a]*, Giovanna Cucci[b], Antonio Donadio[a], Maria Immacolata Schiattone[c] and Vito Cantore[a]

[a]*Institute of Sciences of Food Production, National Research Council (CNR-ISPA), Via Amendola, 122/O, 70125 Bari, Italy;* [b]*Department of Agricultural and Environmental Science, University of Bari "A. Moro", Via Amendola, 165/A, 70126 Bari, Italy;* [c]*CIHEAM – Mediterranean Agronomic Institute of Bari, Via Ceglie 9, 70010 Valenzano, Bari, Italy*

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Environmental stress as salinity can negatively affect the physiology of tomato plants. Conditions leading to a reduction of transpiration can contribute to greater tolerance to salinity. Use of kaolin-based particle film technology (PFT) may be an effective tool to control stomatal conductance and transpiration rate, thus mitigating the detrimental effect of salinity. The present three-year study has investigated the effects of kaolin application on leaf gas exchange, leaf water potential, leaf and canopy temperature of field-grown tomato, irrigated with brackish water by drip method, in southern Italy. Treatments were: (1) three salinity levels of irrigation water (electrical conductivity of water = 0.5, 5 and 10 dS m^{-1}); (2) tomato plants treated or not with kaolin; and (3) two cultivars in each year. The increase in salinity caused the reduction of leaf water potential, stomatal conductance, net photosynthesis and transpiration rate, and the increase of leaf and canopy temperature. Kaolin has resulted in an improvement of leaf water potential, and the reduction in gas exchange variables in low-salinity conditions. Under high salinity, kaolin was effective in limiting the reductions in net photosynthesis and reducing leaf and canopy temperature. These latter variables were slightly affected by kaolin, in different ways in respect to the saline treatments; while in non-saline conditions were 0.2–0.5°C higher in the kaolin-treated plants, the situation was reversed in more saline treatment. The variation of leaf and canopy temperature shows that kaolin influences the thermal balance mainly for the dual effect of reflection of the incident radiation and partial occlusion of the stomata. Kaolin mitigated detrimental effects of salinity also on yield, contributing to the improvement of income for the farmers. The use of kaolin-based PFT may be an effective tool to alleviate salinity stress in tomato production under arid and semi-arid conditions.

Keywords: gas exchange; leaf and canopy temperature; photosynthesis; *Solanum lycopersicum* L.; transpiration

Introduction

In the Mediterranean areas, the indiscriminate exploitation of the groundwater has deteriorated water quality (salinity increase; Scheidleger et al. 2004), which often causes heavy damages to irrigated crops and soil fertility.

The soil salinity rise causes crop energy increase to extract water from soil and ion excess which affects the plant cells, that means growth and yield reduction and sometime death (Munns 2002, 2005). The reduction in growth is consequence of several physiological responses such as modification of ion and water relations, mineral nutrition, stomatal density and size, photosynthetic efficiency, carbon allocation or utilisation (Munns et al. 2006; Munns & Tester 2008).

The rate of photosynthetic CO_2 assimilation is generally reduced by salinity, partly due to a reduced stomatal conductance and consequent restriction of the availability of CO_2 for carboxylation, partly because of non-stomatal inhibition caused by direct effects of NaCl on photosynthetic apparatus (toxic effect; Flexas et al. 2004; Redondo-Gómez et al. 2007; Chaves et al. 2009). The reduction in stomatal conductance also causes a decrease in transpiration rate (Gul et al. 2001; Sharma et al. 2005;

*Corresponding author. Email: francesca.boari@ispa.cnr.it

Redondo-Gómez et al. 2007) and, consequently, a reduction in the transpirational cooling effect with the increase of leaf and canopy temperature (Kluitenberg & Biggar 1992; Sharma et al. 2005; Hackl et al. 2012).

Temperature and relative humidity (RH) of the air can markedly affect crop salt tolerance (Shalhevet 1994; Rauf et al. 2010). For instance, high temperatures associated with low RH of the air, by increasing the evapotranspirative demand, reduce crop salt tolerance because of both the increase in water flow and thus salt accumulation in the root zone (Helal & Mengel 1981; Meiri et al. 1982) and the greater hydraulic gradient in the soil–plant–atmosphere *continuum* (Li et al. 2001).

Instead, the techniques that reduce the transpiration rate could have a positive effect on salinity tolerance. Among those, there is the kaolin-based particle film technology (PFT; Gaballah & Moursy 2004; Moftah & Al-Humaid 2005; Angbabu et al. 2007), a multi-functional, environmentally friendly, material that provides effective insect control, mitigates heat stress and contributes to the production of high-quality fruit and vegetables (Glenn et al. 1999; Glenn & Puterka 2005).

The hypotheses here tested were that the kaolin-based PFT may be an effective tool to control stomatal conductance and transpiration rate, thus mitigating detrimental effects of salinity. To verify these hypotheses, the present study investigated the effects of kaolin particle film application on leaf gas exchange, plant water status, leaf and canopy temperature of field grown tomato, a widespread crop of many areas of southern Italy with salinity problems (Polemio & Limoni 2001; Ancona et al. 2010).

Materials and methods

Experimental site and climate

The research was carried out in the summer period of 2007, 2008 and 2009 in the field at the experimental farm 'E. Pantanelli' of the University Aldo Moro of Bari at Policoro (MT), southern Italy (40° 10′20′′ NL, 16°39′04′′ EL, altitude 15 m a.s.l.). The soil, more than 1.2 m depth, was loamy. Physical and chemical characteristics of the soil are reported in Table 1. This site has sub-humid climate according to the De Martonne classification (Cantore et al. 1987).

Climatic data were collected from a standard weather station located about 50 m from the experimental field (Cantore et al. 2012). The weather was characterised by clear sky conditions with a typical bell-shaped solar radiation (Rs) pattern and maximum values (about 950 W m^{-2}) reached at midday (Figure 1) during all physiological measurements. Air temperature (T_a) differed in maximum values: about 33°C (31 July 2007, 6 August 2007 and 21 July 2008), 36°C (14 July 2008), 34°C (21 and 28 July 2009), 40°C (6 August 2009) and 32°C (18 August 2009; Figure 1). At noon, air was dry, with values of RH that ranged between 19% (6 August 2009) and 35% (31 July 2007; Figure 1). Wind speed (Ws) ranged between 1.5 and 3.5 m s^{-1} (light breeze) of all days, with the exception of 6 August 2009 on which there was a moderate breeze (6.6 m s^{-1}; Figure 1). For the later day, there was a high-evapotranspirative demand due to high T_a, very low RH and moderate Ws.

