

Evaluation of Native Grasses for Sustainable Turfgrass in the Bioclimatic Mediterranean Region

Simonetta Fascetti, Giovanna Potenza, Vincenzo Candido,
Donato Castronuovo, Leonardo Rosati, Michele Perniola, Stella Lovelli,
Roberto Viggiani, and Vito Marchione

Abstract This study reports the results of a research project (*Mi.T.E.A.Med*) funded by the Italian Ministry of Agriculture. The research was organised in two phases: the first one involved the screening of the study area (Southern Italy) to find suitable turfgrass species and the second one focused on ex situ cultivation to test the ecotypes with salinity resistance. During the first step of the research, 11 sites from 6 regions of Southern and Central Italy were identified. In these sites, 24 ecotypes of *Cynodon dactylon* (L.) Pers. were collected and their habitus, phenology, and some biometric parameters have been determined. During the 2 years of research, both botanic and agronomic characterisation of the collected *C. dactylon* ecotypes was carried out. Some native accessions showed a behaviour similar to commercial cultivars, while an ecotype from the Abruzzo Region showed better results compared to the commercial cultivars for several quality indices. The results of this project showed that Mediterranean-adapted native grass species (e.g. *Cynodon dactylon* (L.) Pers.) are worth investigating for turfgrass, making for their performance and low resource requirement (especially water). This species can be used as promising alternatives to conventional non-native turfgrasses.

S. Fascetti (✉) • G. Potenza • V. Candido • D. Castronuovo • L. Rosati • M. Perniola •
S. Lovelli • R. Viggiani
School of Agricultural Forest Food and Environmental Sciences, University of Basilicata,
85100 Potenza, Italy
e-mail: simonetta.fascetti@unibas.it

V. Marchione
Department of Agricultural and Environmental Science, University of Bari, 75100 Bari, Italy

1 Introduction

The production and planting of turfgrasses is a fast-developing business with a growing demand for easy-to-use materials that usually involves the use of non-native species. Unfortunately, most of these species are invasive that may pose a threat to the biodiversity conservation of natural ecosystems (Celesti-Grapow et al. 2009)

Furthermore, the low adaptability of foreign varieties to the prevailing pedoclimatic conditions in Italy (Panella 1980, 1981; Veronesi and Panella 1985) prompted the beginning of a research programme to detect suitable native grass species for turfgrass uses. Unlike forage varieties, those considered for turfgrass should have limited growth thin leaves, ability to recover rapidly after thinning and resistance to trampling.

In Italy the most interesting turf species are grasses belonging to the subfamilies Festucoideae and Eragrostideae. The former are also defined as ‘cool-season grasses’ because of their preferential adaptation to cool and moist environments. The target species for turf uses in this subfamily are Kentucky bluegrass (*Poa pratensis* L.) characterised by a strong rhizomatous habitus and adapted to form long-lasting turfs of intermediate texture; perennial ryegrass (*Lolium perenne* L.) for fast-grown establishment; creeping red fescue (*Festuca rubra* L. subsp. *rubra*) and chewings fescue (*Festuca rubra* L. subsp. *commutata* Gaudin.), which are both with fine texture and rather tolerant to shading (the former has a rhizomatous habitus and is suited to sport lawns, the latter is tufted and less tolerant to trampling); tall fescue (*Festuca arundinacea* Schreb.), which has a rough texture but good tolerance to turf wearing; and creeping bentgrass (*Agrostis stolonifera* L.), characterised by the finest leaf texture and suited to high-quality turfs (Veronesi and Panella 1985). In Italy, the utilisation limit for all these species is represented by summer heat and drought stresses that typically affect the Mediterranean areas (Volterrani et al. 1997; Russi 2004; Annicchiarico and Russi 2005; Marchione 2004, 2008).

On the contrary, species of Eragrostideae subfamily are defined as ‘warm-season grasses’, being characterised by good growing ability under high temperatures and by vegetative standstill with low temperatures (Volterrani et al. 1997).

In particular, Bermuda grass (*Cynodon dactylon* (L.) Pers.) is the most rising species for turf establishment in the Mediterranean coastal and southern environments of Italy, where the climatic conditions are not suitable to cool-season grasses.

In Italy, many turfgrasses species have been introduced from foreign countries, but they showed a low adaptability to the prevailing Mediterranean climatic conditions (Panella 1980; Veronesi and Panella 1985). In fact, the plants come almost exclusively from environments different from those of the Mediterranean and the unsatisfactory performance is mainly due to the high susceptibility to summer stress and reduced growth in winter.

In this context, the availability of genetic material tolerant to summer drought and salinity has become a strong need for the sector of turfgrasses in the

Mediterranean area, especially following the spreading of turfgrass landscapes in arid and seashore areas.

The indigenous grass species are an important source of genetic variability that can provide better performance in terms of tolerance to salinity. In fact, some recent studies revealed that indigenous grass species display high phenotypic variability (Potenza et al. 2014).

Herein we provide the main results of the Mi.T.E.A.Med research project (improved turfgrass in Mediterranean environment: use of autochthonous species of plant and optimisation techniques) funded by the Italian Ministry of Agriculture.

This study was carried out on several native populations of Bermuda grass (*Cynodon dactylon* (L.) Pers.) that were evaluated for turf characteristics with the aim of selecting the most promising ecotypes in terms of lower water requirements and tolerance to salinity.

2 Materials and Method

The research was organised in two phases: the first one involved the screening of the study area flora (Central and South Italy) to find suitable turfgrass species characteristics for the Mediterranean bioclimate, while the second one focused on ex situ cultivation to test the ecotypes with salinity resistance.

