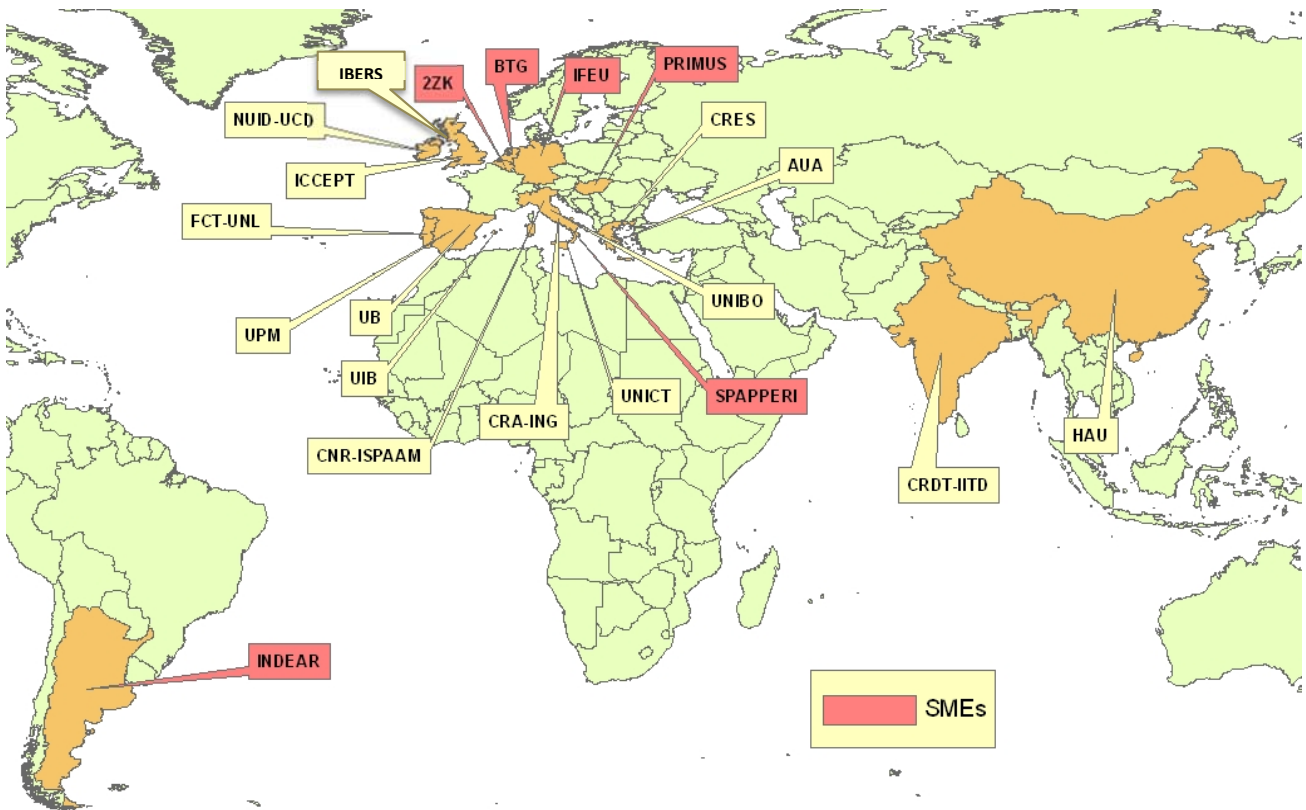


## OPTIMA – Attachment to Final report

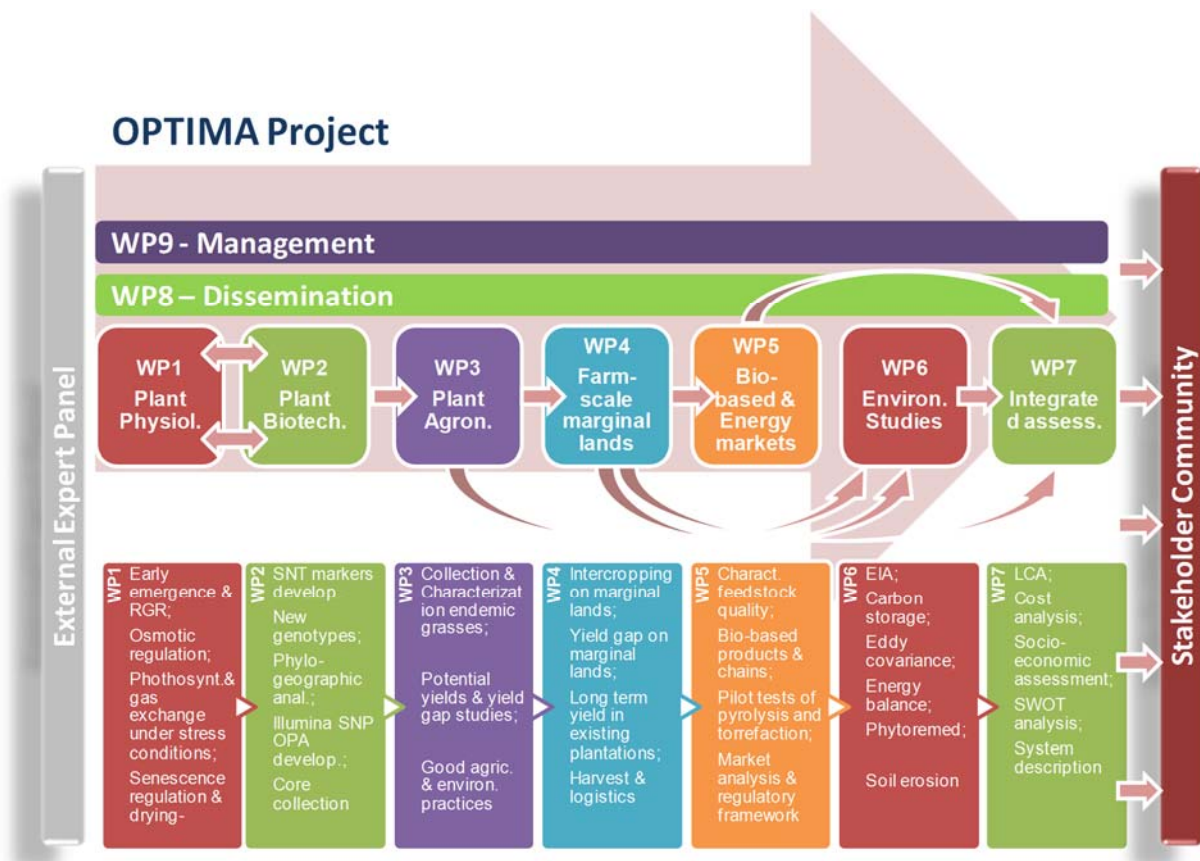


### OPTIMA Logo



### OPTIMA consortium



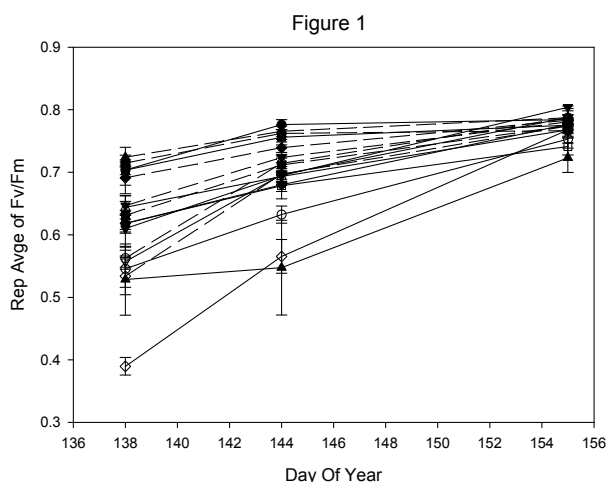


OPTIMA diagram WP flow

### WP1 – Plant and leaf physiology

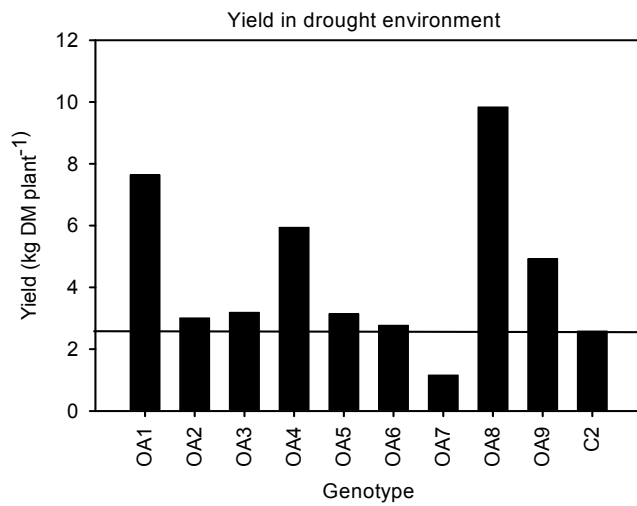
#### Task 1.1 Physiology of switchgrass and miscanthus (*UCD, INDEAR, IBERS, PRIMUS*)

Aberystwyth University – AU IBERS (n. 11)



**Figure 1 – IBERS.** Progression of early season stress in selected genotypes demonstrating a range of tolerance and sensitivity.





**Figure 2 - IBERS.** Variation in selected genotypes for biomass accumulation under field water stressed conditions used to identify genotypes exhibiting tolerance and sensitivity to drought stress.

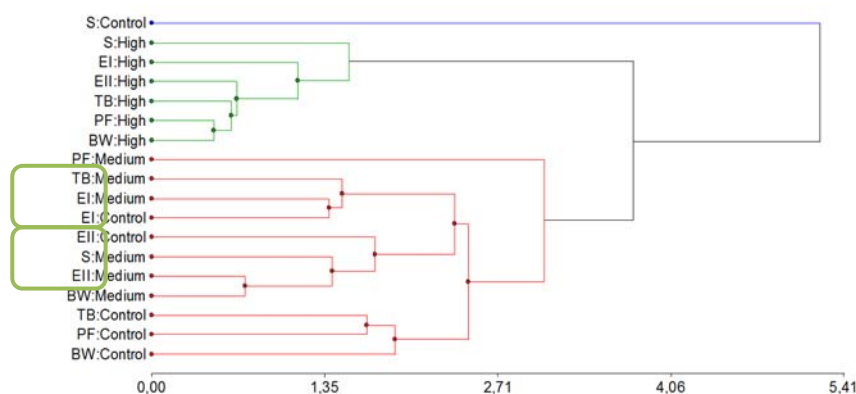
## Instituto of Agrobiotechnology Rosario – INDEAR (n. 17)

**Table 1 – INDEAR.** Survival percentage of *P. virgatum* seedlings after two weeks of treatment with gradually salinized nutrient solution in hydroponics.

| Cultivar | NaCl (mM) | Survival (%) |
|----------|-----------|--------------|
| EI       | 150       | 85,71        |
| EII      | 150       | 85,71        |
| TB       | 150       | 85,71        |
| BW       | 150       | 42,86        |
| S        | 150       | 42,86        |
| PF       | 150       | 14,29        |



**Figure 1 – INDEAR.** *P. virgatum* seedlings under salt evaluation under gradually salinized nutrient solution in hydroponics conditions.

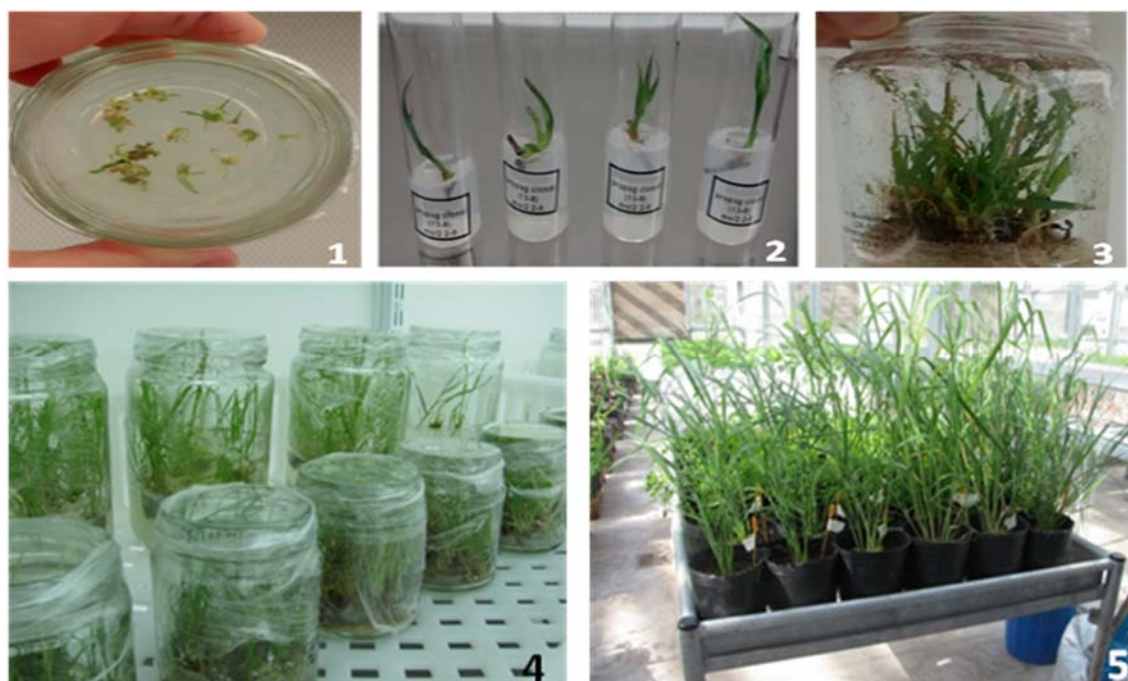


**Figure 2 – INDEAR.** Cultivar and treatment clustering after multivariate analysis.

**Table 2 – INDEAR.** *P* values for significance levels, after ANOVA for various physiological parameters.

| Source               | Photo- synthesis | Stomatalconductance | Transpirationrate | Quantum yield | SPAD   |
|----------------------|------------------|---------------------|-------------------|---------------|--------|
| Cultivar             | 0,2405           | 0,2421              | 0,0075            | 0,8169        | 0,1599 |
| Treatment            | 0,0917           | 0,0002              | <0,0001           | 0,0243        | 0,9401 |
| Cultivar x Treatment | 0,2878           | 0,4025              | 0,1838            | 0,08066       | 0,4867 |





**Figure 3 – INDEAR. Stages for *in vitro* direct regeneration of *P. virgatum* plants of different switchgrass cultivars (EI, EII, S, BW, A).** 1, Mature caryopses as explants for plant direct regeneration; 2, young shoots; 3-4, shoot proliferation under *in vitro* conditions; 5, hardening of propagated plants in the greenhouse of INDEAR for water stress experiments.