Table 1. Main physical, chemical and hydrological characteristics of the soil.

Particle-size analysis		
Total sand (2 > ø > 0.02 mm)	(g 100 g^{-1})	40.0
Silt (0.02 > ø > 0.002 mm)	(g 100 g^{-1})	37.1
Clay (ø < 0.002 mm)	(g 100 g^{-1})	22.9
Chemical properties		
Total nitrogen (Kjeldahl method)	(g kg^{-1})	1.67
Available phosphorus (Olsen method)	(mg kg^{-1})	26.7
Exchangeable potassium (ammonium acetate method)	(mg kg^{-1})	227.0
Organic matter (Walkley Black method)	(g 100 g^{-1})	3.6
Total limestone (Dietrich-Fruhling method)	(g 100 g^{-1})	1.5
Active limestone	(g 100 g^{-1})	0.5
ECe	(dS m^{-1})	0.95
ESP	(%)	1.9
pH (pH in H_2O)		7.7
Hydrological properties		
Field capacity	(g 100 g^{-1})	31.5
Wilting point (−1.5 MPa)	(g 100 g^{-1})	15.0
Bulk density	(kg dm^{-3})	1.25

ECe, saturation extract electrical conductivity ESP, exchangeable sodium percentage.

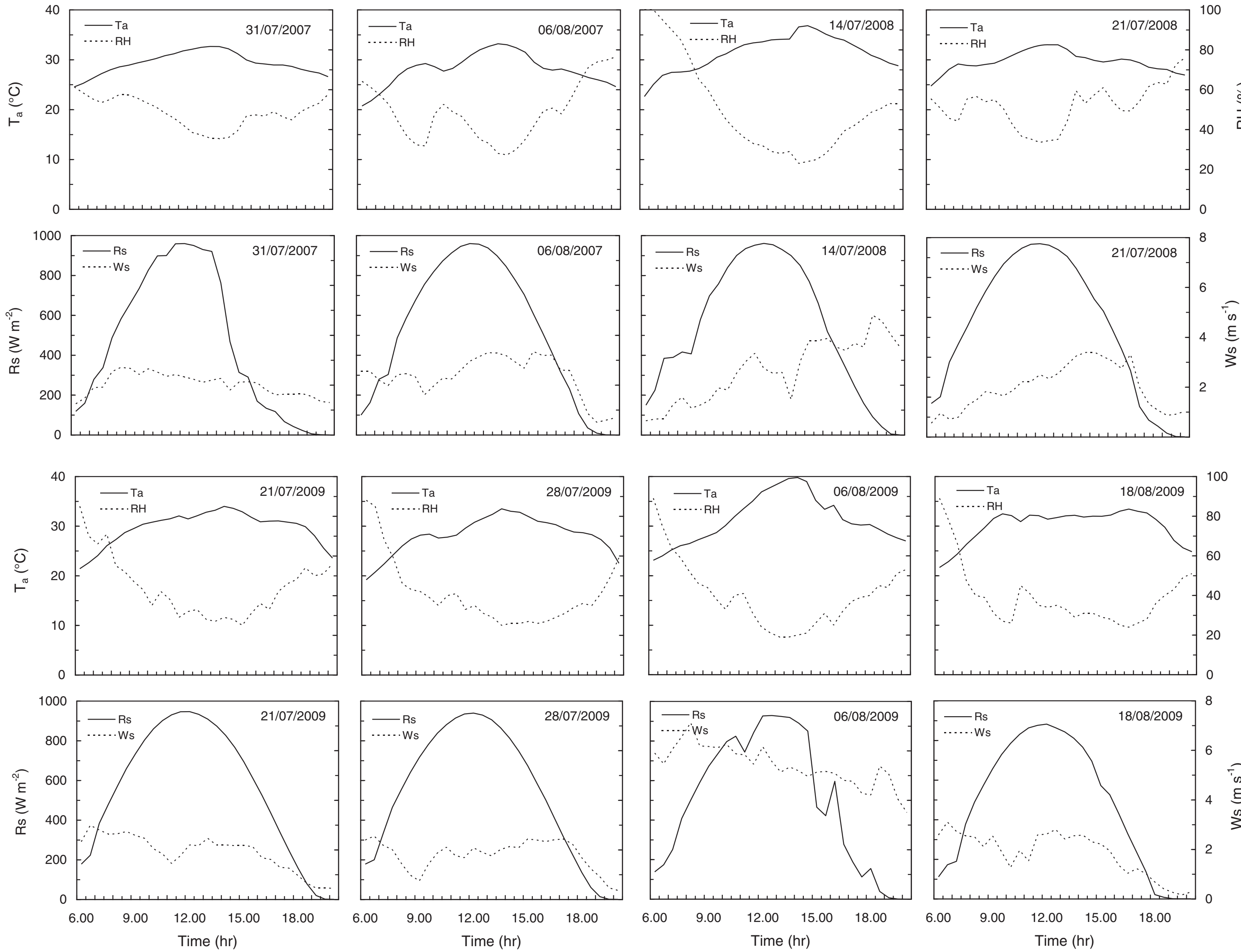

Figure 1. Diurnal trend of air temperature (T_a), relative humidity (RH), solar radiation (Rs) and wind speed (Ws), during the days when tomato physiological attributes were measured.

Table 2. Main chemical characteristics of irrigation water of the three salinity treatments.

Treatments	ECw (dS m^{-1})	pH	Soluble anions (meq L^{-1}) CO_3^{2-}	HCO_3^-	Cl^-	SO_4^{2-}	Soluble cations (meq L^{-1}) Ca^{2+}	Mg^{2+}	Na^+	K^+	SAR
S0	0.5	7.19	0.01	3.51	1.18	2.79	5.73	2.57	1.02	0.04	0.5
S1	5.0	7.48	0.01	3.72	66.01	3.21	8.63	2.70	49.90	0.10	21.0
S2	10.0	7.56	0.01	3.96	123.61	3.25	11.31	2.60	98.36	0.13	37.3

ECw, electrical conductivity of water; SAR, Sodium Adsorption Ratio.
Note: S0, S1 and S2 indicate, respectively, the ECw equal to 0.5, 5, and 10 dS m^{-1}.