The collection was carried out during summer 2011 and winter 2012. Samples of several macrotherm native species were collected in coastal and hilly, especially on sandy substrates dry and/or salty. Consequently to a preliminary screening, we selected for the detailed investigation only native populations of *C. dactylon* sampled in 11 sites (Fig. 1) distributed across 6 administrative regions in three distinct areas: the centre (regions of Lazio and Abruzzo), the south-west (regions of Basilicata, Calabria, Campania) and the south-east of Italy (Table 1).

The average annual temperatures of the collection sites ranged from 14 to 18 °C with annual rainfall of less than 600 mm received predominantly in the autumn and winter (Biondi and Baldoni 1995). Altitude of the collection sites ranged from 0 to 400 m a.s.l.

Every specimen was sampled from each location following the approach suggested by Romani et al. (2002) by walking on a transect. Whole plants (including roots embedded in soil) at the vegetative stage were collected every 2–5 m along the transect. A total of 24 samples of *Cynodon dactylon* were collected across the 10 sites.

Stolon fragments were transplanted into plastic pots in open field at Potenza (40°38'N; 15°48'E; 728 m a.s.l.), Southern Italy. The 450 cm³ plastic pots were filled with 300 cm³ of a specially formulated substrate for turf (Compo Sana 'Terriccio per tappeti erbosi', COMPO Agro Specialities—private society in Italy).

Each plant received 500 ml of water every 3 days. Morphological variables were measured both in situ and ex situ during the growing season (May–October 2012). Successively, plant samples were vegetatively multiplied by transplanting stolon



Fig. 1 Collected sites in the bioclimatic Mediterranean region

fragments into plastic pots in a cold greenhouse at Pantanello farm (+40°23'N, +16°47'E; 8 m a.s.l.), Metaponto. When vegetative material was enough, field plots were established at 'Lucana Prati' sod farm (+40°22'N, +16°47'E; 6 m a.s.l.), Metaponto (Fig. 2).

The soil of the experimental area was clay textured (64 % clay, 17 % silt and 19 % sand), with good total nitrogen (1.1‰) and available phosphorus (25 ppm) and rich in exchangeable potassium (350 ppm). The 24 samples of Bermuda grass were compared in a randomised complete block design with three replications. Plot surface area was 2.25 m² (1.5 m × 1.5 m).

Between the first and the second year (December 2012–March 2013), the colour loss interval was determined by calculating the duration (d) of vegetative stasis on the basis of **colour and ground cover** of each turf plot.

During the second year, from May to September 2013, the following data were recorded: **Growth rate**: it is measured by a turfmeter as weekly vertical growth of turfs maintained at 45 mm cutting height by a rotary mower. **Colour index**: it represents the intensity of the green colour, and it is influenced by genetic characteristics of the species, environmental stress (water and/or thermal stress), nutritional deficiencies and parasitic attacks. This parameter has been monthly measured using a Turf 500 NDVI-Turf Color Meter (Spectrum Technology, Aurora, USA) that measures the reflected light in the spectral bands of red (600 nm) and infrared (850 nm) which correlate with the concentration of chlorophyll pigments in the

Table 1 List of sample sites for *Cynodon dactylon* collection

| Samples | Location | Altitude (m a.s.l.) | Latitude (UTM) | Longitude (UTM) |
|---------|---------------------|---------------------|----------------|-----------------|
| 1 | Massafra (TA) | 30 | 677949 | 4492375 |
| 2 | Vasto (CH) | 5 | 478443 | 4661223 |
| 3 | Vasto (CH) | 5 | 478443 | 4661223 |
| 4 | Vasto (CH) | 5 | 478443 | 4661223 |
| 5 | Vasto (CH) | 5 | 478443 | 4661223 |
| 6 | Vasto (CH) | 5 | 478443 | 4661223 |
| 7 | Pisticci (MT) | 43 | 634308 | 4476229 |
| 8 | Pisticci (MT) | 167 | 631894 | 4470852 |
| 9 | Pisticci (MT) | 6 | 651327 | 4462126 |
| 10 | Ginosa (TA) | 5 | 666275 | 4484012 |
| 11 | Ginosa (TA) | 5 | 666275 | 4484012 |
| 12 | Ginosa (TA) | 5 | 666275 | 4484012 |
| 13 | Montescaglioso (MT) | 302 | 650379 | 4491214 |
| 14 | Feronia (RM) | 408 | 301916 | 4668620 |
| 15 | Diamante (CS) | 0 | 578877 | 4376962 |
| 16 | Conversano (BA) | 100 | 678146 | 4537292 |
| 17 | Conversano (BA) | 105 | 678146 | 4537292 |
| 18 | Matera (MT) | 268 | 640701 | 4501305 |
| 19 | Metaponto (MT) | 72 | 654410 | 440640 |
| 20 | Ascea (SA) | 5 | 515761 | 4438447 |
| 21 | Ascea (SA) | 5 | 515761 | 4438447 |
| 22 | Ascea (SA) | 5 | 515761 | 4438447 |
| 23 | Ascea (SA) | 5 | 515761 | 4438447 |
| 24 | Ascea (SA) | 5 | 515761 | 4438447 |

leaves. From the measurement of NDVI, a colour index on a scale from 1 (= brown) to 9 (= dark green) can be calculated. **Turf quality**, a synthetic index evaluated by visual observations: it is influenced by the uniformity, density, turf colour, leaf texture, percentage of coverage and presence of weeds and insect and disease damage. It varies from 1 (poorest quality) to 9 (highest quality—ideal turf). Ground cover percentage (GCP): it is an assessment of the ground cover of the plot; this index was evaluated monthly by visual estimation.