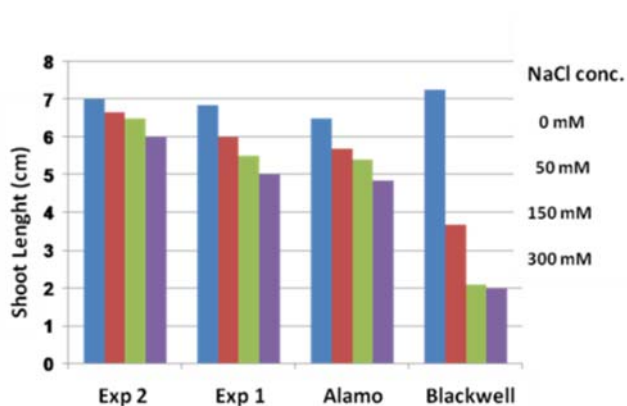
**Table 4 – INDEAR. Callus Induction Frequency (CIF) on MS media supplemented with different concentration of Sodium Chloride.** The values having the same letter following are not significantly different at 0.01 probability level by Least Significant Difference Test (LSD).

| Genotype | Salt concentration (NaCl mM) |                    |                    |                     |                  |
|----------|------------------------------|--------------------|--------------------|---------------------|------------------|
|          | 0                            | 50                 | 100                | 150                 | 300              |
| EI       | 90,62 <sub>a</sub>           | 75,96 <sub>d</sub> | 39,66 <sub>f</sub> | 15,77 <sub>ij</sub> | 2,3 <sub>l</sub> |
| EII      | 86,44 <sub>b</sub>           | 68,44 <sub>e</sub> | 37,25 <sub>g</sub> | 12,13 <sub>j</sub>  | 0                |
| S        | 90,31 <sub>a</sub>           | 75,30 <sub>d</sub> | 32,63 <sub>g</sub> | 14,17 <sub>j</sub>  | 0                |
| BW       | 94,70 <sub>a</sub>           | 31,34 <sub>f</sub> | 11,79 <sub>h</sub> | 3,9 <sub>k</sub>    | 0                |
| Mean     | 90,51                        | 62,76              | 30,33              | 11,49               | 0,575            |



**Table 5 – INDEAR. Chlorophyll content (mg/g fresh weight)** calculated by a spectrophotometer in leaf sections of four different switchgrass materials after incubation with different concentrations of NaCl. EI and EII, Experimental I and II; A, Alamo; BW, Blackwell. The Table shows the rate data generated after two independent experiments. Each experiment included 20 leaf sections and 3 replicates (n=3). Different capital letters indicate significant differences (p<0.05) between switchgrass cultivars; different Non-capital letters indicate significant differences (p<0.05) between NaCl concentrations for the same *Panicum* cv.

|        | BW      | A       | EII     | EI      |
|--------|---------|---------|---------|---------|
| 0 mM   | 1,2 aA  | 1,4 aA  | 1,1 aA  | 1,25 aA |
| 50 mM  | 0,8 bB  | 1,37 aA | 1 aA    | 1,1 AA  |
| 150 mM | 0,5 cC  | 1 bA    | 0,86 aB | 0,9 aA  |
| 300 mM | 0,29 dD | 0,57 cC | 0,54 bC | 0,59 bC |

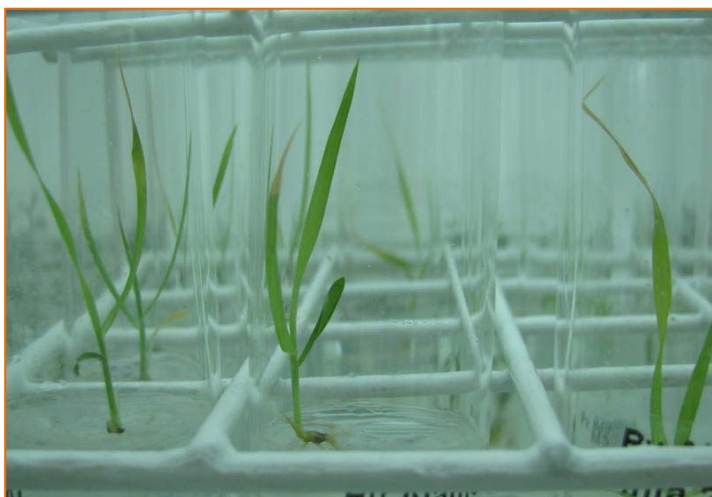


**Figure 4 - INDEAR.** Stem/shoot length (cm) after culture on media containing different concentrations of NaCl (0 mM in blue; 50 mM in red; 150 mM in green; 300 mM in purple). While non-significant differences were observed among Experimental 1 (Exp1 or EI), Experimental 2 (Exp 2 or EII) and Alamo (A), Blackwell (BW) cvs showed an arrested growth in the presence of NaCl. The Figure shows the effect of different concentrations of NaCl on stem elongation (cm) after culture for 9 weeks.

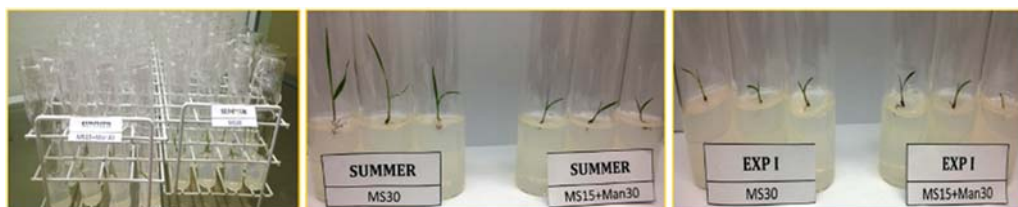


**Table 6 – INDEAR. *In vitro* shoot growth under osmotic stress with PEG.** SD, significant difference. Different letters indicate significant differences between treatments and between cultivars ( $P < 0,05$ ).

| Switchgrass Cv | PEG % | Heigh (cm) | Leaves (#) |
|----------------|-------|------------|------------|
| EI             | 0     | 12.9 a     | 9.6 ab     |
|                | 10    | 11.4 ab    | 8.3 abcd   |
|                | 15    | 11.1 ab    | 9.2 ab     |
|                | 20    | 11.6 ab    | 9.2 ab     |
| EII            | 0     | 10.2 ab    | 9.3 ab     |
|                | 10    | 11.3 ab    | 8.4 abcd   |
|                | 15    | 12.1 ab    | 8.8 abc    |
|                | 20    | 11.0 ab    | 10.7 ab    |
| S              | 0     | 6.2 de     | 4.0 1bcd   |
|                | 10    | 6.3 de     | 2.2 f      |
|                | 15    | 6.8 cde    | 4.2 def    |
|                | 20    | 5.7 e      | 4.6 cdef   |
| A              | 0     | 6.7 cde    | 6.6 bcde   |
|                | 10    | 9.1 bcd    | 11.3 a     |
|                | 15    | 9.5 bc     | 8.4 abcd   |
|                | 20    | 9.4 bc     | 8.8 abc    |
| SD 0.05        |       | 3.0        | 4.2        |



**Figure 5 – INDEAR. *In vitro* plant growth and development under osmotic stress with PEG.**



**Figure 6 – INDEAR. *In vitro* osmotic stress experiment with mannitol: representative photographs of plant material under evaluation.**

**Table 7 – INDEAR.** *In vitro* osmotic stress experiment with mannitol: Stem/shoot length and number of leaves were measured after 3 weeks. No significant differences were observed between the assayed treatments for both switchgrass cvs.

|                    | Summer          |                           | EI              |                           |
|--------------------|-----------------|---------------------------|-----------------|---------------------------|
|                    | Shoots (number) | Shoot lenght (avg. in cm) | Shoots (Number) | Shoot lenght (avg. in cm) |
| Control Treatment  | 20              | 3.37                      | 13              | 1.5                       |
| Mannitol Treatment | 9               | 1.50                      | 8               | 0.94                      |

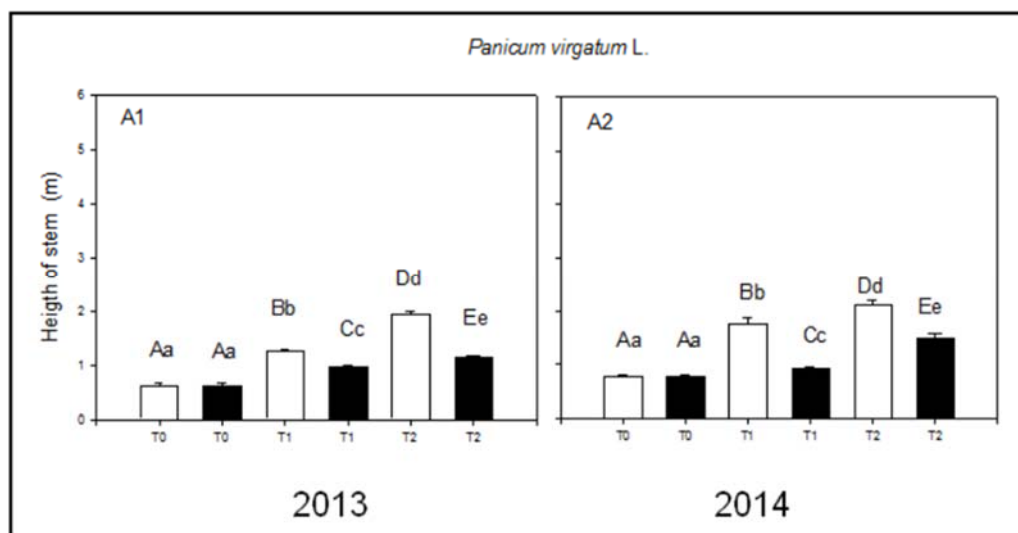
**Table 8 – INDEAR.** Survival percentage of *P. virgatum* clonally propagated plants after 60 days of treatment with suspended watering and after flooding treatment. Values are the rate of 10 replicates. Data followed by the same letter do not present significant differences ( $P < 0,05$ ).

| Cultivar | Survival (%)<br>No watering | Survival (%) Flooding |
|----------|-----------------------------|-----------------------|
| EI       | 65,51a                      | 13,5a                 |
| EII      | 72,33b                      | 22,2b                 |
| BW       | 25,26c                      | 10,5a                 |
| S        | 72,26b                      | 12,6a                 |
| A        | 45,65d                      | 32,5d                 |

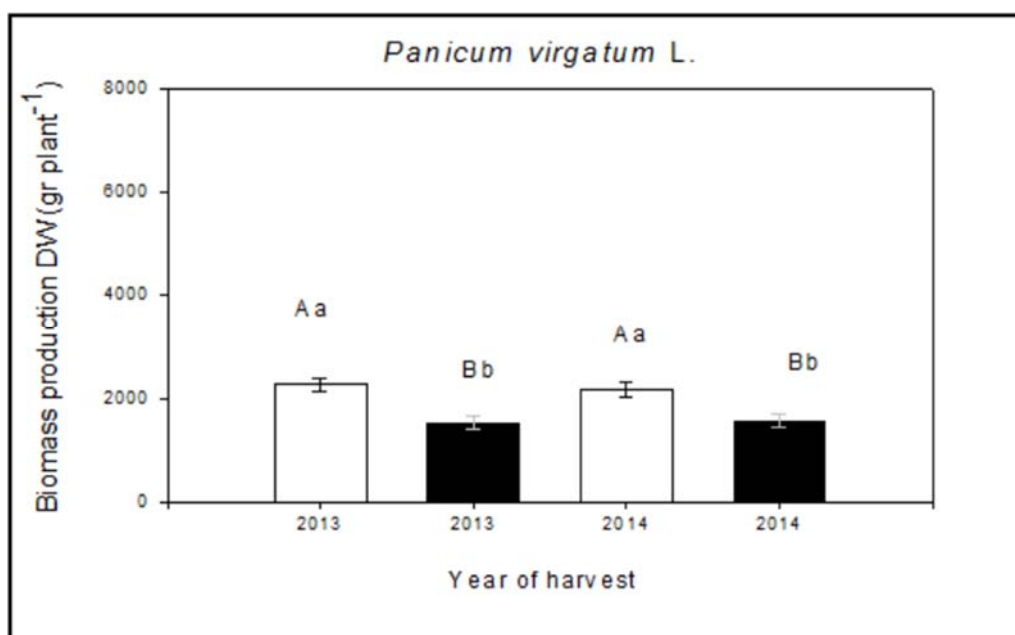


**Figure 7 – INDEAR.** *Panicum* plants in the greenhouse at the beginning of a controlled experiment for the evaluation of plant survival % after 60 days of suspended watering.



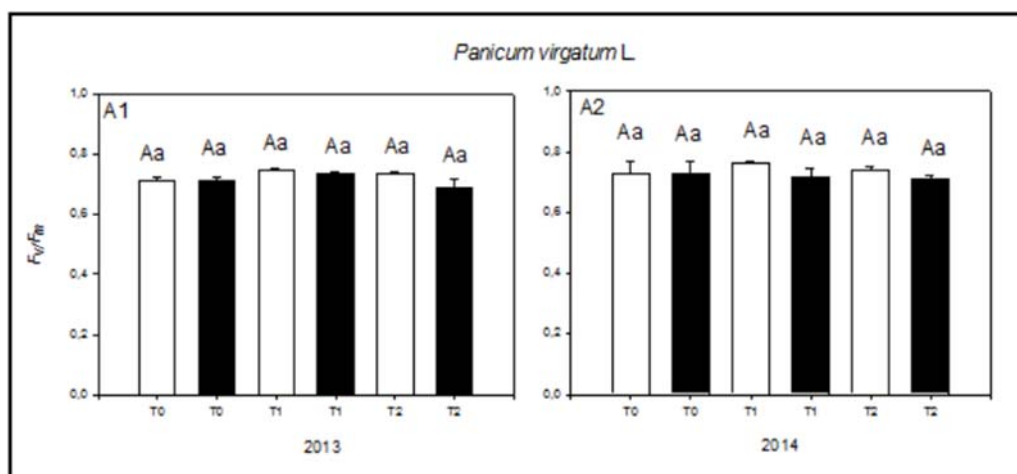


**Figure 8 – INDEAR/UB. Plant Stem length (m) under normal water rainfall regime and with irrigation.** Data were taken at different moments in the year: T0, spring time; T1, early summer and T2, summer end. White bars: irrigation treatment; black bars: no irrigation. A1, Plant length (m) in *P. virgatum* L. plant in 2013; A2, data for 2014. Bars and values are the rate  $\pm$  SE of 4 replicates ( $n=4$ ). In the same year, capital letters indicate significant differences ( $P < 0,05$ ) between the data registration time for the same treatment. Non-capital letters indicate differences ( $P < 0,05$ ) between treatments in the same year and data registration time.

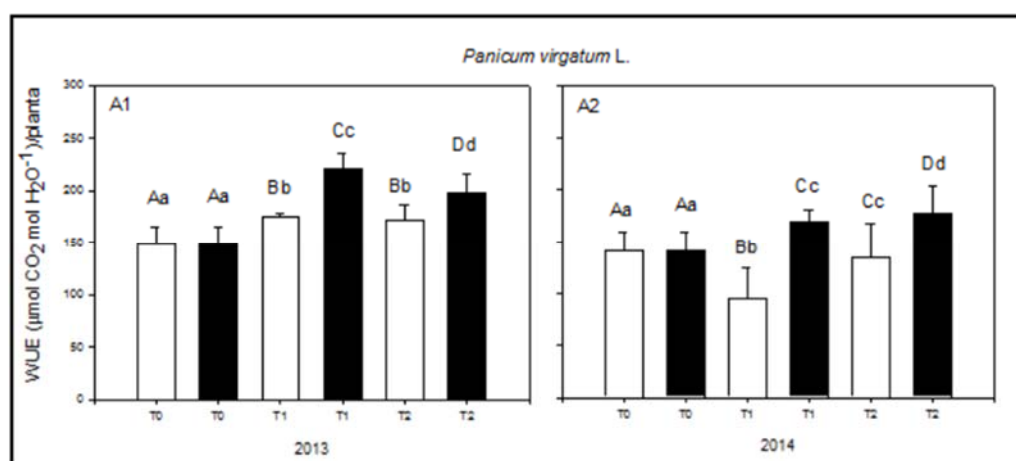


**Figure 9– INDEAR/UB. Biomass production: dry Weight (g) of aerial part of the plants grown under normal waterfall regime and with irrigation, during 2013 and 2014.** A, DW (Peso Seco, PS) in g for *P. virgatum* L. White bars: irrigation; black bars: no irrigation. Bars and values are the rate  $\pm$  SE of 4 replicates ( $n=4$ ). Capital letters indicate significant differences ( $P < 0,05$ ) between years for the same treatment and non-capital letters indicate differences ( $P < 0,05$ ) between treatments within the same year.