Cultural practices

Tomato (*Solanum lycopersicum* L.) plants were transplanted on soil mulched with black polyvinyl chloride (PVC), at the third true leaf stage on 5 June 2007, 12 May 2008 and 25 May 2009 in single East–West-oriented rows, 1.5 m apart, with 0.25 m between plants in the row (2.7 plants m^{-2}). Pest control was performed by integrated pest management strategies and consisted of two or three treatments by applying 500 mL ha^{-1} of Confidor 200 SC (Bayer, CropScience, Milan, Italy), from transplanting until the first kaolin application. Hand weeding was performed throughout the growing period.

Fertiliser was applied to the soil pre-plant (120 and 115 kg ha^{-1} of N and P_2O_5) and incorporated.

Watering was performed with drip method every two to three days to keep soil water content in the root zone close to the readily available water (40% of available water depletion) and to restore 100% of crop evapotranspiration (ETc). Drip lines, with in-line emitters located 0.30 m apart and an emitter flow rate of 4 L h^{-1}, were placed 0.1 m away from the plants. The ETc was calculated by evopotranspirometric method, utilising daily values of 'class A' pan evaporation, pan coefficient reported by Castrignanò et al. (1985), and the crop coefficients reported by Tarantino and Caliandro (1984), adjusted for saline treatments (Allen et al. 1998).

For the first 10 days after planting (DAP), plants of all treatments were watered with fresh water to favour seedlings rooting. Afterwards saline treatments were initiated.

Treatments

Three electrical conductivity of irrigation water (ECw = 0.5, 5 and 10 dS m^{-1}, showed as S0, S1 and S2, respectively), two kaolin treatments (kaolin treated and control untreated, showed as K and C, respectively) on two tomato cultivars in each year (in 2007 and 2008 'HLY 19' and 'Perfectpeel', respectively by Hazera Genetics Ltd, Brurim Israel, for Italy COIS'94® S.p.A., Catania; Seminis Vegetable Seeds, Inc., Parma, Italy. In 2009 'Coimbra' and 'ISI 24424' both by ISI Sementi S.p.A., Fidenza, Italy), were compared. Treatments S1 and S2 were obtained by adding sea salt to the fresh water (S0). Chemical characteristics of irrigation water are reported in Table 2. A split plot design with three replications was used with 24 m^2 plot size.

At the fruits enlargement stage, kaolin (Surround® WP, Serbios S.r.l., Badia Polesine-RO, Italy) suspension (4% w/v) was applied with a backpack power sprayer (model MS073D, Maruyama Mfg. Co, Inc., Japan) in the K treatment. In the control fresh water was sprayed with the same sprayer. Kaolin applications were repeated every 7–10 days until the fruit ripening stage.

Gas exchange at leaf scale

Leaf gas exchange (net CO_2 assimilation – A, transpiration – E, stomatal conductance – g_s) was measured with a portable photosynthetic open-system ADC-LCA3 (Analytical Development Co., Hoddesdon, UK) equipped with an assimilation leaf chamber 6.2 cm^{-2} large. Photosynthetic water productivity (pWP) was computed from A/E ratio.

Leaf gas exchange measurements were performed in clear sky days (photosynthetically active radiation [PAR] > 2100 µmol m^{-2} s^{-1}) between 11:30 and 14:30 hr on: (1) two days (1 and 6–8 days after the first kaolin application) at fruits enlargement stage in 2007 (31 July and 6 August) and 2008 (14 and 21 July); (2) the day after each kaolin application (21 and 28 July, 6 and 18 August) between fruit enlargement and fruit mature green stage in 2009. In addition, on 21 July 2009, six sets of measurements every two hours during the day were made.

At each measurement time, on two plants per plot, two upper fully expanded, healthy, terminal and sun well-exposed leaves were chosen for measurements. The kaolin-treated leaves with the most uniform coating were chosen. Kaolin-treated leaves and untreated ones were randomly measured. Each measurement lasted two to three minute.

Leaf and canopy temperature measurements

Leaf temperature (T_l) was assessed on the abaxial surface simultaneously with leaf gas exchange measurements, using a fine-wire thermocouple mounted in the leaf chamber (PLC) of ADC-LCA3.

Canopy's temperature (T_c) was measured, on top of plants on the same days and time of leaf temperature measurements, using an infrared (IR) thermometer (model 112 C; Everest Interscience, Tustin, California). Eight measurements for each plot were carried out at every date. The measuring order for the different treatments was completely random.

Leaf water potential

Leaf water potential (ψ_l) was simultaneously measured with leaf gas exchange and canopy temperature, with a pressure bomb (Model 3005, Ecosearch, Città di Castello-PG, Italy), on two fully expanded, healthy, young leaves of the same plants where gas exchange was monitored.

Soil water content and salinity

In the same days of the gas exchange measurements, in three places per plot, crosswise to the row at 0, 0.25 and 0.50 m from the emitters, at three different depths (0.20, 0.40 and 0.60 m), were measured: (1) soil water content by gravimetric method, collecting soil samples through a cylindrical probe (∅ 2.5 cm); and (2) *in situ* electrical conductivity of saturated extract (ECe) by an EC-probe (Eijkelkamp Agrisearch Equipment, Geisbeek, the Netherlands), according to Rhoades and van Schilfgaarde (1976) method.

Kaolin residue measurements

Kaolin residue on adaxial and abaxial leaf surface was determined (Cantore et al. 2009a) on leaves utilised for gas exchange measurements at the end of each measurement set.

Statistical analysis

Analysis of variance of treatment effects was performed, and means were separated according to the post hoc Student-Newman-Keuls (SNK) test, using the statistical package SPSS 12.0 (SPSS Inc., Chicago, IL).