Contemporary, stolon fragments were transplanted in honeycombed styrofoam containers filled with a small quantity of peat (Fig. 3). These containers were placed over plastic containers containing 15 L of aerated Hoagland nutrient solution (EC = 2.5 dS m⁻¹; pH = 6.0) formulated with tap water (Hoagland and Arnon 1950). Solution contained the following nutrients as mmol L⁻¹: NO₃⁻ 13.5, NH₄⁺ 1.5, PO₄³⁻ 1.0, K⁺ 6.0, Ca²⁺ 5, Mg²⁺ 2.0 and SO₄²⁻ 2.0. Styrofoam containers were submerged to the soil surface in the solutions. Loss of nutrient solution was compensated by a weekly substitution. Nutrient solution pH was daily adjusted to get 6.5–7.0; it was constantly aerated and maintained at a constant volume. An automated heating system started working each time air temperature dropped under



Fig. 2 Field plots established at ‘Lucana Prati’ sod farm (+40°22’N, +16°47’E; 6 m a.s.l.), Metaponto

18 °C, while the greenhouse roof opened as soon as the temperature exceeded 25 ° C. An automatic weather station was placed in the greenhouse in order to measure meteorological data.

After 10 days of establishment and turf adjustment in the greenhouse, a salt treatment was imposed. Plants were subjected to one level of salt stress, 15 dS m⁻¹, corresponding to 150 mM NaCl through NaCl addition (commercial salt). There was a control treatment maintained at a 2.2 dS m⁻¹ salt level. Each experimental treatment was replicated three times arranging the pots according to a randomised block factorial scheme. In each pot, there were three plants for a total of nine plants per experimental treatment. In order to avoid osmotic shock to plants, NaCl addition to the nutrient solution occurred gradually.

Growth measurements were carried out during the experiment, the following morphological parameters were measured biweekly with a digital calliper: leaf length (l), leaf width (L) and distance between internodes of the latest fully expanded leaf (i) with five measurements per characteristic plant. Measurements were taken at a regular interval of 14 days. After 60 days, plants were harvested: dry matter and leaf number were obtained and counted, respectively. Total dry matter (hypogeous and epigeous) was obtained drying the samples in a ventilated oven at 75 °C until constant weight. Leaf area was measured at the end of the experiment trial, by a surface electronic detector (Model 3100, Li-Cor, Inc., Lincoln, NE, USA).

At the end of the experiment, Na⁺ and Cl⁻ ion concentration of leaf tissue was measured. Samples were oven dried at 70 °C and finely ground. A subsample of leaves from each of the two treatments was dried, ground and extracted in HNO₃



Fig. 3 Honeycombed styrofoam containers filled with a small quantity of peat of aerated Hoagland nutrient solution

(65 % v/v) to measure Na^+ concentrations on leaf extracts using a flame spectrophotometer (Flame Spectrophotometer, Varian 220 FS). Another subsample was ashed at 60 °C for one night, and subsamples of dry matter were used for extraction of Cl^- , using a carbonate and sodium bicarbonate solution. Cl^- was measured by a titration with silver nitrate solution.

2.1 Statistical Analysis

All data were analysed with ANOVA procedure and means were compared with Duncan's test, using SigmaPlot 11.0 for Windows (Systat Software Inc., San Jose, CA, USA). Significant differences were identified by Tukey's test with 5 % and 1 % significance. Afterwards, to assess the different patterns in behaviour of the ecotypes in response to aesthetic and agronomic characteristics, cluster analysis was performed with the procedure CLUST using Ward's method (Ward J. 1963) of significantly affected parameters (monthly growth, *colour index*, *aesthetic general appearance (AGA)*, *green cover percentage*, *interval loss of green colour*). In addition all salinity data were subjected to a cluster analysis (CA) using Ward's method (Ward J. 1963). These analyses were performed using Statistica V.10 (StatSoft Inc., USA).

3 Results and Discussion

The results of the agronomic behaviour carried out in 2012 and 2013, of the 24 native ecotypes of *C. dactylon*, are shown in Tables 2, 3, 4 and 5.

Table 2 Monthly and cumulative growth of ecotypes collected in Central and Southern Italy

| Samples | Monthly assessed growth (mm) | | | | | | | | | | Cumulative | |
|---------|------------------------------|----|------|----|------|----|--------|----|-----------|----|------------|---|
| | May | | June | | July | | August | | September | | | |
| 1 | 44 | d | 62 | hi | 49 | d | 42 | bc | 33 | ef | 230 | c |
| 2 | 28 | hi | 34 | o | 24 | mn | 21 | h | 27 | h | 135 | p |
| 3 | 26 | ij | 44 | l | 25 | lm | 18 | i | 29 | gh | 142 | n |
| 4 | 18 | mn | 39 | n | 19 | op | 25 | g | 12 | kl | 114 | r |
| 5 | 32 | fg | 58 | j | 29 | k | 32 | e | 33 | ef | 184 | i |
| 6 | 22 | kl | 35 | o | 25 | lm | 28 | f | 35 | ce | 146 | m |
| 7 | 22 | kl | 39 | n | 46 | e | 40 | c | 27 | h | 174 | j |
| 8 | 31 | fg | 43 | lm | 29 | k | 41 | bc | 24 | i | 168 | k |
| 9 | 22 | kl | 30 | p | 16 | q | 15 | jk | 10 | lm | 94 | t |
| 10 | 12 | o | 36 | o | 21 | no | 12 | l | 20 | j | 101 | s |
| 11 | 24 | jk | 53 | k | 44 | ef | 40 | c | 26 | h | 188 | h |
| 12 | 5 | p | 12 | q | 17 | pq | 18 | i | 10 | lm | 63 | v |
| 13 | 25 | j | 63 | gh | 35 | i | 35 | d | 29 | gh | 188 | h |
| 14 | 38 | e | 72 | bc | 41 | gh | 48 | a | 27 | h | 226 | d |
| 15 | 30 | gh | 60 | ij | 28 | kl | 36 | d | 37 | c | 192 | g |
| 16 | 17 | n | 35 | o | 30 | jk | 24 | gh | 10 | lm | 116 | q |
| 17 | 24 | jk | 41 | mn | 29 | k | 29 | f | 13 | k | 137 | o |
| 18 | 19 | mn | 35 | o | 15 | q | 14 | kl | 9 | m | 92 | u |
| 19 | 20 | lm | 74 | b | 42 | fg | 42 | bc | 34 | de | 213 | f |
| 20 | 33 | f | 68 | de | 40 | gh | 41 | bc | 30 | g | 212 | f |
| 21 | 55 | b | 65 | fg | 33 | ij | 41 | bc | 36 | cd | 230 | c |
| 22 | 43 | d | 58 | j | 20 | op | 13 | kl | 24 | i | 158 | l |
| 23 | 63 | a | 137 | a | 59 | a | 49 | a | 50 | a | 359 | a |
| 24 | 36 | e | 69 | d | 52 | c | 29 | f | 27 | h | 213 | f |