**Figure 10– INDEAR/UB. Maximum Quantum Yield of Photosystem II ( $F_v/F_m$ ) in *Panicum virgatum* L.** plants grown under normal waterfall regime and watering conditions at different moments (T0: spring; T1: early summer and T2: summer end). White bars: irrigation treatment; black bars: no irrigation. Bars and values are the rate  $\pm$  SE of 3 replicates ( $n=3$ ). In the same year, capital letters indicate significant differences ( $P<0,05$ ) between the data registration time for the same treatment. Non-capital letters indicate differences ( $P < 0,05$ ) between treatments in the same data registration time.



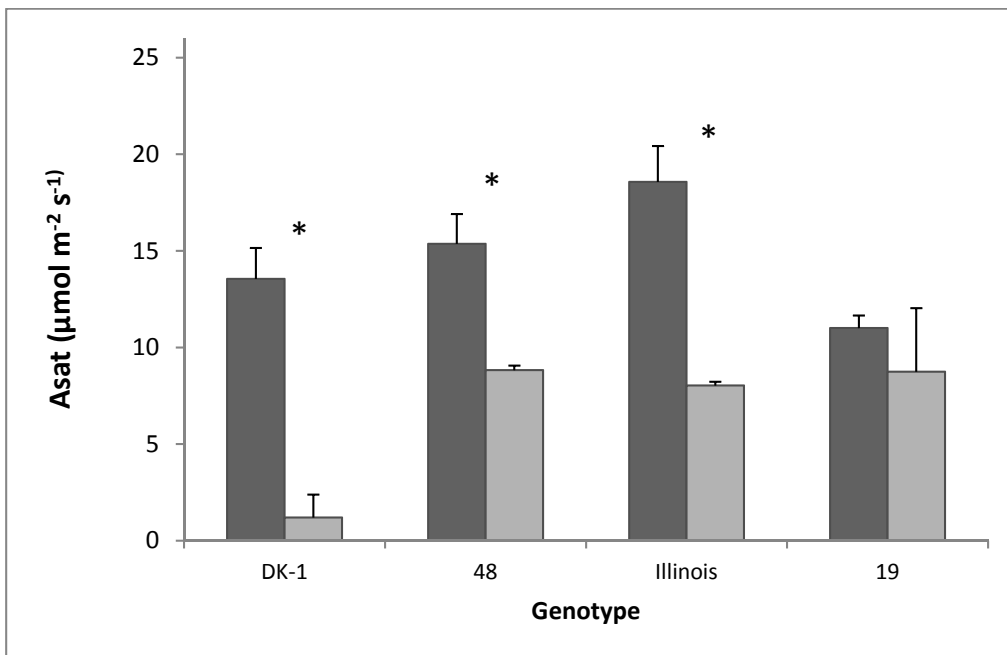
**Figure 11– INDEAR/UB. Water Use Efficiency, WUE ( $\mu\text{mol CO}_2 \text{ mol H}_2\text{O}^{-1}$ ) in *Panicum virgatum* L.** plants grown under normal waterfall regime and watering conditions at different moments (T0: spring; T1: early summer and T2: summer end). White bars: Irrigation treatment; black bars: no irrigation. Bars and values are the rate  $\pm$  SE of 4 replicates ( $n=4$ ). In the same year, capital letters indicate significant differences ( $P<0,05$ ) between the data registration time for the same treatment. Non-capital letters indicate differences ( $P < 0,05$ ) between treatments in the same data registration time.



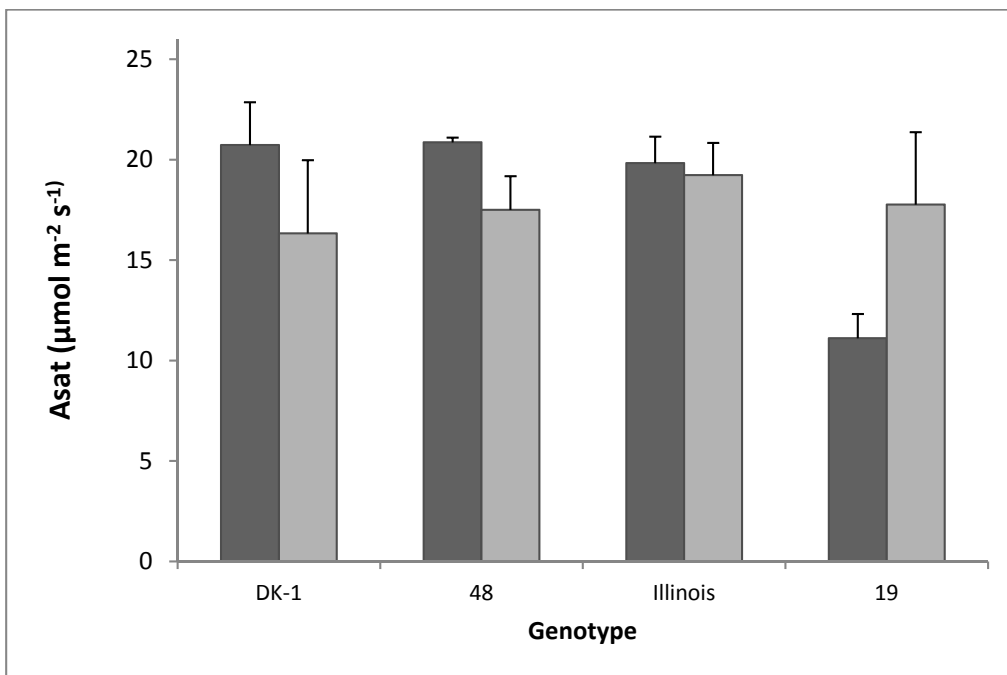
**Table 9 – INDEAR/UB. Photosynthetic parameters:** Net CO<sub>2</sub> Accumulation Rate at light saturation ( $A_{sat}$ ), Maximum Net Photosynthetic Rate ( $A_{max}$ ), stomatal conductance ( $g_s$ ), measured in *Panicum virgatum* L. plants grown under normal waterfall regime and with irrigation. Data were taken at three moments in the year: (T0: spring; T1: early summer and T2: summer end). Values are the media  $\pm$  SE of three replicates ( $n=3$ ). Different capital letters indicate significative differences ( $P < 0,05$ ) between years for the same parameter, time and treatment. Non-capital letters show significative differences ( $P < 0,05$ ) between times for the same parameter and year.

| Panicum virgatum L.                                                         |      |            |       |          |       |          |
|-----------------------------------------------------------------------------|------|------------|-------|----------|-------|----------|
| Parameters                                                                  | Time | Treatment  | 2013  |          | 2014  |          |
| A <sub>sat</sub><br>(μmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> ) | T0   | Irrigation | 19,33 | ±1,45Aa  | 17,00 | ±1,00Aa  |
|                                                                             | T0   | Rainfall   | 19,33 | ±1,45Aa  | 17,00 | ±1,00Aa  |
|                                                                             | T1   | Irrigation | 21,00 | ±1,50Aa  | 19,83 | ±3,35Aa  |
|                                                                             | T1   | Rainfall   | 4,50  | ±0,50Bb  | 5,07  | ±1,39Bb  |
|                                                                             | T2   | Irrigation | 15,50 | ±2,18Cc  | 16,17 | ±1,17Cc  |
|                                                                             | T2   | Rainfall   | 3,17  | ±0,93Bb  | 5,33  | ±0,17Bb  |
| A <sub>max</sub><br>(μmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> ) | T0   | Irrigation | 26,03 | ±3,24Aa  | 29,15 | ±2,15Bb  |
|                                                                             | T0   | Rainfall   | 26,03 | ±3,24Aa  | 29,15 | ±2,15Bb  |
|                                                                             | T1   | Irrigation | 26,60 | ±0,30Aa  | 25,57 | ±2,44Aa  |
|                                                                             | T1   | Rainfall   | 6,80  | ±0,34Cc  | 7,42  | ±2,14Cc  |
|                                                                             | T2   | Irrigation | 19,90 | ±2,50Dd  | 26,23 | ±2,02Aa  |
|                                                                             | T2   | Rainfall   | 6,40  | ±1,02Cc  | 7,92  | ±1,79Cc  |
| g <sub>s</sub><br>(mol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> )   | T0   | Irrigation | 0,130 | ±0,010Aa | 0,120 | ±0,010Aa |
|                                                                             | T0   | Rainfall   | 0,130 | ±0,010Aa | 0,120 | ±0,010Aa |
|                                                                             | T1   | Irrigation | 0,120 | ±0,004Aa | 0,210 | ±0,005Bb |
|                                                                             | T1   | Rainfall   | 0,014 | ±0,010Cc | 0,030 | ±0,010Cc |
|                                                                             | T2   | Irrigation | 0,090 | ±0,003Dd | 0,120 | ±0,003Aa |
|                                                                             | T2   | Rainfall   | 0,016 | ±0,004Cc | 0,030 | ±0,010Cc |



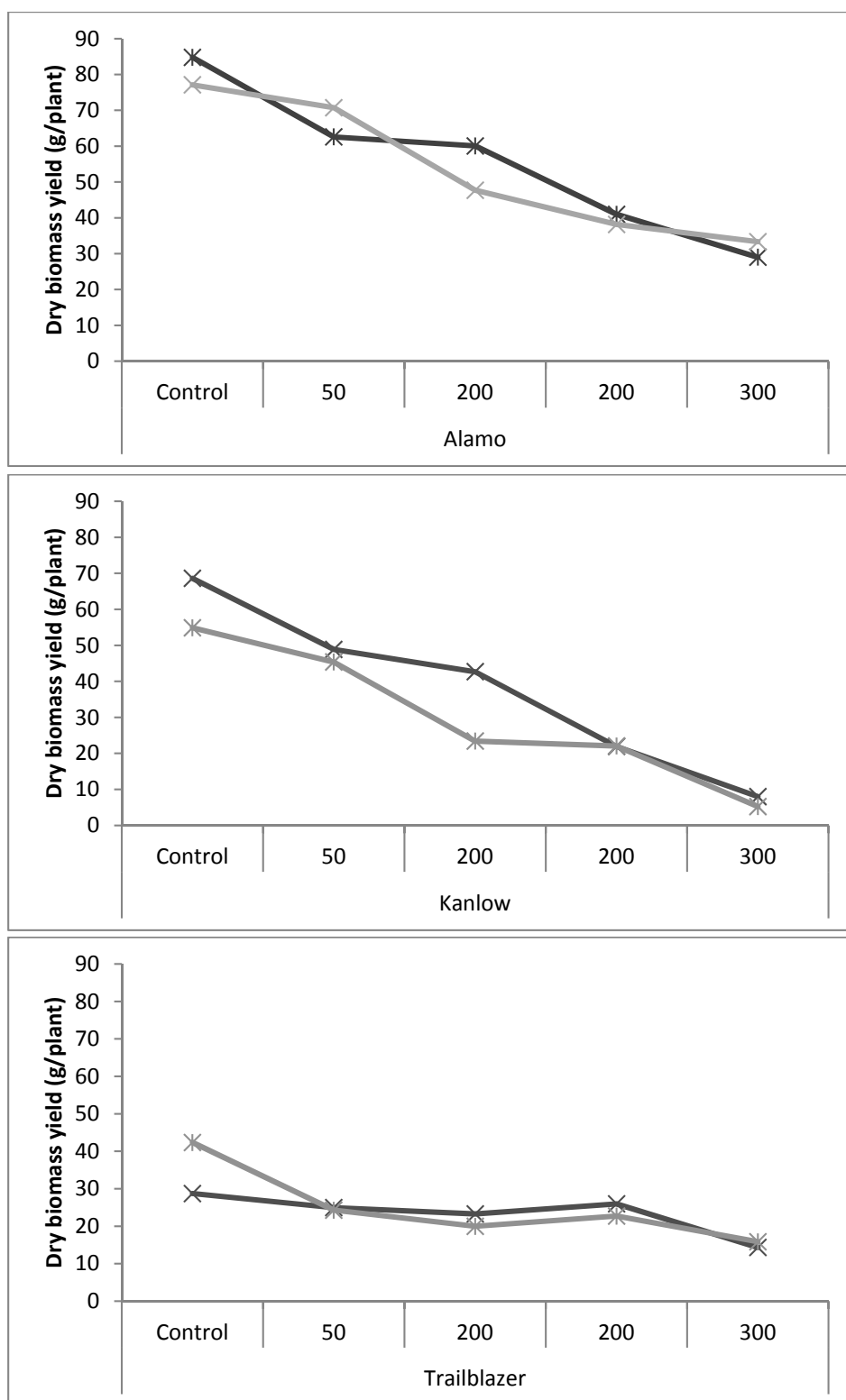


**Figure 1 – UCD.** Leaf-level  $\text{CO}_2$  assimilation rate (Asat) measured at  $400 \mu\text{mol mol}^{-1} \text{CO}_2$ ,  $2000 \mu\text{mol photon m}^{-2} \text{s}^{-1}$  and  $25 \pm 2^\circ \text{C}$  ( $n=3$ ) in control (black bars) and plants exposed to a ten day low temperature exposure (grey bars). Values represent mean  $\pm \text{SE}$ . Within each genotype, values that are significantly different are identified with \* ( $p \leq 0.05$ ).