Results

Soil salinity

The irrigation with brackish water determined an increasing trend in soil salinity during the crop cycle. ECe, as average of the soil layer 0–0.60 m and of the three distances from the dripper (0, 0.25 and 0.50 m across the rows), in the treatment irrigated with fresh water (S0) was almost unchanged (0.9–1.2 dS m^{-1}), while in the two saline treatments it reached values ranging between 3.2 and 4.4 dS m^{-1} in S1 and between 5.1 and 6.8 dS m^{-1} in S2 (Table 3). In Figure 2 is represented the typical distribution of ECe, in relation to the depth and distance from the drippers, for the different levels of salinity, representative of the different dates of survey. The distribution of the salts brought with the irrigation water is varied both with depth and with distance from the dripper. Particularly, the lowest values of ECe were recorded in the most superficial soil layer (0–20 cm) below the drippers. The highest values were recorded in the most superficial layer of soil, 50 cm from the dripper (Figure 2).

Kaolin residue

In 2007 and 2008, one day after the first kaolin application, kaolin residue amounted to 150 and 31 μg cm^{-2}, on adaxial and abaxial leaf surfaces, respectively. About one week later, the kaolin coating was reduced by about 19% (adaxial leaf surface) and 18% (abaxial leaf surface; Table 4). In 2009 the residue of kaolin increased from the first to the last date of measurements as consequence of four kaolin applications, despite partial washing-off of particle film caused by a light rain (5 mm, on 14 August):

Table 3. Values (±SD) of electrical conductivity of the soil saturation extract (ECe, dS m^{-1}) in the days of gas exchange measurement, for the different salinity treatments.

	2007		2008		2009			
Salinity treatments	31/07	06/08	14/07	21/07	21/07	28/07	06/08	18/08
S0	0.9 ± 0.21	1.0 ± 0.11	1.0 ± 0.22	0.9 ± 0.15	0.9 ± 0.09	1.0 ± 0.08	0.9 ± 0.19	1.2 ± 0.23
S1	3.2 ± 0.54	3.3 ± 0.48	4.2 ± 0.52	4.4 ± 0.63	3.5 ± 0.77	3.4 ± 0.24	3.8 ± 0.46	4.0 ± 0.78
S2	5.1 ± 0.73	5.4 ± 0.65	5.9 ± 0.86	6.3 ± 0.94	5.4 ± 0.51	5.5 ± 0.87	5.9 ± 0.99	6.8 ± 0.65

Note: S0, S1 and S2 indicate respectively the electrical conductivity of irrigation water, ECw equal to 0.5, 5 and 10 dS m^{-1}. Each value is the average of values measured at three depths (0–20, 20–40 and 40–60 cm) and three distances from the dripper (0, 25 and 50 cm).

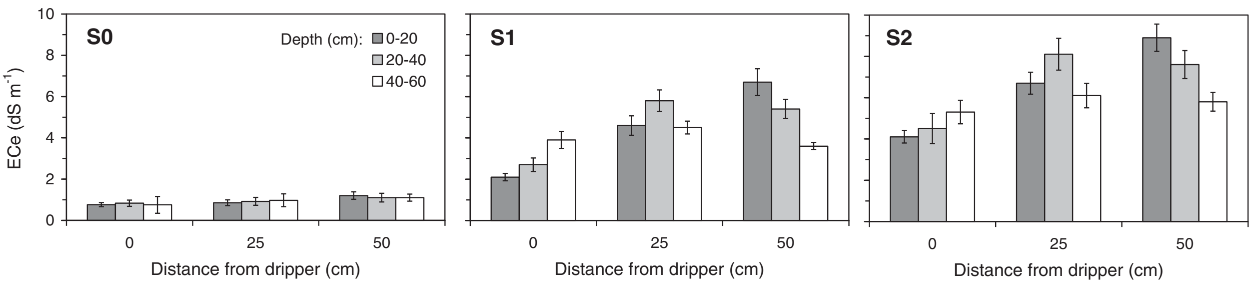

Figure 2. Representative distribution of salt in the soil (on 21 July 2008): electrical conductivity of the soil saturation extract (ECe, dS m^{-1}), at different soil depth and distances from the dripper, for the different salinity treatments.
Note: S0, S1 and S2 indicate, respectively, the electrical conductivity of irrigation water, ECw equal to 0.5, 5 and 10 dS m^{-1}. The vertical bars indicate standard deviation.

Table 4. Values (± SD) of kaolin residue (μg cm^{-2}) on adaxial and abaxial surface of leaves utilised to measure gas exchanges.

	2007		2008		2009			
Leaf surface	31/07	06/08	14/07	21/07	21/07	28/07	06/08	18/08
Adaxial	147 ± 26	116 ± 21	154 ± 19	129 ± 33	122 ± 29	245 ± 45	306 ± 66	322 ± 78
Abaxial	33 ± 7	26 ± 6	29 ± 10	25 ± 9	25 ± 12	38 ± 9	47 ± 21	52 ± 25

from 122 to 322 μg cm^{-2} (adaxial leaf surface) and from 25 to 52 μg cm^{-2} (abaxial leaf surface; Table 4).

Effects of salinity, kaolin and cultivar on physiological aspects of tomato

Leaf and canopy temperature had a similar trend with respect to treatments. Therefore, only data for T_c are presented.

In the three years, A, E, g_s, pWP, ψ_l and T_c did not differ between cultivars, while were influenced by salinity levels and by kaolin treatments (Table 5).

As expected, with increased salinity A, E, g_s and ψ_l decreased by 33.6%, 36.3%, 38.1% and 31.4%, respectively, and T_c increased by 5.8%, shifting from S0 to S2. Instead, pWP had no consistent response to treatments (Table 5).

In 2007 and 2008, kaolin reduced A, E and g_s, increased pWP and ψ_l, while did not affected significantly T_c. The magnitude of these changes was different between one day and about a week after the kaolin spraying. In fact, while the reduction of A, E, g_s and the increase of ψ_l were highest (respectively, 17.2%, 21.3%, 23.4% and 17.6%), the day following the kaolin application, these differences were significantly reduced about a week later, specially for A.

In 2009, the effect of the further applications of kaolin can be highlighted. Particularly, A, E and g_s were reduced after the first and second kaolin spraying, but increased after subsequent applications. ψ_l was always higher (by an average of 12.2%) with the application of kaolin.

The response to kaolin application was different in relation to saline treatments.