Values not having any letter in common are significantly different at $P \leq 0.05$ (Duncan's test)

The accessions caused significant difference in each of the tested parameters when subjected to analysis of variance. For simplicity, the results for each parameter have been analysed separately.

The 'ecotypes' showed significant differences in each of the tested parameters when subjected to analysis of variance. For simplicity, the results for each parameter have been analysed separately.

3.1 Total Growth

The mean total growth during the period May–September was 170 mm (Table 2), with extreme variations observed between ecotype 12 (63 mm) and ecotype 23 (359 mm). A similar behaviour was also found by Marchione (2008) on 12 commercial varieties of Bermuda grass.

Table 3 Colour index and days of winter colour loss interval of ecotypes during the trial period (2012–2013)

| Samples | May | | June | | July | | August | | September | | Mean May–Sept (2013) | | Winter colour loss interval (d) | |
|---------|-----|----|------|----|------|----|--------|----|-----------|----|----------------------|----|---------------------------------|----|
| | | | | | | | | | | | | | | |
| 1 | 6.8 | bd | 6.7 | ab | 6.8 | a | 6.0 | ce | 6.0 | de | 6.5 | ab | 133 | j |
| 2 | 6.5 | d | 6.2 | ef | 6.3 | be | 6.1 | bd | 6.0 | de | 6.2 | ce | 146 | f |
| 3 | 7.0 | ab | 6.6 | ac | 6.4 | bd | 6.2 | ac | 6.1 | cd | 6.5 | ab | 125 | k |
| 4 | 7.0 | ab | 6.7 | ab | 6.3 | be | 5.5 | hj | 5.4 | i | 6.2 | ce | 146 | f |
| 5 | 6.7 | cd | 6.3 | cf | 5.9 | fg | 6.0 | ce | 5.8 | fg | 6.1 | df | 151 | e |
| 6 | 6.7 | cd | 6.3 | cf | 6.1 | ef | 6.3 | ab | 6.3 | ab | 6.3 | bd | 146 | f |
| 7 | 6.8 | bd | 6.2 | ef | 6.3 | be | 6.1 | bd | 6.1 | cd | 6.3 | bd | 143 | g |
| 8 | 6.8 | bd | 6.6 | ad | 6.2 | de | 5.6 | gi | 5.7 | gh | 6.2 | ce | 151 | e |
| 9 | 6.8 | bd | 6.1 | fg | 6.1 | ef | 5.8 | eg | 6.3 | ab | 6.2 | ce | 152 | de |
| 10 | 6.7 | cd | 6.3 | cf | 6.5 | b | 6.0 | ce | 6.3 | ab | 6.4 | ac | 146 | f |
| 11 | 6.8 | bd | 6.6 | ad | 6.4 | bd | 6.1 | bd | 6.2 | bc | 6.4 | ac | 138 | i |
| 12 | 6.2 | e | 5.9 | g | 5.6 | h | 5.7 | fh | 6.0 | de | 5.9 | f | 145 | fg |
| 13 | 6.7 | cd | 6.3 | df | 6.2 | ce | 6.1 | bd | 6.1 | cd | 6.3 | bd | 146 | f |
| 14 | 6.8 | bd | 6.7 | ab | 6.5 | b | 6.0 | ce | 6.2 | bc | 6.4 | ac | 157 | c |
| 15 | 6.9 | ac | 6.5 | ad | 6.3 | be | 6.4 | a | 6.2 | bc | 6.5 | ab | 138 | i |
| 16 | 6.7 | cd | 6.2 | ef | 6.2 | ce | 5.3 | j | 5.5 | hi | 6.0 | ef | 153 | d |
| 17 | 7.0 | ab | 6.2 | ef | 6.2 | ce | 5.6 | gi | 5.3 | i | 6.0 | ef | 166 | b |
| 18 | 6.8 | bd | 6.4 | be | 6.3 | be | 5.6 | gi | 5.8 | eg | 6.2 | ce | 173 | a |
| 19 | 6.7 | cd | 6.4 | be | 6.2 | ce | 5.8 | eg | 5.5 | hi | 6.1 | df | 141 | h |
| 20 | 6.9 | ac | 6.5 | ad | 6.5 | b | 5.9 | df | 6.2 | bc | 6.4 | ac | 133 | j |
| 21 | 6.8 | bd | 6.3 | df | 6.3 | be | 5.9 | df | 5.9 | ef | 6.2 | ce | 138 | i |
| 22 | 6.8 | bd | 6.3 | df | 5.8 | gh | 5.6 | gi | 5.9 | ef | 6.1 | df | 146 | f |
| 23 | 7.1 | a | 6.5 | ad | 5.7 | gh | 5.7 | fh | 5.8 | eg | 6.2 | ce | 138 | i |
| 24 | 6.8 | bd | 6.3 | cf | 6.1 | ef | 5.4 | ij | 5.7 | fg | 6.1 | df | 143 | g |

Values not having any letter in common are significantly different at $P \leq 0.05$ (Duncan's test)

The extreme variability observed among the accessions compared is due in part to the genotypic characteristics of each accession and in part to an attack of phytoplasma 'BGWL', which resulted in an arrest of the vegetation and the discolouration of the leaf blades especially for accessions 12, 18, 9, 10 and 16.