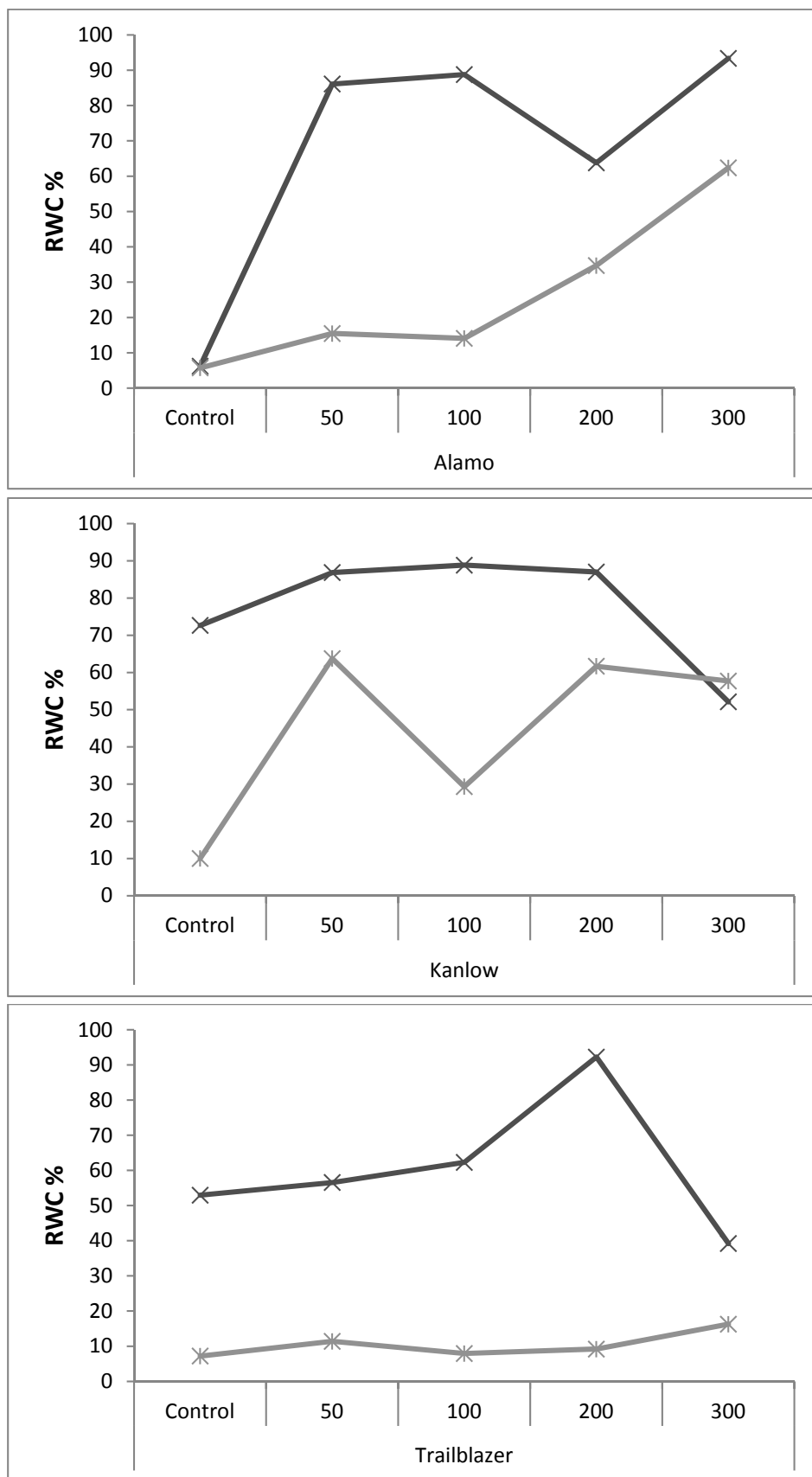
**Figure 2 - UCD.** Leaf-level  $\text{CO}_2$  assimilation rate (Asat) measured at  $400 \mu\text{mol mol}^{-1} \text{CO}_2$ ,  $2000 \mu\text{mol photon m}^{-2} \text{s}^{-1}$  and  $25 \pm 2^\circ \text{C}$  ( $n=3$ ) in control (black bars) and plants exposed to a ten day low temperature treatment (grey bars) on day 15 (recovery). Values represent mean  $\pm \text{SE}$ . Within each genotype, values that are significantly different are identified with \* ( $p \leq 0.05$ ).





**Figure 3 - UCD.** Dry biomass yield (g/plant) in *Panicum virgatum* var. Alamo, Kanlow and Trailblazer (n=3) after 7 (black) and 9 (grey) months of growth under different salinities (mM).



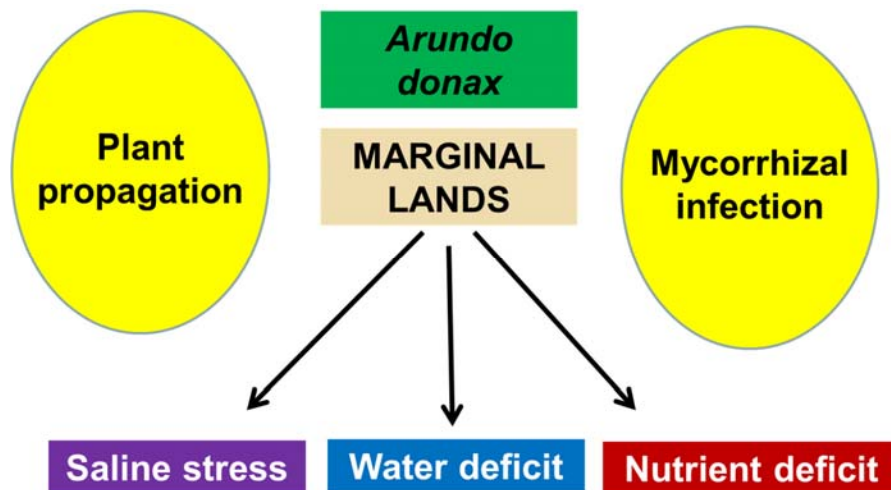


**Figure 4 - UCD.** Relative water content (RWC - %) in *Panicum virgatum* var. Alamo, Kanlow and Trailblazer (n=3) after 7 (black) and 9 (grey) months of growth under different salinities (mM).



## Task 1.2 Physiology of giant reed (*UIB*, *UB*)

Universitat de Les Illes Balears – UIB (n. 9)

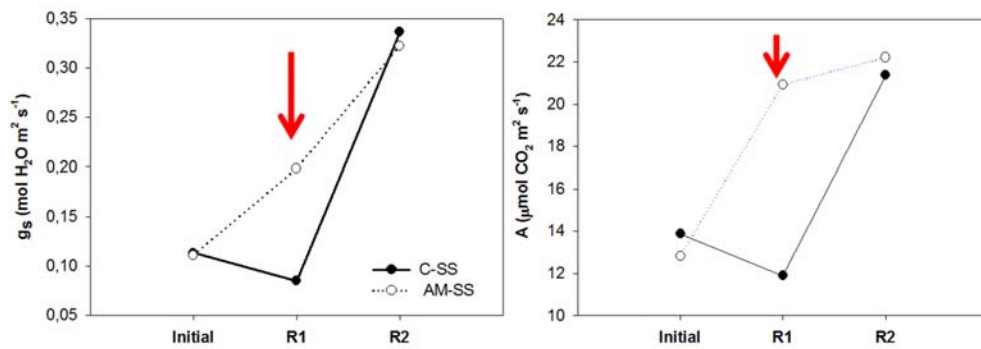


**Figure 1 - UIB.** Activities of Task 1.2 in which Partner 9 has been involved during 2013-14.

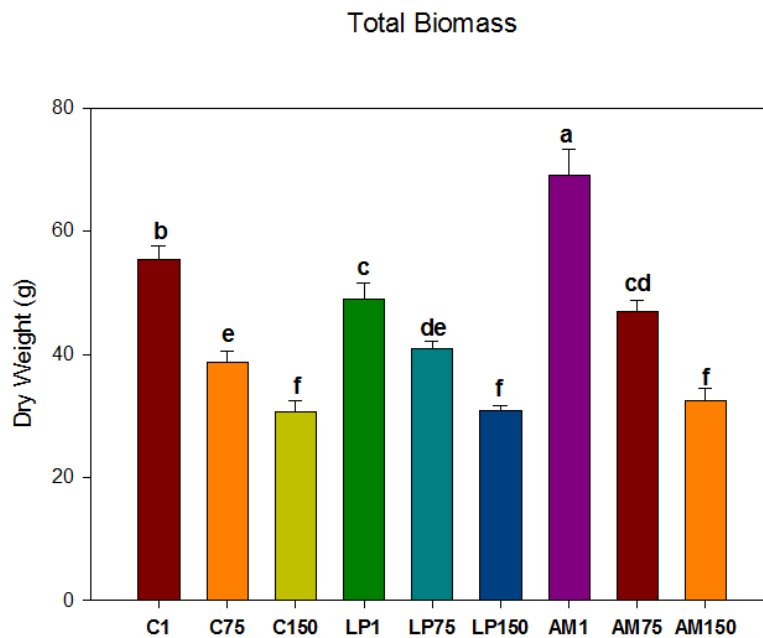
**Table 1 - UIB.** Biomass partitioning (grams, g) of inoculated (AM) and non-inoculated (C) *Arundo donax* plants under well-watered (WW), mild drought (MS) and severe drought (SS) conditions. Values are means of 6 replicates  $\pm$  standard error. Different letters indicate significant differences at  $p < 0.05$

|       | LEAVES                 | STEM                   | ROOT                    | RIZOME                 | SHOOT                  | S/R                    | TB                    | LA (cm <sup>2</sup> )   |
|-------|------------------------|------------------------|-------------------------|------------------------|------------------------|------------------------|-----------------------|-------------------------|
| C-WW  | 61,48<br>$\pm 2,30$ A  | 89,76<br>$\pm 2,13$ A  | 110,74<br>$\pm 11,04$ A | 156,89<br>$\pm 5,68$ A | 151,24<br>$\pm 4,01$ A | 0,569<br>$\pm 0,02$ AB | 418,9<br>$\pm 14,4$ A | 754,1<br>$\pm 52,0$ A   |
| C-MS  | 44,62<br>$\pm 2,41$ B  | 63,48<br>$\pm 2,76$ B  | 80,39<br>$\pm 5,52$ B   | 110,09<br>$\pm 7,48$ B | 108,10<br>$\pm 4,27$ B | 0,584<br>$\pm 0,06$ A  | 298,6<br>$\pm 10,6$ B | 521,1<br>$\pm 48,3$ AB  |
| C-SS  | 38,08<br>$\pm 0,52$ CD | 51,21<br>$\pm 2,35$ C  | 79,50<br>$\pm 3,69$ B   | 107,85<br>$\pm 5,18$ B | 89,29<br>$\pm 2,50$ C  | 0,479<br>$\pm 0,016$ B | 276,6<br>$\pm 9,0$ BC | 428,2<br>$\pm 23,0$ AB  |
| AM-WW | 64,00<br>$\pm 2,60$ A  | 93,10<br>$\pm 5,97$ A  | 108,67<br>$\pm 4,65$ A  | 160,73<br>$\pm 3,03$ A | 157,09<br>$\pm 8,27$ A | 0,584<br>$\pm 0,03$ A  | 426,5<br>$\pm 11,9$ A | 743,4<br>$\pm 46,1$ AB  |
| AM-MS | 42,92<br>$\pm 1,77$ B  | 55,33<br>$\pm 4,93$ BC | 82,50<br>$\pm 4,08$ B   | 108,13<br>$\pm 4,61$ B | 98,25<br>$\pm 6,56$ BC | 0,515<br>$\pm 0,03$ AB | 288,9<br>$\pm 11,0$ B | 510,02<br>$\pm 69,99$ B |
| AM-SS | 36,52<br>$\pm 2,15$ D  | 50,97<br>$\pm 4,76$ C  | 70,44<br>$\pm 2,86$ B   | 98,21<br>$\pm 3,11$ B  | 87,49<br>$\pm 6,87$ C  | 0,522<br>$\pm 0,05$ AB | 256,1<br>$\pm 7,3$ C  | 414,9<br>$\pm 31,4$ AB  |



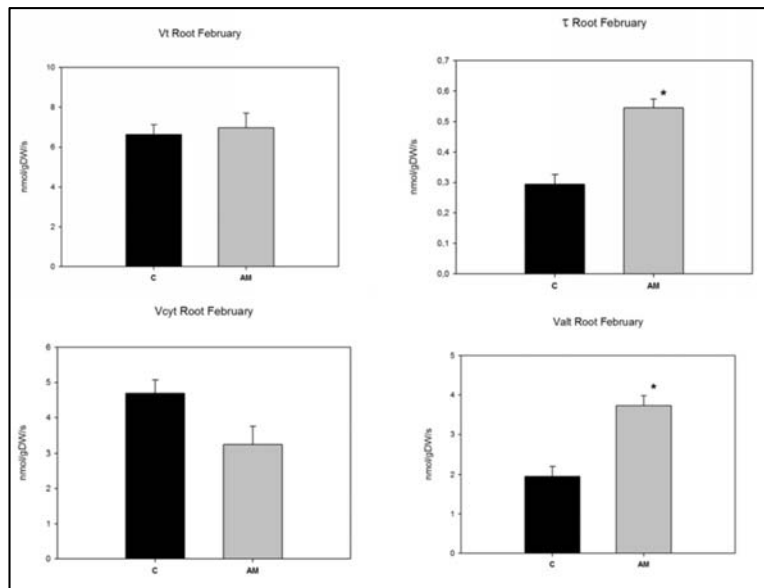


**Figure 2 - UIB.** Stomatal conductance and net photosynthesis of inoculated (AM) and non-inoculated (C) *Arundo donax* plants under severe drought (SS) conditions just before rewatering (initial) and 1 (R1) and 2 (R2) days after rewatering. Values are means of 6 replicates  $\pm$  standard error.



**Figure 3 - UIB.** Total dry biomass of inoculated (AM, LP) and non-inoculated (C) *Arundo donax* plants at three salinity levels (1, 75 and 150 mM NaCl). C and AM plants were grown at 30 mg P/I, LP plants were grown at 2 mg P/I. Values are means of 6 replicates  $\pm$  standard error. Different letters indicate significant differences at  $p < 0.05$ .



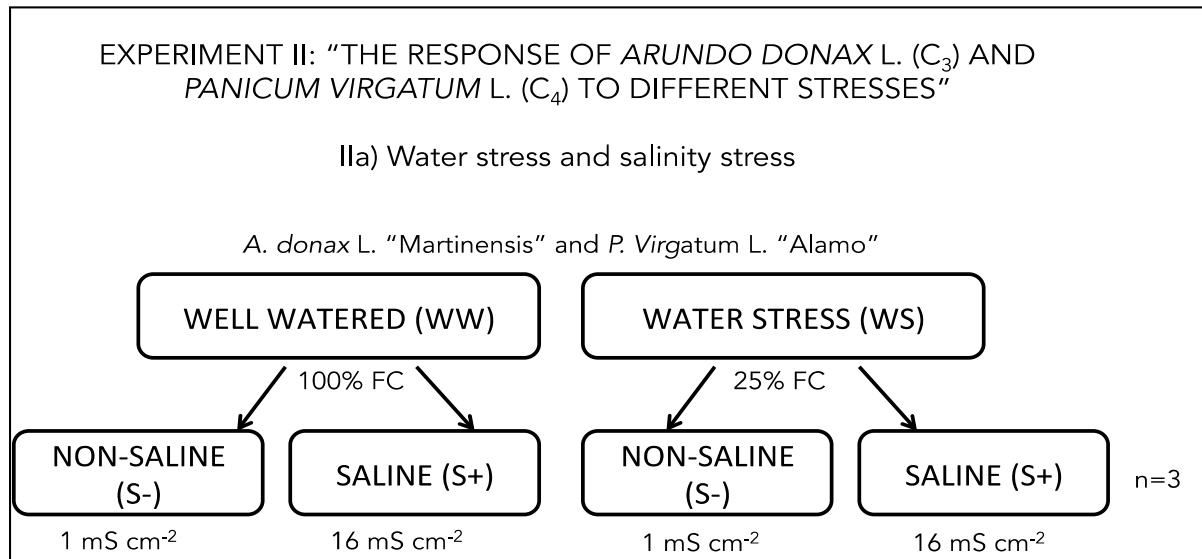


**Figure 4 - UIB.** Total respiration (Vt), electron partitioning to the alternative pathway (Ta), cytochrome pathway activity (vcyt), and alternative pathway activity (valt) in non-inoculated (C) and inoculated (AM) *Arundo donax* plants. Values are means of 5 replicates  $\pm$  standard error. \* indicates significant differences at  $p < 0.05$