In fact, there was a significant interaction between salinity and kaolin treatment for most of the variables (Figure 3). While in S0, kaolin treatments showed a significant reduction in gas exchange variables, the decrease was slowed down in S1, and was almost annulled in S2. Such behaviour had been much more evident the day following the kaolin application, while it was attenuated after about a week.

In fact, in 2007 and 2008, the day after the spraying of kaolin, A, E and g_s were reduced on average of 27.8%, 32.9% and 32.3% in S0 and of 17.9%, 23.4% and 28.9% in S1, respectively, while in S2 were increased slightly. About a week later, in kaolin-sprayed plants, the same variables on average decreased, respectively, by 7.5%. 13.2% and 12.2% in S0, of 4.0%, 4.3% and 3.3% in S1, while increased by 10.8%, 12.0% and 12.4% in S2.

Leaf water potential was improved after kaolin application under salinity stress, especially the first day after the kaolin spraying. In fact, the increase in ψ_l has changed from 14% to 5.7% of S0 and from 24% to 16.3% of S2, one day and about a week after the application of kaolin, respectively.

As a result of kaolin application, T_c increased by 1.5% in S0, while was decreased under salinity stress (0.3% in S1 and 2.4% in S2).

Table 5. Physiological measures carried out: one (31 July 2007 and 14 July 2008), six (6 August 2007) and eight (21 July 2008) days after the first kaolin application in 2007 and 2008; the day after the kaolin applications in 2009 (21 and 28 July, 6 and 18 August 2009).

Treatments	A	E	g_s	pWP	ψ_l	T_c	A	E	g_s	pWP	ψ_l	T_c
	31 July 2007						6 August 2007					
Salinity	**	**	**	*	*	*	**	**	**	ns	*	*
S0	21.5 a	5.1 a	0.61 a	4.3 ab	−1.30 a	30.5 c	22.7 a	5.3 a	0.62 a	4.3	−1.29 a	30.0 b
S1	18.3 b	4.2 b	0.49 b	4.4 a	−1.33 a	31.2 b	18.1 b	4.4 b	0.49 b	4.1	−1.37 b	30.7 b
S2	14.4 c	3.6 c	0.36 c	4.0 b	−1.51 b	32.0 a	15.8 c	3.8 c	0.41 c	4.2	−1.54 c	31.7 a
Kaolin	**	**	**	**	*	ns	ns	ns	ns	ns	*	ns
K	16.2 b	3.6 b	0.39 b	4.5 a	−1.25 a	31.2	18.6	4.3	0.48	4.3	−1.32 a	30.7
C	19.9 a	5.0 a	0.58 a	4.0 b	−1.51 b	31.2	19.1	4.6	0.53	4.1	−1.47 b	30.9
Cultivar	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
HLY 19	18.0	4.2	0.50	4.3	−1.36	31.3	18.6	4.4	0.50	4.2	−1.37	31.1
Perfectpeel	18.1	4.4	0.48	4.1	−1.40	31.1	19	4.6	0.52	4.1	−1.42	30.5
	14 July 2008						21 July 2008					
Salinity	**	**	**	*	*	**	**	**	**	ns	*	*
S0	21.8 a	5.6 a	0.54 a	3.9 b	−0.97 a	31.7 b	23.6 a	6.1 a	0.61 a	3.9	−0.97 a	31.1 b
S1	17.9 b	4.4 b	0.49 b	4.7 a	−1.22 b	32.1 b	19.1 b	5.0 b	0.49 b	3.8	−1.18 b	32.2 a
S2	14.5 c	3.3 c	0.37 c	4.1 b	−1.44 c	34.2 a	16.6 c	4.3 c	0.44 c	3.9	−1.51 c	32.9 a
Kaolin	**	**	**	ns	*	ns	ns	*	ns	*	*	ns
K	16.5 b	4.1 b	0.43 b	4.1	−1.09 a	32.3	19.7	4.9 b	0.50	4.0 a	−1.15 a	31.9
C	19.6 a	4.8 a	0.50 a	4.2	−1.33 b	32.9	19.8	5.4 a	0.52	3.7 b	−1.29 b	32.2
Cultivar	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
HLY 19	18.2	4.3	0.48	4.2	−1.22	32.5	19.6	5.0	0.52	3.9	−1.21	32.1
Perfectpeel	17.9	4.3	0.46	4.1	−1.20	32.7	20.0	5.2	0.50	3.9	−1.23	31.9
	21 July 2009						28 July 2009					
Salinity	**	**	**	*	*	**	**	**	**	*	**	*
S0	22.5 a	5.6 a	0.64 a	4.1 b	−1.28 a	30.9 c	20.2 a	5.3 a	0.58 a	3.9 b	−1.02 a	31.2 c
S1	19.9 b	4.6 b	0.48 b	4.4 a	−1.36 b	31.6 b	17.9 b	4.0 b	0.43 b	4.5 a	−1.41 b	32.0 b
S2	16.1 c	3.5 c	0.40 c	4.6 a	−1.43 c	32.5 a	13.1 c	2.9 c	0.30 c	4.5 a	−1.53 c	33.5 a
Kaolin	**	**	**	ns	*	ns	**	**	**	**	*	ns
K	18.5 b	4.1 b	0.45 b	4.5	−1.31 a	31.7	16.1 b	3.6 b	0.35 b	4.6 a	−1.21 a	31.9
C	20.5 a	5.0 a	0.56 a	4.2	−1.40 b	31.6	18.0 a	4.5 a	0.52 a	4.0 b	−1.42 b	32.2
Cultivar	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Coimbra	19.7	4.6	0.52	4.2	−1.34	31.6	16.9	3.9	0.42	4.3	−1.29	29.8
ISI 24424	19.3	4.5	0.49	4.3	−1.37	31.6	17.2	4.2	0.45	4.2	−1.34	32.3

Table 5 (*Continued*)