A prostrate growth habit lowered the values of the cumulative growth for the accessions 2, 3, 4 and 17; on the contrary, accessions that showed the greatest growth were 23 (359 mm) and 21, 1 and 14, all with values ranging around 230 mm.

Table 4 Turf quality of ecotypes during the trial period (2013)

| Samples | May | | June | | July | | August | | September | | Mean May–Sept (2013) | |
|---------|------|------|------|------|------|------|--------|------|-----------|------|-------------------------|------|
| | 1 | 6.01 | cd | 6.02 | de | 6.00 | de | 5.04 | cd | 5.04 | c | 5.08 |
| 2 | 4.09 | k | 4.08 | lm | 4.09 | k | 4.08 | hi | 4.07 | gi | 4.08 | j |
| 3 | 7.02 | a | 7.03 | a | 6.07 | a | 6.02 | a | 6.00 | a | 6.07 | a |
| 4 | 5.03 | hi | 5.04 | hi | 5.03 | j | 5.01 | fg | 5.00 | de | 5.02 | gh |
| 5 | 5.01 | ik | 4.06 | mn | 4.03 | m | 4.06 | jk | 4.07 | fi | 4.07 | j |
| 6 | 5.04 | h | 5.03 | hj | 5.00 | k | 5.02 | ef | 4.09 | df | 5.02 | gh |
| 7 | 4.09 | k | 5.02 | ij | 5.00 | k | 5.07 | b | 4.07 | fi | 5.01 | hi |
| 8 | 5.03 | hj | 5.01 | jk | 5.00 | k | 5.01 | fg | 5.00 | de | 5.01 | hi |
| 9 | 5.08 | f | 4.09 | kl | 4.02 | m | 4.04 | kl | 4.05 | ij | 4.08 | j |
| 10 | 6.01 | cd | 5.08 | g | 5.06 | h | 5.00 | gh | 5.00 | de | 5.05 | ef |
| 11 | 6.00 | ce | 5.09 | fg | 5.00 | k | 5.06 | bc | 5.01 | d | 5.05 | ef |
| 12 | 3.08 | m | 3.05 | o | 2.09 | n | 3.00 | m | 3.03 | l | 3.03 | l |
| 13 | 6.04 | b | 6.02 | cd | 6.01 | cd | 6.00 | a | 5.06 | bc | 6.01 | c |
| 14 | 6.01 | cd | 6.03 | bd | 6.02 | c | 5.06 | bc | 5.05 | bc | 5.09 | d |
| 15 | 5.08 | ef | 6.00 | ef | 5.07 | g | 5.06 | bc | 5.07 | b | 5.08 | d |
| 16 | 4.06 | l | 4.05 | n | 4.02 | m | 4.03 | l | 4.04 | j | 4.04 | k |
| 17 | 4.09 | k | 4.09 | kl | 4.06 | l | 4.07 | ij | 4.09 | dg | 4.08 | j |
| 18 | 5.00 | jk | 5.02 | ij | 4.09 | k | 4.08 | hi | 3.09 | k | 4.09 | ij |
| 19 | 5.05 | gh | 5.03 | hj | 5.03 | ij | 4.09 | hi | 4.08 | eh | 5.01 | hi |
| 20 | 6.01 | cd | 6.01 | df | 5.09 | ef | 5.05 | cd | 5.04 | c | 5.08 | d |
| 21 | 5.07 | fg | 5.08 | g | 5.08 | fg | 4.09 | hi | 5.00 | de | 5.04 | fg |
| 22 | 5.03 | hj | 4.09 | kl | 4.08 | k | 4.04 | l | 4.04 | j | 4.08 | j |
| 23 | 5.03 | hj | 5.04 | hi | 5.03 | ij | 4.09 | hi | 4.09 | df | 5.02 | gh |
| 24 | 5.08 | df | 5.05 | h | 5.05 | hi | 5.03 | de | 4.06 | hi | 5.04 | fg |
| Mean | 5.09 | | 5.02 | | 4.49 | | 4.39 | | 4.29 | | 4.47 | |

Values not having any letter in common are significantly different at $P \leq 0.05$ (Duncan's test)

3.2 Colour Index

Data in Table 3 show for this characteristic a mean value above sufficiency and equal to 6.2.

From May to September 2013, ecotypes 1, 3 and 15 showed higher values; on average this index was equal to 6.5. In contrast accessions that showed the lowest value, equal to 6, were accessions 12, 16 and 17. These results are similar to those obtained in other researches who studied 40 accessions of native *C. dactylon* collected in Sicily (Southern Italy) in order to identify biotypes with good qualities for broadscale turf in the Mediterranean region (Leto et al. 2008).