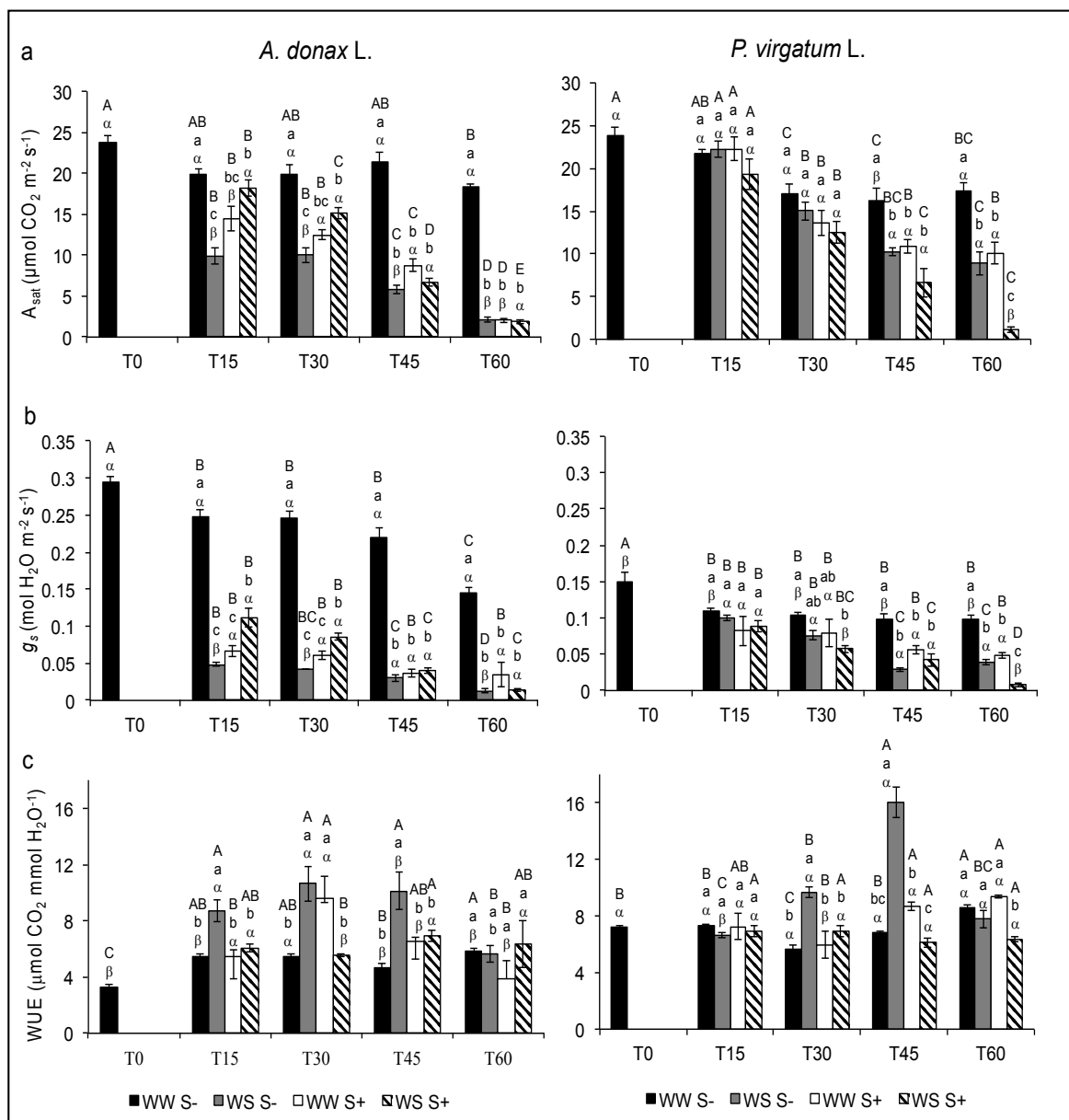
**Table 2 - UIB.** Plant biomass (g), shoot biomass (g) and plant height (mm) of micropropagated plantlets of *A. donax* (AM, inoculated and Non-AM, non-inoculated plants). Values are means of 10 replicates  $\pm$  standard error. Different letters indicate significant differences at  $p < 0.05$ .

|                   | AM Plants          | Non-AM plants      |
|-------------------|--------------------|--------------------|
| Total dry biomass | 0.235 $\pm$ 0.010a | 0.163 $\pm$ 0.011b |
| Shoot dry biomass | 0.164 $\pm$ 0.008a | 0.104 $\pm$ 0.008b |
| Plant height      | 168.9 $\pm$ 6.77a  | 129.4 $\pm$ 5.18b  |

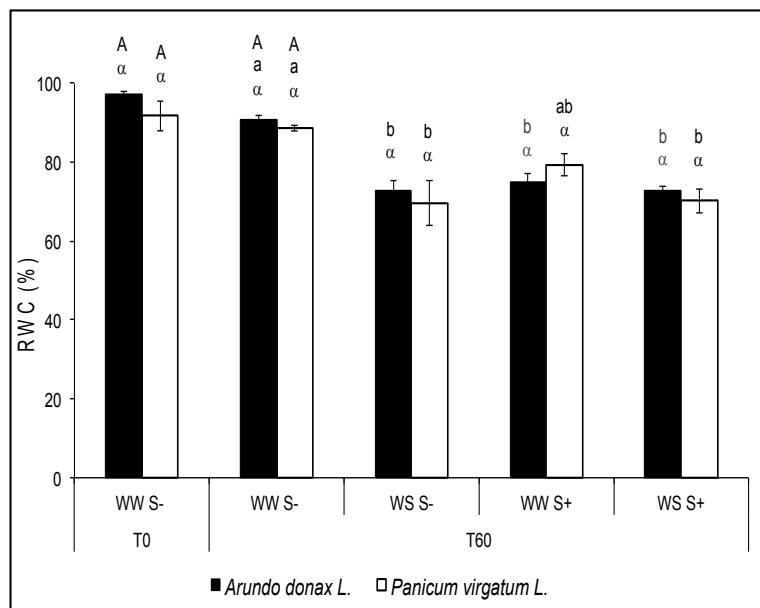




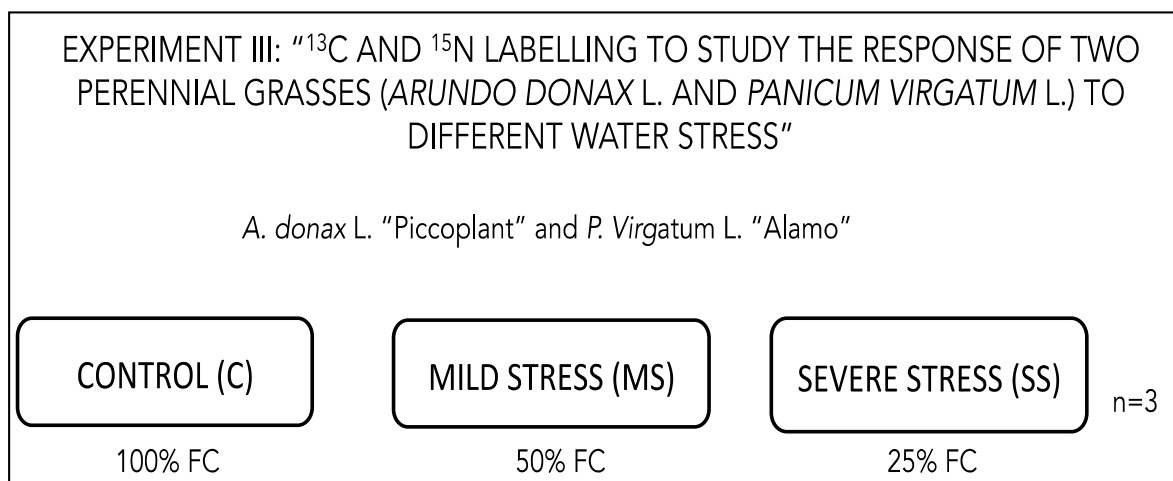
**Figure 1 – UB.** Scheme of the experimental design of experiment II.



**Figure 2 – UB.** Representation of  $A_{sat}$  (a),  $g_s$  (b) and  $iWUE$  (c) values of *A. donax* and *P. virgatum* during (T0, T15, T30, T45, T60) for each treatment: i) well-watered without salinity (WW S-); ii) low watered without salinity (WS S-); iii) well-watered with salinity (WW S+) and iv) low watered with salinity (WS S+). Values are the mean of four replicates and standard errors (SE) are shown. Data were analyzed with an ANOVA Tukey analysis. Different capital letters mean significant differences ( $P < 0.05$ ) between time (T0-T60) for the same species and treatment, different small letters mean significant differences ( $P < 0.05$ ) between treatments for the same species and time and different Greek letters mean significant differences ( $P < 0.05$ ) between species for the same treatment and time.

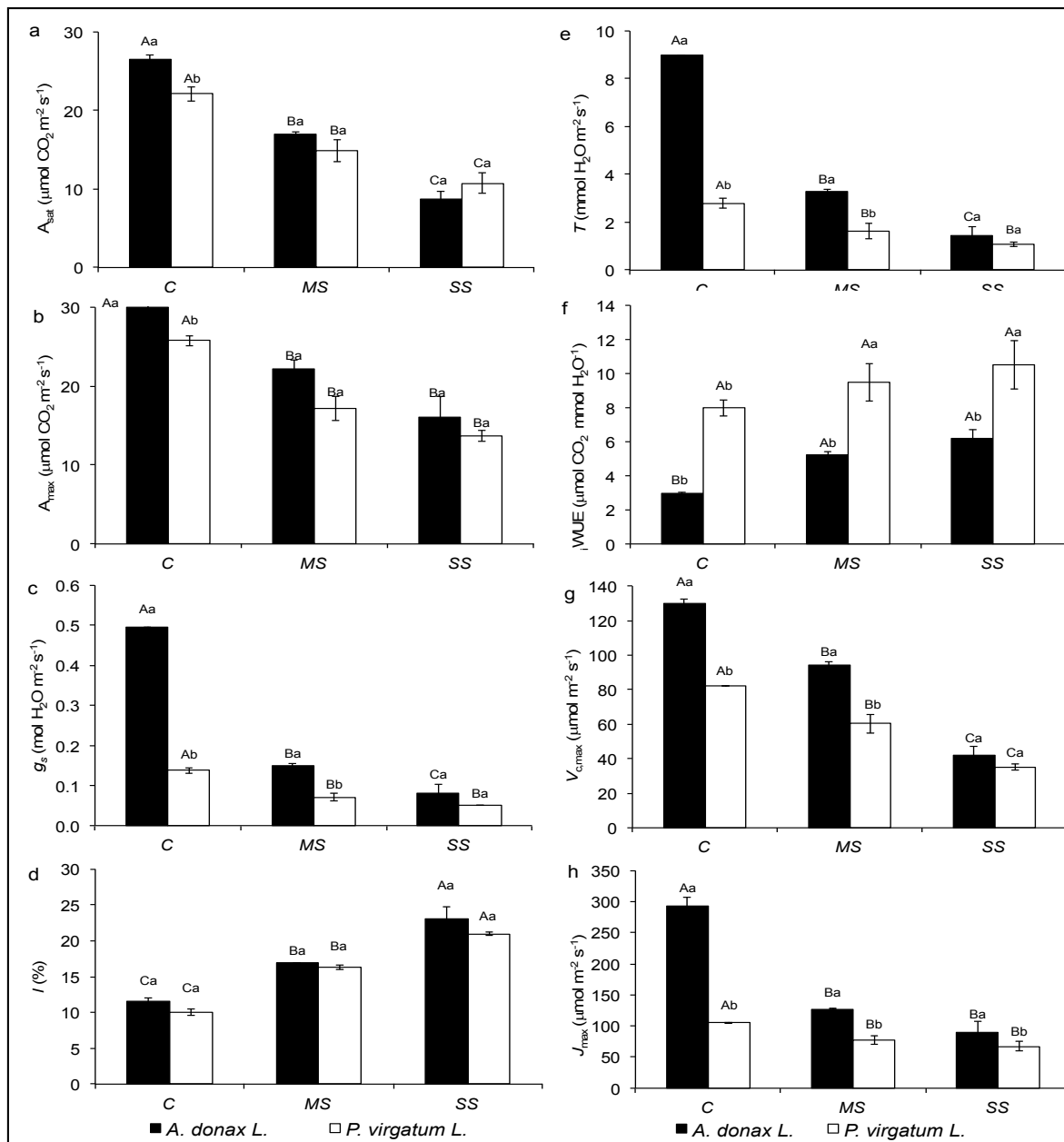


**Figure 3 - UB.** Changes in relative water content (RWC, %) between the beginning (T0) and the end of the experiment (T60) for both species and treatments (WW S-, WS S-, WW S+ and WS S+). Data are the means of three replicates and the standard errors (SE) are shown. Data were analysed with an ANOVA Tukey analysis. Different capital letters mean significant differences ( $P < 0.05$ ) between time (T0 – T60) for the same species in the control treatment (WW S-), different small letters mean significant differences ( $P < 0.05$ ) between treatments for a same species at T60 and different Greek letters mean significant differences ( $P < 0.05$ ) between species for same treatment.