Treatments	A	E	g_s	pWP	ψ_l	T_c	A	E	g_s	pWP	ψ_l	T_c
	6 August 2009						18 August 2009					
Salinity	**	**	**	*	*	*	**	**	*	ns	*	*
S0	18.4 a	6.0 a	0.62 a	3.1 b	−1.40 a	39.1 c	16.0 a	3.7 a	0.33 a	4.3	−1.03 a	31.7 b
S1	14.9 b	4.9 b	0.48 b	3.0 b	−1.50 b	40.3 b	13.0 b	3.0 b	0.26 b	4.3	−1.26 b	32.5 a
S2	11.2 c	3.2 c	0.30 c	3.4 a	−1.65 c	41.1 a	9.7 c	2.5 c	0.22 c	3.9	−1.36 c	33.2 a
Kaolin	**	*	*	*	*	*	*	*	ns	ns	*	ns
K	16.6 a	4.9 a	0.48 a	3.4 a	−1.42 a	39.8 b	13.5 a	3.2 a	0.28	4.3	−1.11 a	32.3
C	13.1 b	4.5 b	0.45 b	2.9 b	−1.61 b	40.5 a	12.2 b	2.9 b	0.26	4.1	−1.32 b	32.6
Cultivar	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Coimbra	14.5	4.6	0.45	3.2	−1.50	40.3	12.7	3.0	0.26	4.2	−1.19	32.5
ISI 24424	15.0	4.8	0.48	3.2	−1.53	39.9	13.1	3.2	0.28	4.1	−1.24	32.3

S0, S1 and S2 indicate respectively the electrical conductivity of irrigation water, ECw equal to 0.5, 5, and 10 dS m^{-1}. K and C represent, respectively, treatment with kaolin and untreated control.ns, *, ** indicate F test not significant or significant at $P < 0.05$ and $P < 0.01$, respectively. Mean separation within columns by SNK test ($P < 0.05$).
Note: Main effects of salinity level, kaolin application and cultivar on net assimilation (A, μmol m^{-2} s^{-1}), transpiration (E, mmol m^{-2} s^{-1}), stomatal conductance (gs, mol m^{-2} s^{-1}), photosynthetic water productivity (pWP, μmol m^{-2} s^{-1}/mmol m^{-2} s^{-1}), leaf water potential (ψ_l, MPa) and canopy temperature (T_c, °C).

In 2009, on the first two dates of measurements, variables had a trend similar to that of previous years, in relation to kaolin treatments and to salinity (57 and 64 DAP).

However, on 6 August (73 DAP), characterised by a high-evapotranspirative demand of the atmosphere due to moderate Ws, high T_a and low RH, kaolin has proven to be very effective in mitigating the strong stress conditions. In fact, in S0, where in the other dates there had been a significant reduction in gas exchange as a result of the application of kaolin, this did not happen, and the effectiveness of kaolin increased with rising salinity. Indeed, in S2, with the application of kaolin, A, E, g_s and ψ_l increased, respectively, by 76.5%, 64.0%, 72.7% and 17.7%, while T_c decreased by about 2.7% The last dates of surveys in which tomato, going to senescence, reduced gas exchange, significant differences were observed between K and C in S0 and S1 for most of the variables. Instead in S2, the kaolin application increased A (51.9%), E (33.3%), g_s (31.6%) and ψ_l (18.7%), and reduced T_c (3.1%).

The daily trend of A shows that in S0 the major differences between K and C occur among 11:00 and 13:00, and at 17:00: (1) in C highest value was reached at 11:00 and then decreased progressively; (2) in K, A values remained unchanged from 11:00 to 15:00, and then fell sharply at 17:00 as occurred also for Rs (Figures 1 and 4). Instead, in S2, C and K treatments had similar values of A, except that at 15:00 when the values of K were higher than those of C. Similar trend was observed for E and g_s. However, at 17:00 in C of S0, these two variables did not reduce drastically as was occurred for A.

The trend of ψ_l shows that kaolin tends to improve plant water status during the whole day.

Highest values of T_c were found at 15:00, as well as the major differences among the treatments. Values were of 0.75°C and 1.1°C lower in K, compared to C, for S0 and S2, respectively.

Discussion

The increase in salinity reduced gas exchange and leaf water potential of tomato plant.

The salinity had reduced osmotic potential (ψ_π) of the soil circulating solution (increased osmotic pressure) and, consequently, total soil water potential (ψ_t; Caruso 1993). This results in decreased water availability for the plant (Ayers & Westcot 1994; Yeo et al. 2000; Munns 2002), reducing ψ_l and, consequently g_s and gas exchange (Adams & Ho 1989; Vassey & Sharkey 1989; Brugnoli & Lauteri 1991), which has also led to a reduction in the transpirational cooling effect with a T_c increase of 1–2°C between S0 and S2. In fact, for many vegetative