Table 5 Ground cover percentage of ecotypes during the trial period (2013)

| Samples | Monthly assessed ground cover (%) | | | | | | | | | | Average | |
|---------|-----------------------------------|----|-------|----|-------|----|--------|----|-----------|----|-----------------|----|
| | May | | June | | July | | August | | September | | May–Sept (2013) | |
| 1 | 77 | fg | 82 | fi | 82 | bc | 80 | b | 63 | bd | 77 | d |
| 2 | 68 | ij | 57 | o | 61 | j | 49 | m | 60 | df | 59 | l |
| 3 | 97 | a | 98 | a | 88 | a | 86 | a | 65 | ac | 87 | a |
| 4 | 78 | ef | 67 | lm | 69 | gh | 50 | lm | 51 | ij | 63 | k |
| 5 | 62 | k | 63 | mn | 59 | j | 56 | ij | 57 | fh | 59 | l |
| 6 | 67 | j | 69 | l | 64 | ij | 68 | cf | 58 | eg | 65 | ij |
| 7 | 68 | ij | 61 | no | 62 | j | 62 | gh | 62 | ce | 63 | k |
| 8 | 72 | hi | 71 | kl | 62 | j | 59 | hi | 58 | eg | 64 | jk |
| 9 | 72 | hi | 60 | no | 59 | j | 52 | lm | 52 | ij | 59 | l |
| 10 | 87 | b | 85 | df | 78 | ce | 64 | fg | 58 | eg | 75 | e |
| 11 | 77 | fg | 76 | j | 62 | j | 70 | c | 58 | eg | 69 | h |
| 12 | 22 | m | 22 | p | 18 | k | 19 | n | 28 | k | 22 | n |
| 13 | 97 | a | 94 | ab | 87 | ab | 78 | b | 67 | ab | 84 | b |
| 14 | 87 | b | 84 | eg | 86 | ab | 67 | cf | 57 | fh | 76 | e |
| 15 | 73 | gh | 78 | ij | 70 | gh | 68 | ce | 63 | bd | 71 | g |
| 16 | 48 | l | 63 | mn | 60 | j | 55 | jk | 50 | j | 55 | m |
| 17 | 73 | gh | 71 | kl | 67 | hi | 61 | gh | 58 | eg | 66 | i |
| 18 | 73 | gh | 75 | jk | 67 | hi | 53 | kl | 50 | j | 64 | jk |
| 19 | 83 | bd | 83 | fh | 77 | df | 66 | df | 56 | fh | 73 | f |
| 20 | 87 | b | 87 | de | 78 | ce | 69 | ce | 53 | hj | 75 | e |
| 21 | 79 | df | 80 | gi | 73 | fg | 61 | gh | 60 | df | 71 | g |
| 22 | 86 | bc | 85 | df | 73 | fg | 57 | ij | 55 | gi | 71 | g |
| 23 | 73 | gh | 78 | hj | 75 | ef | 65 | ef | 53 | hj | 69 | h |
| 24 | 82 | ce | 85 | df | 80 | cd | 61 | gh | 58 | eg | 73 | f |
| Mean | 74.5 | | 73.92 | | 69.04 | | 61.5 | | 56.25 | | 67.08 | |

Values not having any letter in common are significantly different at $P \leq 0.05$ (Duncan's test)

3.3 Turf Quality

During the entire period of experimentation, ecotypes that showed the highest mean values were 3, 13 and 14, respectively, with 6.7 to 6.1 and 5.9; they also showed a good preservation of quality from May 2013 to September 2013. Accessions 12 and 16 were ranked at the lowest level, with values respectively equal to 3.3 and 4.4. These extremely low values were due partly to the genotypic characteristics of accession and to a considerable extent of phytoplasma BGWL attacks, which have widely affected the aesthetic quality of ecotypes. The lower values of these ecotypes have strongly influenced the mean of turf quality during the whole period and also for single months.

Table 6 Results of salinity measurements

| Samples | LA | FW | DW | RDW | R/S | L _f | L _f | L _i | Na ⁺ (mg/g) | Cl ⁻ (mg/g) |
|---------|---------|-------|-------|------|------|----------------|----------------|----------------|------------------------|------------------------|
| 1n_s | 523.33 | 24.33 | 5.9 | 0.25 | 0.04 | 52.68 | 3.67 | 65.11 | 2.98 | 6.53 |
| 2n_s | 404.33 | 17 | 3.37 | 1.1 | 0.43 | 33.77 | 3.96 | 53.08 | 2.085 | 6.51 |
| 3n_s | 1049.33 | 57.69 | 10.52 | 3.63 | 0.37 | 36.96 | 3.65 | 50.43 | 1.36 | 7.35 |
| 4n_s | 135 | 11.15 | 2.33 | 1.06 | 0.95 | 39.24 | 3.82 | 63.49 | 1.83 | 10.945 |
| 5n_s | 191 | 17.55 | 3.14 | 1.17 | 0.52 | 24.72 | 4.01 | 33.27 | 1.975 | 9.09 |
| 6n_s | 166.67 | 12.88 | 3 | 0.91 | 0.35 | 24.65 | 3.56 | 47.05 | 1.985 | 7.115 |
| 7n_s | 359 | 37.95 | 6.31 | 2.5 | 1.45 | 44.56 | 4.2 | 35.81 | 1.985 | 7.685 |
| 8n_s | 330.67 | 31.63 | 6.07 | 1.84 | 0.33 | 35.78 | 4.58 | 51.64 | 2.735 | 10.16 |
| 9n_s | 178.67 | 14.11 | 2.72 | 1.41 | 0.52 | 37.98 | 4.37 | 35.03 | 2.255 | 9.465 |
| 10n_s | 264.33 | 26.24 | 5.1 | 3.51 | 0.7 | 47.32 | 4.44 | 48.77 | 1.08 | 8.39 |
| 11n_s | 302 | 28.19 | 5.06 | 2.76 | 0.58 | 50.6 | 4.28 | 66.12 | 2.38 | 9.92 |
| 12n_s | 204 | 16.07 | 2.82 | 1.79 | 0.68 | 37.27 | 3.84 | 48.44 | 1.65 | 12.56 |
| 13n_s | 139 | 14.42 | 2.67 | 2.1 | 0.89 | 44.39 | 5.7 | 49.83 | 1.86 | 21.035 |
| 14n_s | 155.67 | 12.94 | 2.24 | 1.04 | 0.66 | 49.84 | 5 | 53.27 | 2.13 | 8.13 |
| 15n_s | 241.33 | 19.67 | 4.13 | 1.83 | 0.45 | 38.55 | 4.08 | 48.01 | 1.835 | 9.965 |
| 16n_s | 289.33 | 28.3 | 4.61 | 1.54 | 0.38 | 47.18 | 3.76 | 84.19 | 1.31 | 15.235 |
| 17n_s | 136 | 11.21 | 2.91 | 0.17 | 0.06 | 24.53 | 3.19 | 34.12 | 1.22 | 7.725 |
| 18n_s | 140.67 | 11.91 | 2.18 | 1.56 | 0.72 | 26.99 | 2.88 | 30.77 | 1.33 | 10.205 |
| 19n_s | 161.33 | 15.9 | 2.69 | 1.05 | 0.46 | 33.7 | 4.32 | 56.26 | 1.725 | 7.495 |
| 21n_s | 173 | 13.6 | 2.83 | 1.82 | 0.82 | 44.87 | 5.28 | 65.32 | 1.82 | 7.425 |
| 22n_s | 274.33 | 23.57 | 4.54 | 1.45 | 0.67 | 31.79 | 3.33 | 61.34 | 2.065 | 9.81 |
| 23n_s | 251 | 21.17 | 4.02 | 1.67 | 0.64 | 38.03 | 3.59 | 72.4 | 1.57 | 8.935 |
| 24n_s | 295 | 28.24 | 5.12 | 0.9 | 0.18 | 58.31 | 4.25 | 53.49 | 2 | 9.96 |