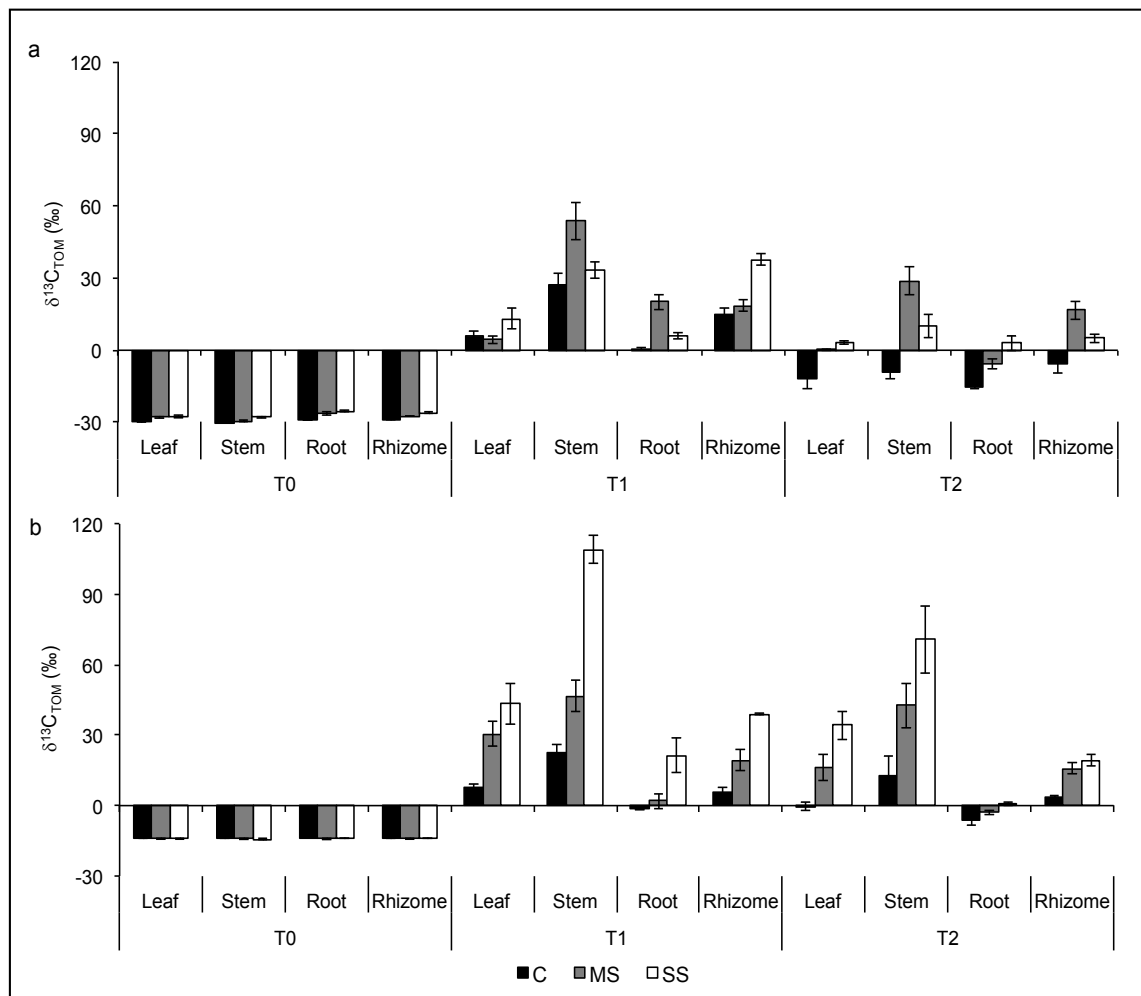


**Figure 4 – UB.** Scheme of the experimental design of experiment III.



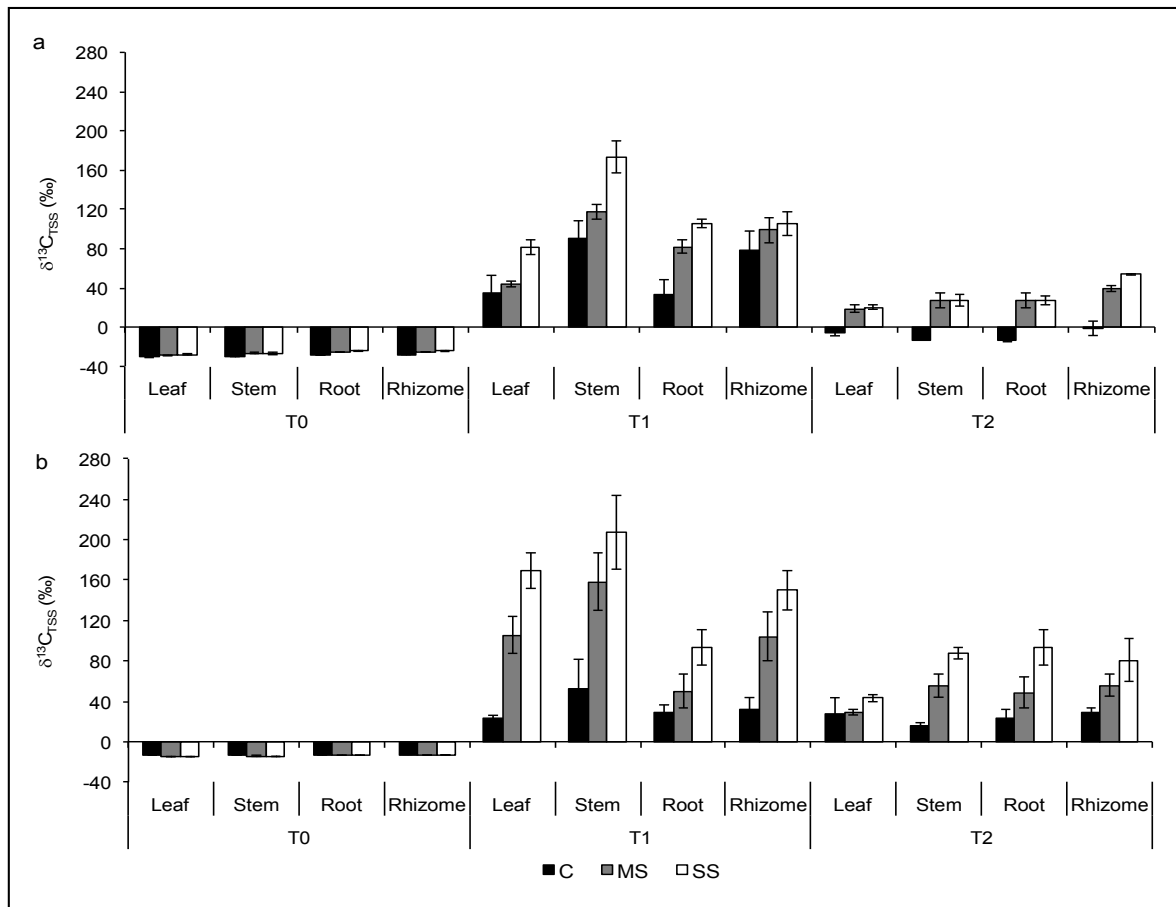


**Figure 5 – UB.** Photosynthesis parameters ( $A_{sat}$ , assimilation rate at light saturation (a);  $A_{max}$ , maximum assimilation rate at light and  $\text{CO}_2$  saturation (b);  $g_s$ , stomatal conductance (c);  $l$ , stomatal limitation (d);  $T$ , transpiration rate (e); WUE, water use efficiency (f);  $V_c$ , max, maximum carboxylation velocity of Rubisco (g);  $J_{max}$ , the rate of photosynthetic electron transport (h)) in both species (*A. donax* and *P. virgatum* L.) for each treatment: i) Control (C, 100% FC), ii) Mild stress (MS, 50% FC) and iii) Severe Stress (SS, 25% FC) at the end of the experiment (Tf). Data are the means of three replicates and the standard error is shown. Data were analyzed with an ANOVA Tukey analysis. Different capital letters indicate significant differences ( $P < 0.05$ ) between treatments for the same species and different small letters indicate significant differences ( $P < 0.05$ ) between species for the same treatment.



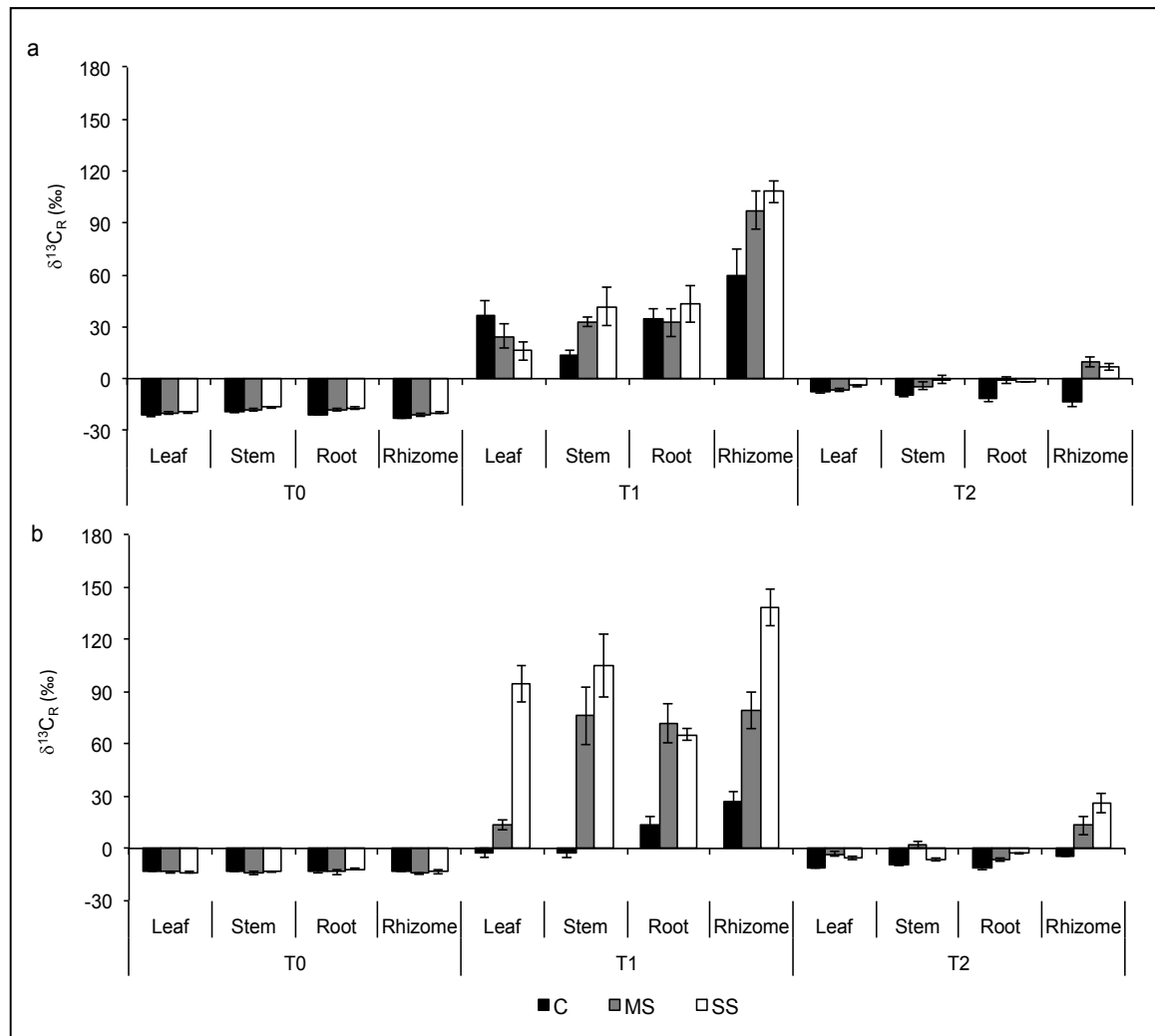
**Figure 6 – UB.** Water stress effects (Control (C, 100% FC), Mild Stress (MS, 50% FC) and Severe Stress (SS, 25% FC) on  $\delta^{13}\text{C}$  values (‰) of  $\text{CO}_2$  in total organic matter ( $\delta^{13}\text{C}_{\text{TOM}}$ ) in leaves, stems, roots and rhizomes in both species (*A. donax* (a) and *P. virgatum* (b)) before labelling (T0), 1 day after labelling (T1) and 7 days after labelling (T2).





**Figure 7 – UB.** Water stress effects (Control (C, 100% FC), Mild stress (MS, 50% FC) and Severe Stress (SS, 25% FC) on  $\delta^{13}C$  values (‰) of sugars ( $\delta^{13}CTSS$ ) in leaves, stems, roots and rhizomes in both species (*A. donax* (a) and *P. virgatum* (b)) before labelling (T0), 1 day after labelling (T1) and 7 days after labelling (T2).





**Figure 8 - UB.** Water stress effects (Control (C, 100% FC), Mild Stress (MS, 50% FC) and Severe Stress (SS, 25% FC) on  $\delta^{13}C$  values (‰) of respired  $CO_2$  ( $\delta^{13}C_R$ ) in leaves, stems, roots and rhizomes in both species (*A. donax* (a) and *P. virgatum* (b)) before labelling (T0), 1 day after labelling (T1) and 7 days after labelling (T2).



## WP2 – Plant biotechnology

### Instituto of Agrobiotechnology Rosario – INDEAR (n. 17)

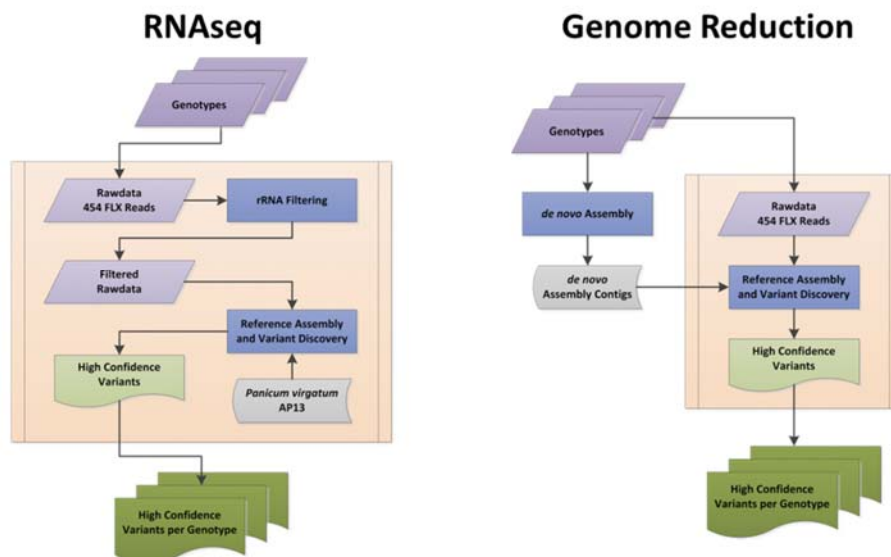
**Table 1 – INDEAR.** Sequencing metrics; level of the sequencing process (read 1, read 2/index, read 3), cycles of chemistry, total yield of each step and percentage of bases with quality scores equal or above Q30.

| Level        | Cycles     | Total Yield   | % ≥ Q30     |
|--------------|------------|---------------|-------------|
| Read 1       | 151        | 47.8 G        | 81.8        |
| Read 2 (I)   | 7          | 1.9 G         | 90.0        |
| Read 3       | 151        | 47.8 G        | 79.4        |
| <b>Total</b> | <b>309</b> | <b>97.5 G</b> | <b>80.8</b> |

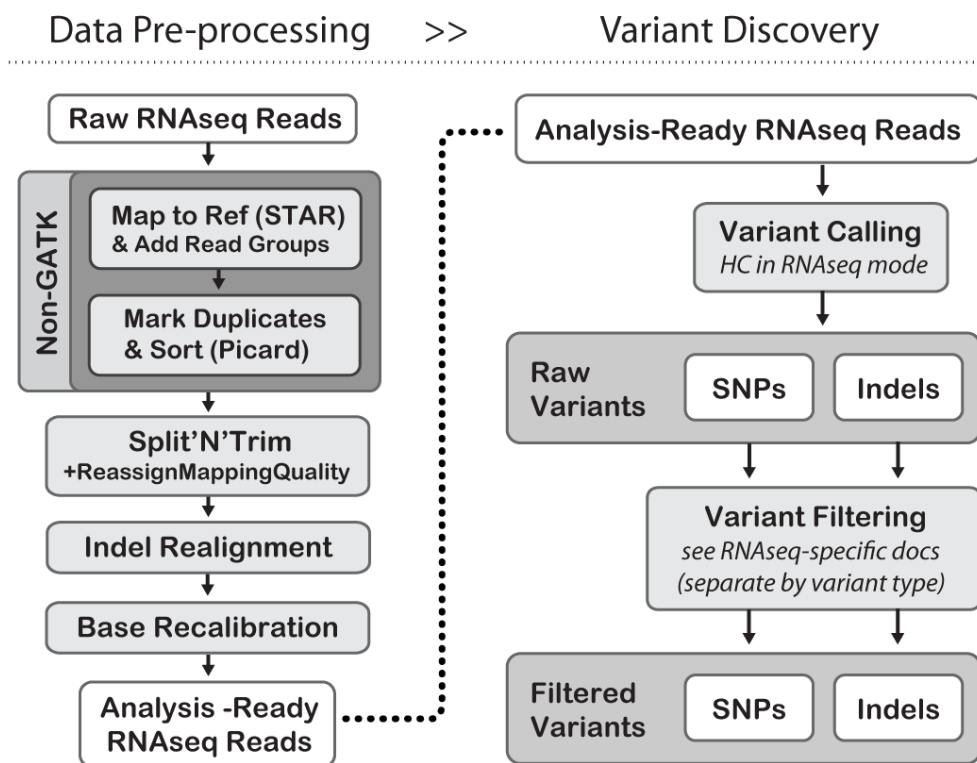
**Table 2 – INDEAR.** Metrics per sample, sample ID/genotype, total passing filter (PF) paired-ends (PE) reads in millions (M), total bases sequenced (Gb).

| Sample ID    | Total PF<br>PE reads<br>(M) | Total bases<br>sequenced<br>(Gb) |
|--------------|-----------------------------|----------------------------------|
| Summer       | 98                          | 14,9                             |
| Caddo        | 90                          | 13,7                             |
| Greenville   | 92                          | 13,9                             |
| Pathfinder   | 79                          | 12,0                             |
| EG1101       | 73                          | 11,1                             |
| Blackwell    | 119                         | 18,0                             |
| EG2101       | 80                          | 12,0                             |
| <b>Total</b> | <b>632</b>                  | <b>95,50</b>                     |
| Average      | 90                          | 13,6                             |
| SD           | 15                          | 2,3                              |





**Figure 1A - INDEAR.** RNA-seq pipeline for 454/Roche sequencing. **B.** Genome Reduction pipeline for 454/Roche sequencing.