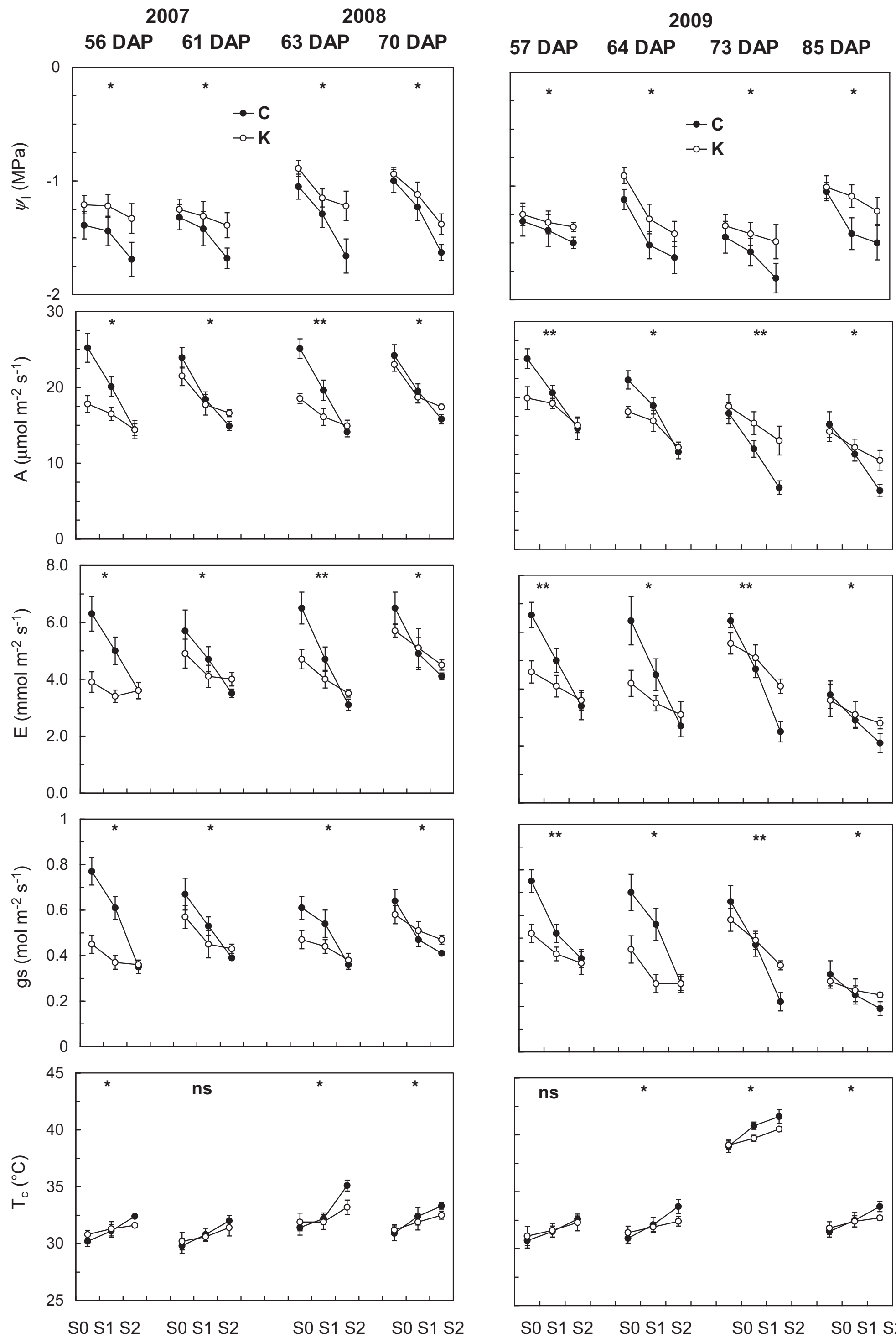

Figure 3. Physiological measures carried out: (1) one (54 DAP) and six (61 DAP) days, and one (63 DAP) and eight (70 DAP) days after the first kaolin application in 2007 and 2008, respectively; (2) the day after the kaolin applications in 2009 (57, 64, 73 and 85 DAP): interaction between salinity levels (S0, S1, S2) and kaolin treatments (C-control without kaolin; K-kaolin sprayed plants) on leaf water potential (ψ_l), net assimilation (A), transpiration (E), stomatal conductance (g_s) and canopy temperature (T_c).

Note: S0, S1 and S2 indicate respectively the electrical conductivity of irrigation water, ECw equal to 0.5, 5 and 10 dS m^{-1}. DAP indicate the days after planting. The vertical bars indicate the standard deviation. ns, *, ** indicate F test not significant or significant at $P \leq 0.05$ and $P \leq 0.01$, respectively.

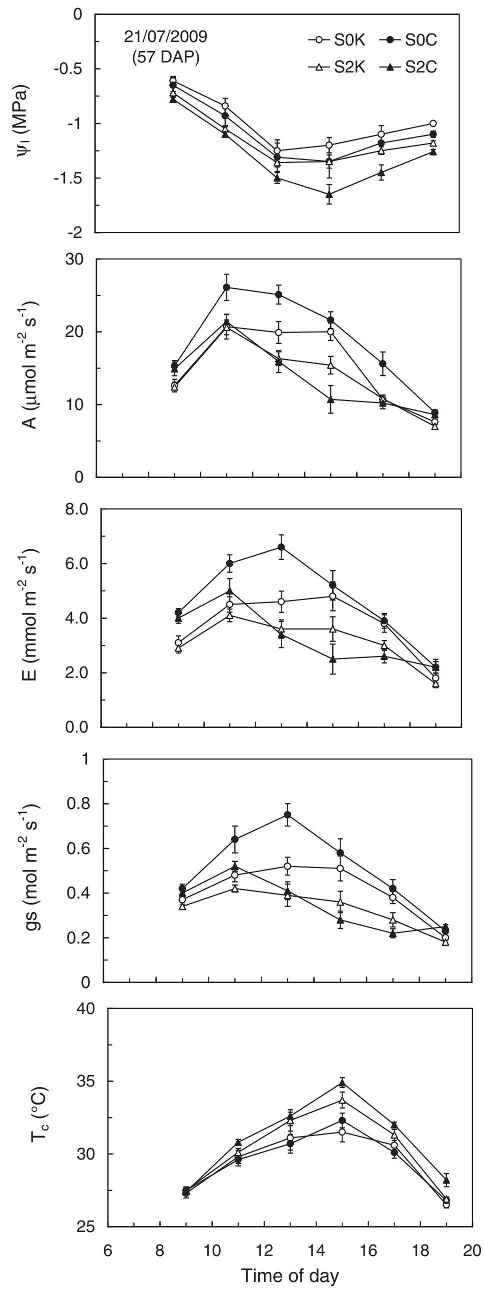

Figure 4. Physiological measures carried out during 21 July 2009 (57 DAP): effect of salinity levels (S0, S2) and kaolin treatments (C-control without kaolin; K-kaolin sprayed plants) on leaf water potential (ψ_l), net assimilation (A), transpiration (E), stomatal conductance (g_s) and canopy temperature (T_c).
Note: S0 and S2 indicate, respectively, the electrical conductivity of irrigation water, ECw equal to 0.5 and 10 dS m^{-1}. DAP indicate the days after planting. The vertical bars indicate the standard deviation.

surfaces, leaf or canopy temperature is strongly influenced by the portion of incoming radiant energy that is dissipated by transpiration. With the depletion of soil water and/or with the increase of soil salinity and subsequent reduction in the water status of the plant, transpiration is reduced and leaf temperature increases (Kluitenberg & Biggar 1992; Sharma et al. 2005; Hackl et al. 2012).

Kaolin caused reduction of A, E, g_s, and increase in ψ_l and pWP, especially just after its application. Such effect was reduced in intensity a week after kaolin application, when it was detected a reduction of kaolin coating leaves.