3.4 Green Cover Percentage

The coverage percentage showed a mean value generally equal to 67 %, in accordance with the findings of Volterrani et al. (1997) and Croce et al. (2001) in other Italian sites. Among the ecotypes compared, the one which showed the highest mean value was ecotype 3 (87 %); satisfactory values were also reported for accessions 13 and 1, with mean values equal to 84 and 77 %.

The lowest value was reached by genotype 12 (22 %). This anomalous value is partly imputable to the massive attack of BGWL, which continued during the entire period of the research.

The results of the experimental salinity carried out in 2013, of the 24 native ecotypes of *C. dactylon*, are shown in Table 6.

The salinity conditions have always had a depressing effect on all measured parameters (leaf area, dry weight, dry weight of roots, root/shoot ratio, etc.) as widely shown in Table 7.

Leaf area on average reduced to 296 cm² from 824 cm², while dry weight on average reduced to 4.5 from 8.1 g plant⁻¹. The significant effect of salinity conditions on all studied accessions and significant difference between accessions as regards the ability to support salts into the nutrient solutions were observed. The interaction effect on leaf area parameter was also significant. Root/shoot ratio on average increased as the effect of salinity conditions, as expected. It increased on average to 0.55 g g⁻¹ from 0.33 g g⁻¹. On this parameter only salinity resulted in a significant effect. The dry weight percentage reduction of all studied accessions was also calculated.

3.4.1 Uptake of Na⁺ and Cl⁻

There was no significant difference between accessions as regards Cl⁻ uptake. As regards Na⁺ leaf tissue concentration on average, it has increased respect to control, 12.0 mg g⁻¹ and 1.2 mg g⁻¹, respectively. It was also observed that the ability to limit the accumulation of sodium ions into leaf tissue varies between the different compared accessions.

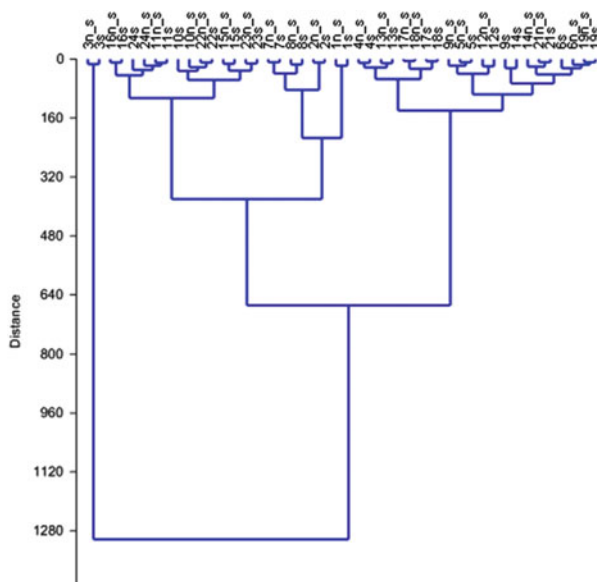
The dendrogram which is obtained from the cluster analysis is a graphical representation of the level of aggregation among the studied accessions (Fig. 4). It clearly shows that the group of accessions on the left is well separated from other groups in the right. The samples on the left showed a greater resistance to salinity. In particular accession 3 is the best ecotype about salinity stress. This ecotype could be considered in future research programmes.