**Figure 2 - INDEAR.** RNA-seq variant Discovery pipeline by GATK.

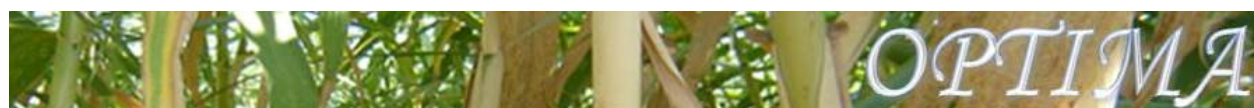


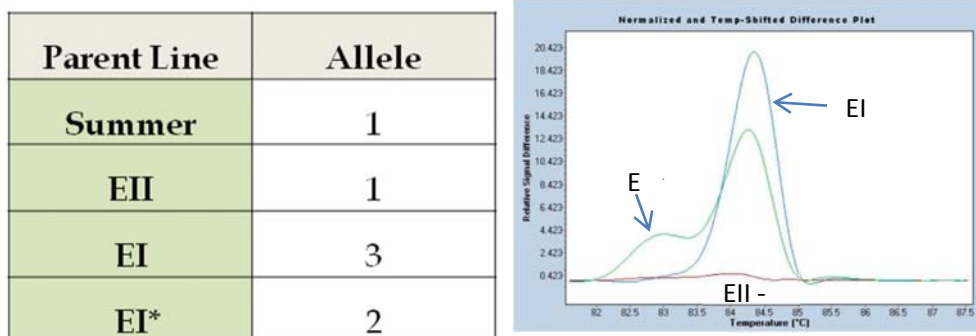
**Table 3 - INDEAR.** Some detail about the crosses that have been carried out in INDEAR between the pre-selected switchgrass cultivars. Experimental I (Exp1 or EI) X Experimental II (Exp2 or EII) and EI X S (Summer). Exp 1 and Exp 1\* correspond to two different parental plants. The crosses or lines in grey correspond to failed crosses as detection of female parent line has occurred.

| Crop        | Female parent | X | Male Parent | Envelope # | Sowing Date |
|-------------|---------------|---|-------------|------------|-------------|
| Switchgrass | Exp 1*        | x | Summer      | 23         | 28/04/15    |
| Switchgrass | Summer        | x | Exp 1       | 10         | 28/04/15    |
| Switchgrass | Exp 1 *       | x | Summer      | 3          | 28/05/15    |
| Switchgrass | Exp 1 *       | x | Summer      | 3          | 28/05/15    |
| Switchgrass | Summer        | x | Exp 1*      | 4          | 28/05/15    |
| Switchgrass | Summer        | x | Exp 1*      | 4          | 28/05/15    |
| Switchgrass | Summer        | x | Exp 1*      | 4          | 28/05/15    |
| Switchgrass | Exp 1*        | x | Summer      | 8          | 28/05/15    |
| Switchgrass | Exp 2         | x | Exp 1       | 3          | 28/05/15    |
| Switchgrass | Exp 1         | x | Exp 2       | 5          | 28/05/15    |
| Switchgrass | Exp 1         | x | Exp 2       | 5          | 28/05/15    |
| Switchgrass | Exp 1         | x | Exp 2       | 5          | 28/05/15    |



**Figure 4 - INDEAR.** Representative photograph of ongoing F<sub>1</sub> crosses at Indear greenhouse.





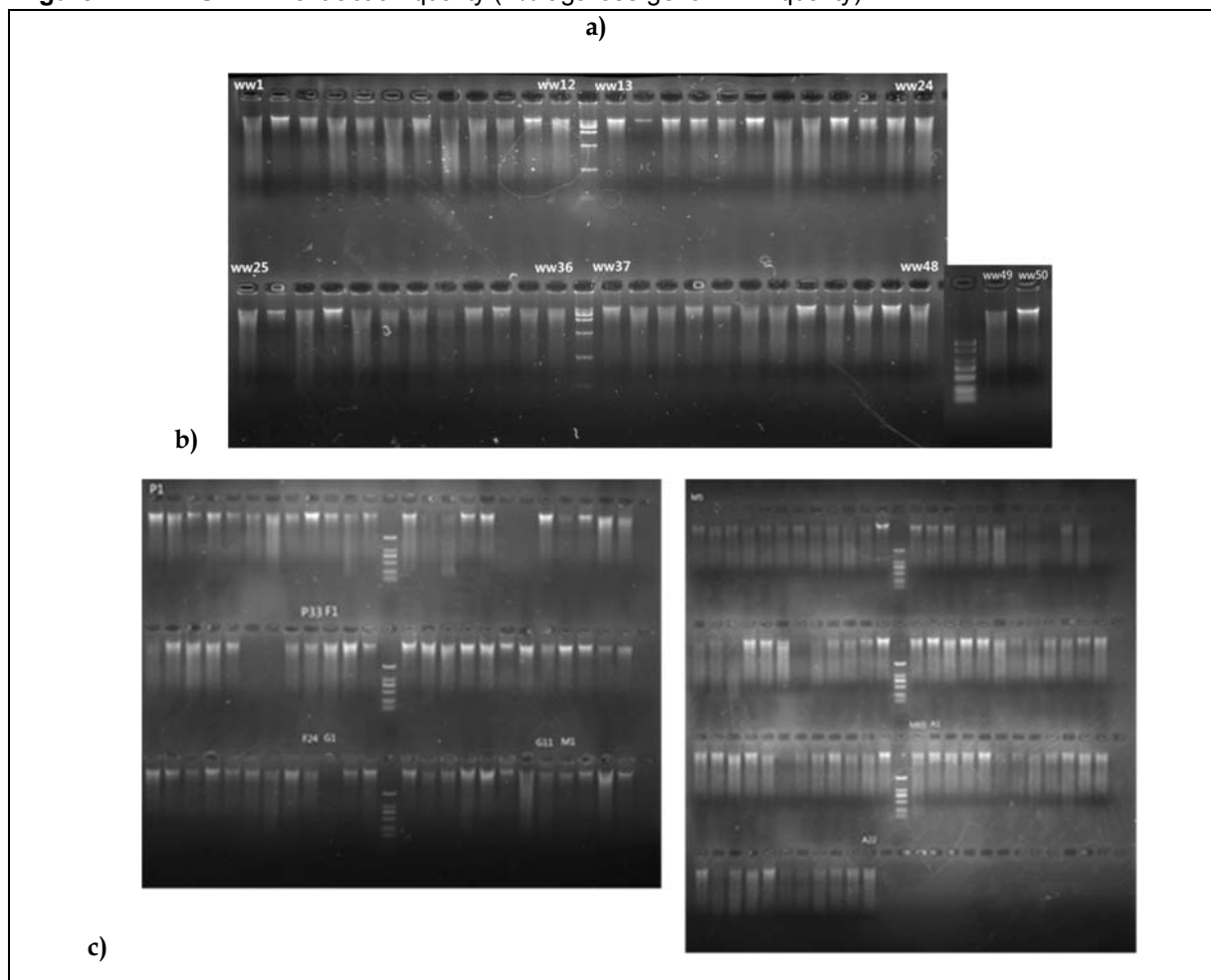
**Figure 5 - INDEAR.** Results obtained by HRM for marker 1 for the different parent plants. Right panel, Graphic representation of the different normalized temperatures (curves) for Marker 1. Curves that correspond to the different parent lines are indicated. Each color curve corresponds to one allele of the SNP of contig 67032, position 4.044.

## Huazhong Agricultural University – HZAU (n. 14)

**Table 1 – HZAU.** The number of sample and SNP for GBS

| Project    |                   | Samples | SNP numbers |
|------------|-------------------|---------|-------------|
| Map genome | Drought tolerance | 50      | 319,900     |
|            | Piccoplant        | 27      | 213,057     |
|            | Fondachello       | 24      | 210,491     |
|            | Granadensis       | 9       | 31,655      |
|            | Martinensis       | 52      | 303,593     |
| Total      |                   | 181     | 1,184,113   |

**Figure 1 – HZAU.** DNA extraction quality (1% agarose gel of DNA quality)

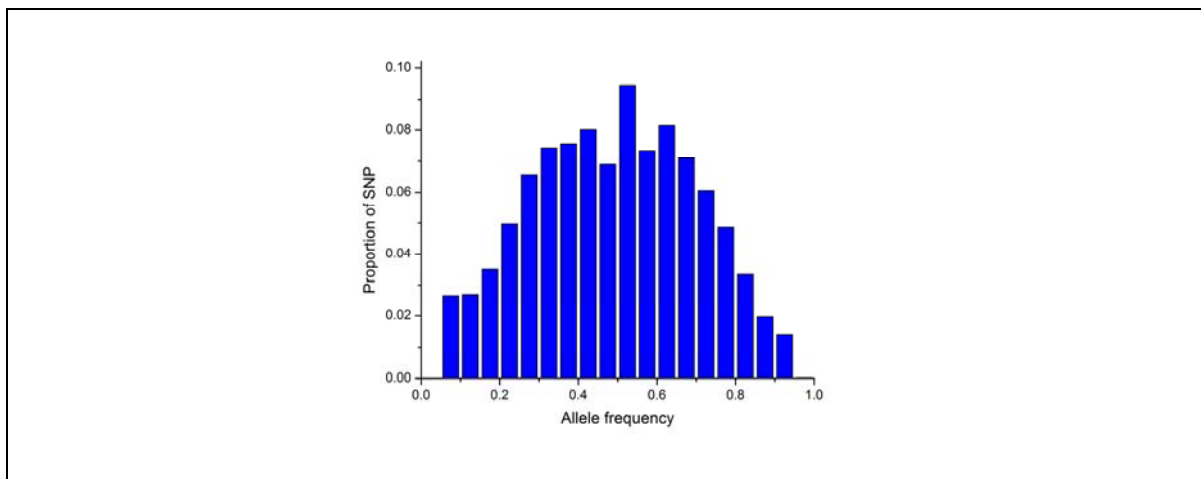


**Table 1 - HZAU.** GBS raw data statistic

| Total read numbers | Accepted reads numbers | Accepted reads ratio | The average depth of each clones | Raw SNP numbers |
|--------------------|------------------------|----------------------|----------------------------------|-----------------|
| 197,671,204        | 144,058,366            | 73%                  | 0.288 Gb                         | 319,900         |

**Table 2 – HZAU.** The number of sample and SNP for GBS

| Project           | Samples     | SNP numbers |         |
|-------------------|-------------|-------------|---------|
| Drought tolerance | 50          | 319,900     |         |
| Piccoplant        | 27          | 213,057     |         |
| Fondachello       | 24          | 210,491     |         |
| Map genome        | Granadensis | 9           | 31,655  |
|                   | Martinensis | 52          | 303,593 |
|                   | Argentum    | 19          | 105,417 |
| Total             | 181         | 1,184,113   |         |



**Figure 2 - HZAU.** Total SNPs allele frequency distribution of drought tolerance experiment



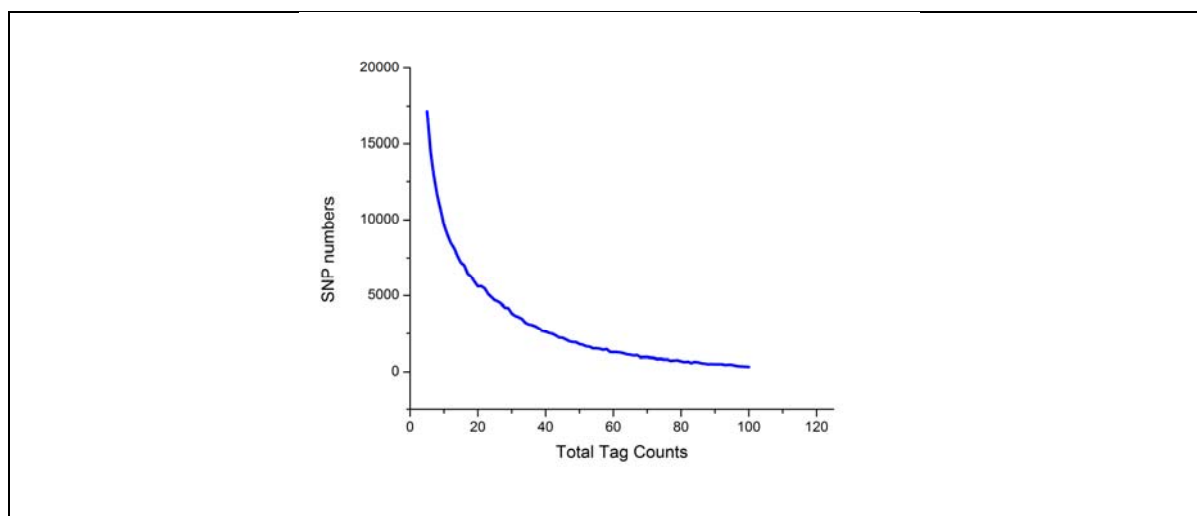


Figure 2 - **HZAU**. Tag counts depths distribution in 50 clones of drought tolerance experiment.