The reduction of A, E and g_s in kaolin treated plants, is probably due to a partial obstruction of the stomata by kaolin (Cantore et al. 2009a). This hypothesis is supported from the data reported by Srinivasa Rao (1985, 1986) and from observations made on apple trees (Le Grange et al. 2004; Gindaba & Wand 2007). On tomato, in agreement with reduction of A and E at leaf scale, Cantore et al. (2009a) found also the reduction of net assimilation and evapotranspiration at canopy scale. The reduction of g_s and, therefore, of E caused by the kaolin is also reported for many species such as apple (Le Grange et al. 2004), pecan (Lombardini et al. 2005), tea (Anandacoomaraswamy et al. 2000), pepper (Sheikh & Mall 1978), bean (Tworkoski et al. 2002; Cantore et al. 2009b), *Polianthes tuberosa* L. (Moftah & Al-Humaid 2005), orange (Pace et al. 2009), tomato (Srinivasa Rao 1985, 1986; Nakano & Uehara 1996; Cantore et al. 2009a) and potato (Cantore unpublished data). On grapevine, however, g_s was reduced in well-watered crop, while it was not affected in stressed one (Shellie & Glenn 2008). However, there are many experimental findings that do not show reduction in gas exchange resulting from application of kaolin (Glenn et al. 2003; Glenn 2010). These results prevail for crops such as apple tree whose leaves have stomata only on the lower surface (hypostomatic). However, in the species with leaves amphistomatic (tomato, potato, pepper and bean; Adedeji et al. 2007), gas exchange is reduced probably because kaolin is deposited mainly on the upper surface of the leaves, and only a small portion is deposited on the bottom. In hypostomatic species it only marginally affects the obstruction of the stomata, contrary to amphistomatic species. The amount of kaolin that is deposited on the two surfaces of the leaf also depends on the equipment utilised and mode of spraying. In herbaceous crops, the spraying takes place from top to bottom, for which it is the upper surface of the leaves that receives the greatest amount of kaolin. In tree crops, however, generally the spraying occurs from the bottom upwards, with a different inclination of the direction of the spray, in relation to the height of the sprayer and trees. This would lead to a different distribution of the kaolin on the leaf surfaces and would explain the different effect on gas exchange reported by different authors, as is the case of the apple tree.

Instead, the reduction of A cannot be attributed to the reflection of the PAR by the kaolin, whereas an eventual reduction of the PAR of 20%, as reported in other studies (Wünsche et al. 2004), would lead to PAR of 1700 µmol m^2 s^{-1} or above, not limiting for tomato photosynthesis.

The reduction of the negative effects of kaolin on gas exchange to a week after treatment can be attributed to reduction of obstruction of stomata by the kaolin: (1) for the loss of 18–19% of kaolin powder from the leaves; and (2) for the stomatal movement that probably helps to restore its opening.

Furthermore, there was a significant interaction between salinity and kaolin treatments for most of the variables. In particular, while with irrigation with fresh water kaolin showed a significant reduction in the above variables, this decrease was mitigated in S1, and cancelled in S2.

The interaction between salinity and kaolin treatment is in agreement with observations made on the vine, on which kaolin resulted in reduction of g_s on the crop under good water supply, while had not caused any change on the water-stressed crop (Shellie & Glenn 2008). Water stress, in fact, in many aspects causes the same effects of salt stress for the reduction of the soil ψ_t (Yeo 1983; Larker 2003). While with no stress or moderate stress prevails the stomatal limitation caused by the kaolin, under severe salt stress, when also the non kaolin-treated crop has a strong stomatal limitation, kaolin shows effectiveness as antitranspirant by limiting water loss, allowing the crop to have a high water status.

Canopy temperature was slightly affected by kaolin in different ways with respect to the salinity: in S0 T_c was 0.2–0.5°C higher in the kaolin-treated plants, while the situation was reversed in S2. In fact, in the latter treatment, the kaolin has determined a reduction of 0.2–1.0°C, especially at noon time.

The temperature at leaf and canopy scale shows that kaolin influences thermal balance of vegetation mainly for the dual effect of reflection of the incident radiation and partial occlusion of the stomata. The reflection would help to reduce the temperature increase, while the occlusion of stomata would act in the opposite direction due to the lower thermoregulation. Therefore, in crop irrigated with fresh water prevails the reduction of thermoregulation for the partial occlusion of stomata caused by kaolin, while in the crop with salt stress that already has reduced g_s prevails the kaolin reflective effect with a lower temperature increase.

These results apparently are in contrast with those obtained in researches in different crops, in which kaolin-based treatments reduced temperature at leaf and canopy scale of 1–6°C (Miranda et al. 2007; Steiman et al. 2007; DaMatta et al. 2008; Shellie & Glenn 2008).

Concordant results, however, are reported for pepper on which significant changes on these variables were not observed (Russo & Diaz-Perez 2005); potato, on which these were increased of 0.5–1.0°C (personal communication) and tomato under optimal water availability conditions on which the increases were of 1°C and 0.4°C, respectively, for leaf and canopy temperature (Cantore et al. 2009a).

The different behaviours between amphistomatic herbaceous species such as tomato, potato and pepper and hypostomatic tree species as the apple tree, agree with what was previously reported on the interaction between the different types of leaves, in relation to stomatal distribution and gas exchanges. In fact, in hypostomatic species as the apple tree, in which the kaolin generally does not obstruct the stomata, to the normal thermoregulation due to non-limited transpiration, it adds the reflection of radiation caused by the kaolin. In amphistomatic species, however, the thermal reduction determined by the reflection of the radiation is compensated by the lower thermoregulation by the transpiration, caused by the obstruction of stomata.

From the results obtained it can be concluded that, notwithstanding kaolin reduces net photosynthesis, its effects on reduction of transpiration and improvement of plant water status mitigates the harmful effect of heat and drought and contributes to improve salt tolerance of tomato.

In fact, in the saline treatments, the application of kaolin resulted in an increase of marketable yield (average of the three years) of 23% (S1) and 34% (S2; data not shown), consistent with the results obtained in wheat crop by Gaballah and Moursy (2004).

Balancing the increase in revenues due to the increased yield and the cost of kaolin application, it was estimated a significant increase of net income of tomato grower (up to 500–600 €/ha in non-saline conditions and 800–900 €/ha in saline conditions). Therefore, the kaolin is a valuable tool to improve the income of the farmer, particularly under conditions of salinity.

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