Table 7 Depressing effect of salinity on all measured parameters

| Samples | LA | FW | DW | RDW | R/S | L _f | L _f | Li | Na ⁺ (mg/g) | Cl ⁻ (mg/g) |
|---------|---------|-------|-------|------|------|----------------|----------------|-------|------------------------|------------------------|
| 1s | 523.33 | 24.33 | 5.9 | 0.25 | 0.04 | 52.68 | 3.67 | 65.11 | 16.99 | 27.1 |
| 2s | 404.33 | 17 | 3.37 | 1.1 | 0.43 | 33.77 | 3.96 | 53.08 | 11.88 | 14.4 |
| 3s | 1049.33 | 57.69 | 10.52 | 3.63 | 0.37 | 36.96 | 3.65 | 50.43 | 9.13 | 23.95 |
| 4s | 135 | 11.15 | 2.33 | 1.06 | 0.95 | 39.24 | 3.82 | 63.49 | 10.97 | 21.6 |
| 5s | 191 | 17.55 | 3.14 | 1.17 | 0.52 | 24.72 | 4.01 | 33.27 | 8.785 | 17.5 |
| 6s | 166.67 | 12.88 | 3 | 0.91 | 0.35 | 24.65 | 3.56 | 47.05 | 9.06 | 29.7 |
| 7s | 359 | 37.95 | 6.31 | 2.5 | 1.45 | 44.56 | 4.2 | 35.81 | 6.11 | 18 |
| 8s | 330.67 | 31.63 | 6.07 | 1.84 | 0.33 | 35.78 | 4.58 | 51.64 | 15.475 | 24.65 |
| 9s | 178.67 | 14.11 | 2.72 | 1.41 | 0.52 | 37.98 | 4.37 | 35.03 | 30.3 | 43.65 |
| 11s | 302 | 28.19 | 5.06 | 2.76 | 0.58 | 50.6 | 4.28 | 66.12 | 14.15 | 17.3 |
| 12s | 204 | 16.07 | 2.82 | 1.79 | 0.68 | 37.27 | 3.84 | 48.44 | 18.405 | 33.55 |
| 13s | 139 | 14.42 | 2.67 | 2.1 | 0.89 | 44.39 | 5.7 | 49.83 | 16.215 | 22.9 |
| 14s | 155.67 | 12.94 | 2.24 | 1.04 | 0.66 | 49.84 | 5 | 53.27 | 21.18 | 32.85 |
| 15s | 241.33 | 19.67 | 4.13 | 1.83 | 0.45 | 38.55 | 4.08 | 48.01 | 12.915 | 22.75 |
| 16s | 289.33 | 28.3 | 4.61 | 1.54 | 0.38 | 47.18 | 3.76 | 84.19 | 11.44 | 23.25 |
| 17s | 136 | 11.21 | 2.91 | 0.17 | 0.06 | 24.53 | 3.19 | 34.12 | 11.08 | 30.95 |
| 18s | 140.67 | 11.91 | 2.18 | 1.56 | 0.72 | 26.99 | 2.88 | 30.77 | 9.075 | 34.1 |
| 19s | 161.33 | 15.9 | 2.69 | 1.05 | 0.46 | 33.7 | 4.32 | 56.26 | 6.75 | 17.95 |
| 21s | 173 | 13.6 | 2.83 | 1.82 | 0.82 | 44.87 | 5.28 | 65.32 | 7.41 | 17.75 |
| 22s | 274.33 | 23.57 | 4.54 | 1.45 | 0.67 | 31.79 | 3.33 | 61.34 | 5.1 | 21.7 |
| 23s | 251 | 21.17 | 4.02 | 1.67 | 0.64 | 38.03 | 3.59 | 72.4 | 8.11 | 23.45 |
| 24s | 295 | 28.24 | 5.12 | 0.9 | 0.18 | 58.31 | 4.25 | 53.49 | 9.24 | 36.8 |

Samples 1–24n_s = samples without salt; Samples 1–24 s = samples with salt; Measured parameters: LA leaf area, FW fresh weight, DW dry weight, RDW root dry weight, R/S root/shoot ratio, l_f leaf length, L_f leaf width, l_i distance between internodes, [Na⁺] leaf tissue concentration, [Cl⁻] leaf tissue

Fig. 4 Tree based on the relationship between morphometric data, aesthetic, chemical and salt stress among 24 autochthonous Bermuda grass ecotypes with Ward's method and Euclidean linkage. Note: Samples 1–24n_s = samples without salt; Samples 1–24_s = samples with salt. Measured parameters: *LA* leaf area, *FW* fresh weight, *DW* dry weight, *RDW* root dry weight, *R/S* root/shoot, *l_f* leaf length, *L_f* leaf width, *l_i* distance between internodes, $[Na^+]$, $[Cl^-]$



4 Conclusions

The research project was of particular interest in increasing knowledge on Bermuda grass germplasm in Central and Southern Italy.

The bio-agronomic evaluation of 24 ecotypes of *Cynodon dactylon* (L.) Pers. showed high variability in the collected germplasm as far as the biometric and qualitative parameters were concerned.

Many of the ecotypes compared resulted susceptible to phytoplasma BGWL attacks, which influenced all the considered parameters with the exception of ecotypes 1, 3, 13, 14, 21, 23 and 24. Susceptibility to phytoplasma is a factor to be considered for the selection of ecotypes for turf.

The native accession 3 from Vasto (Abruzzo Region) achieved the best results among native accessions. For some parameters, several ecotypes (1, 13, 14 and 20) showed good biometric and qualitative performances and could result in great interest for selecting future Bermuda grass cultivars for turfgrass use.

As can be seen clearly from the data shown, only some of the ecotypes studied showed a reduction of leaf length (*l_f*), leaf width (*L_f*) and distance between internodes (*l_i*), at least 50 %, and in some cases any reduction in the growth rate as in the case of the ecotypes 1, 3, 6 and 17 was not observed.

Further studies are needed to better characterise the above native accessions of *C. dactylon*. in order to promote them in the turfgrass market and to assess the potential benefits compared to alien species in the Mediterranean environments.

Acknowledgements Findings in this paper are the preliminary results of research work financially supported by the Italian Ministry of Agriculture (Mi.T.E.A.Med project). The authors wish to thank Aleksandra Dimitrijevic and Mauro Musto for their linguistic revisions, which improved the clarity of the manuscript.

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