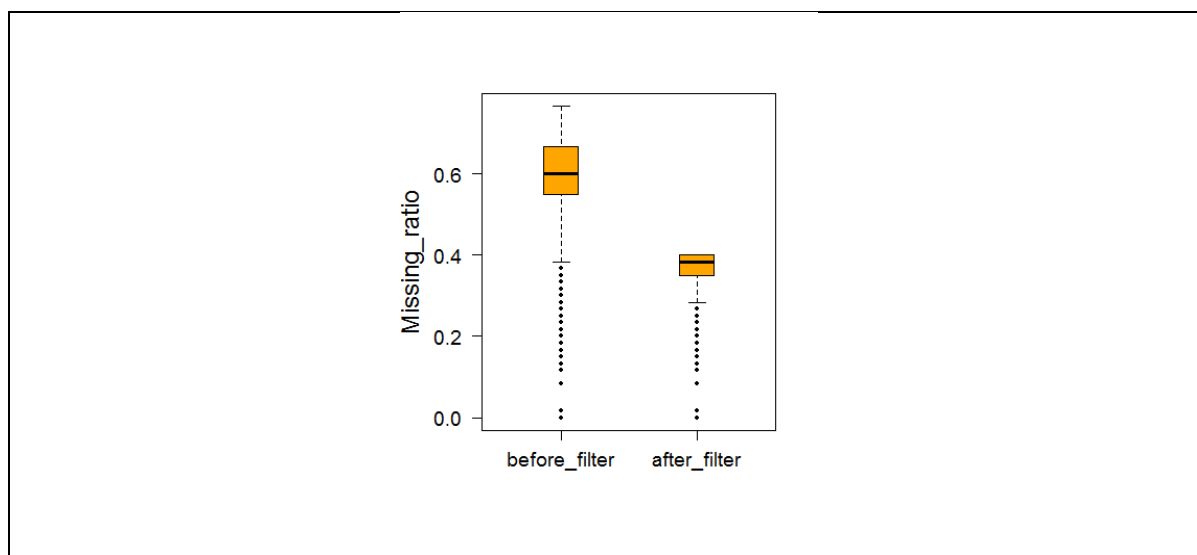
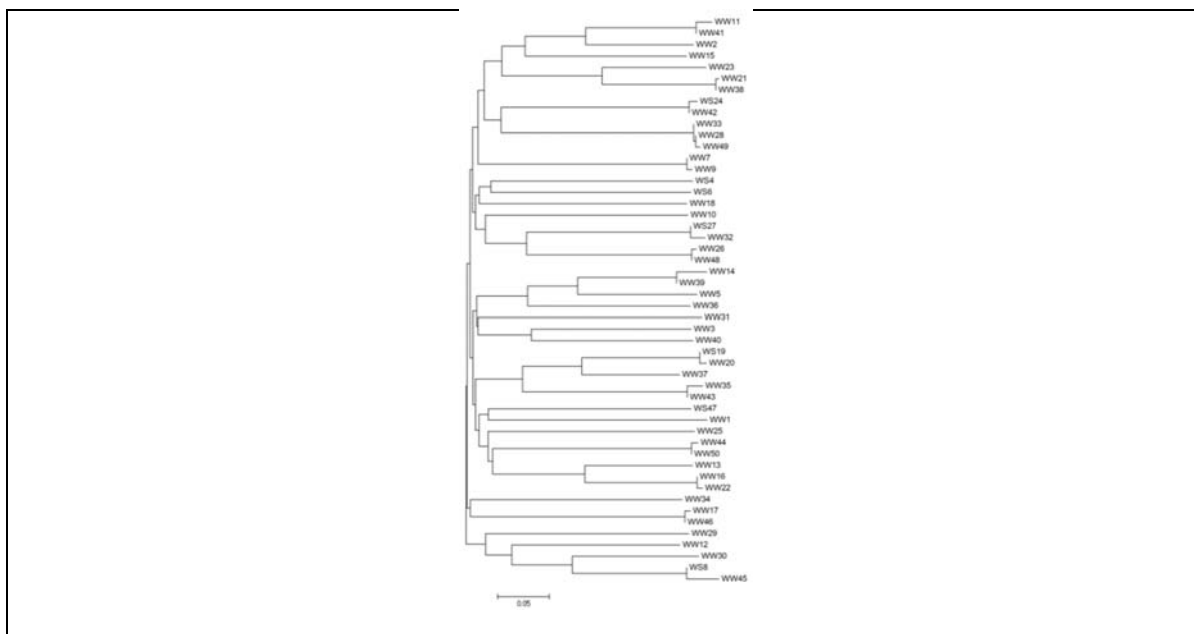
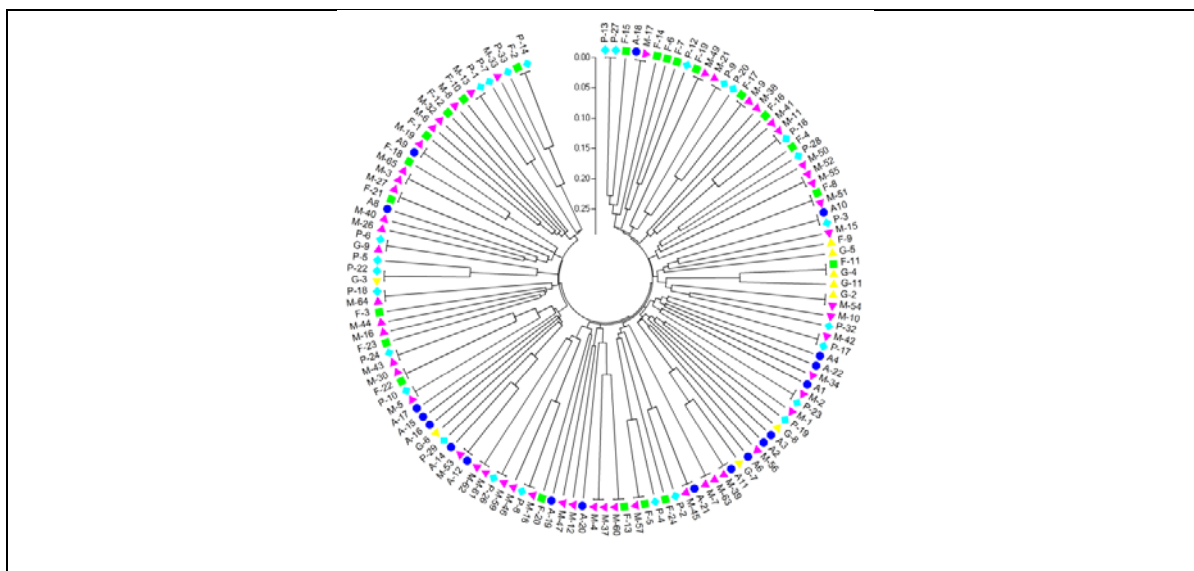


Figure 3 - **HZAU**. Genotype missing ratio before and after filtering.



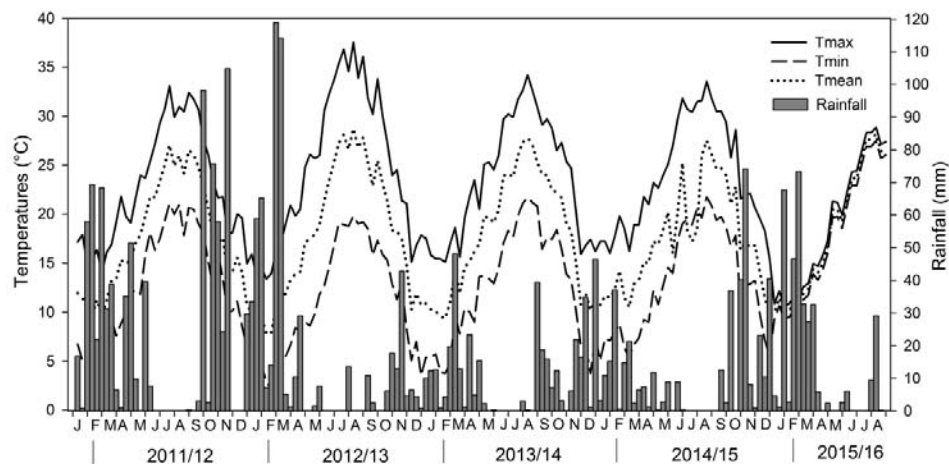
**Figure 4 - HZAU.** Neighbor-Joining tree of 50 samples was constructed by the 319,900 SNPs markers.



**Figure 5 - HZAU.** Neighbor-Joining tree of 131 samples contained 5 ecotypes was constructed.

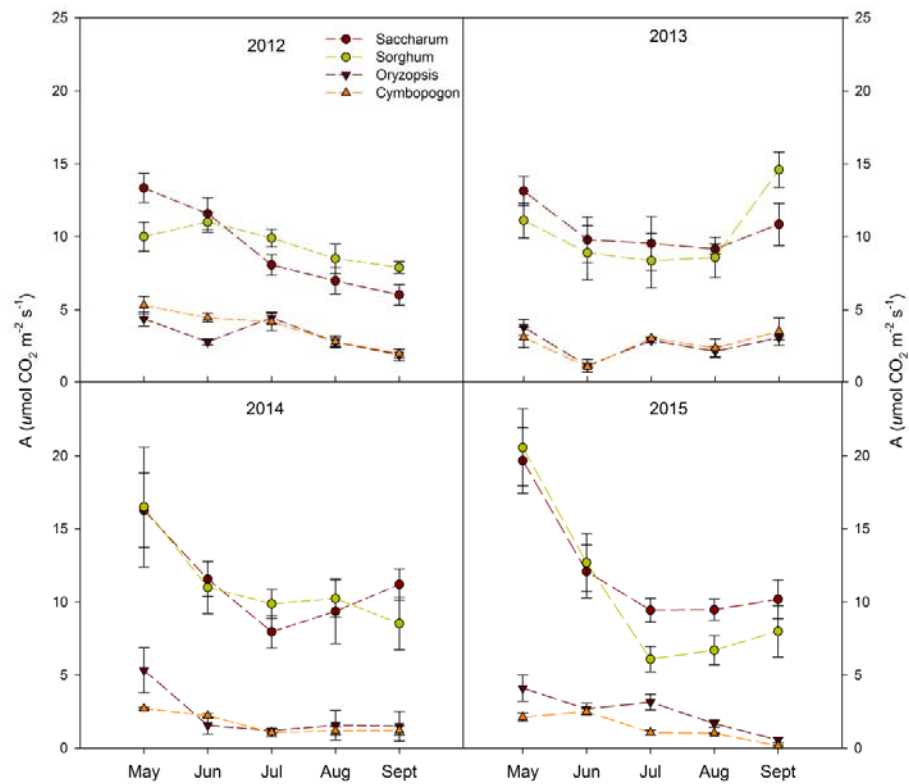
### Task 3.1 - Characterization of endemic grasses and novel plant varieties adapted to southern European conditions

Università di Catania - UNICT (n. 1)



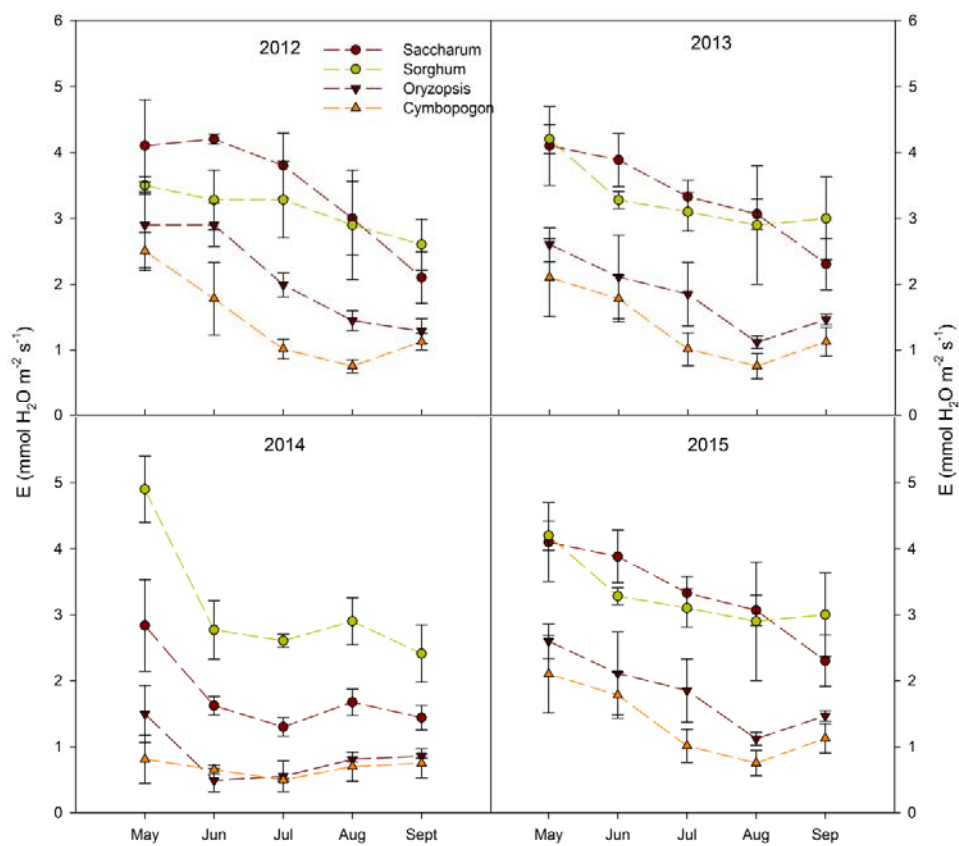
**Figure 1 - UNICT.** Meteorological trend from establishment up to last harvest of perennial spontaneous grasses at the UNICT Experimental farm.





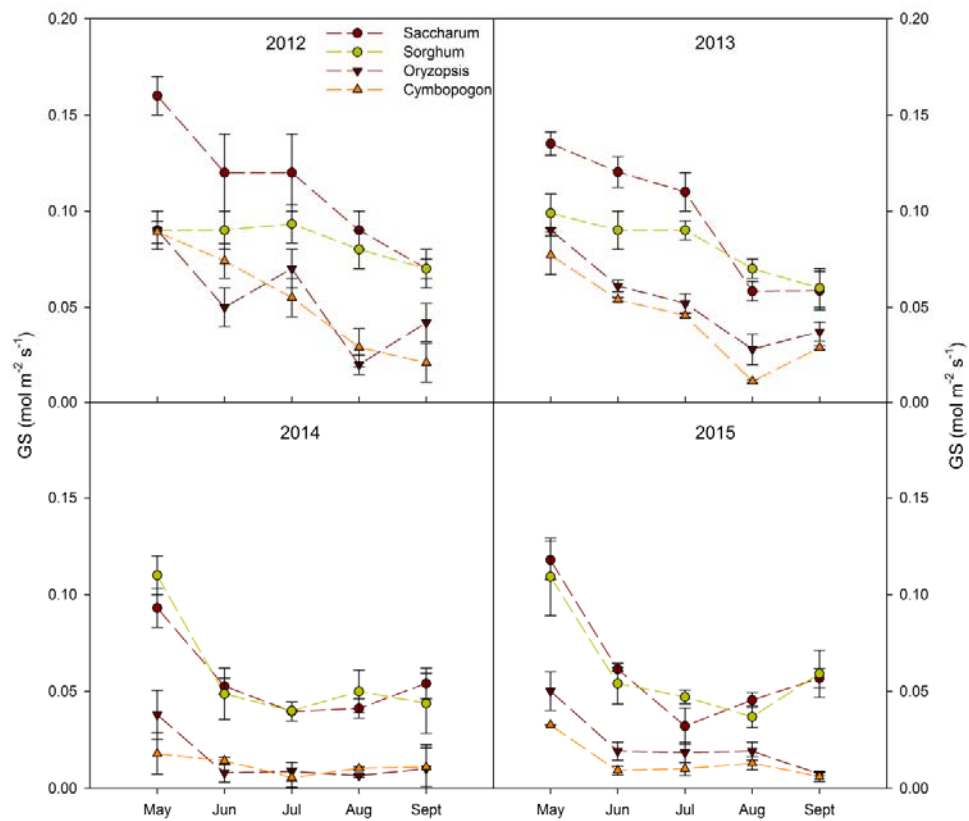
**Figure 2 - UNICT.** Assimilation rate (A,  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) of the four endemic grasses in 2012, 2013, 2014 and 2015 growing seasons.





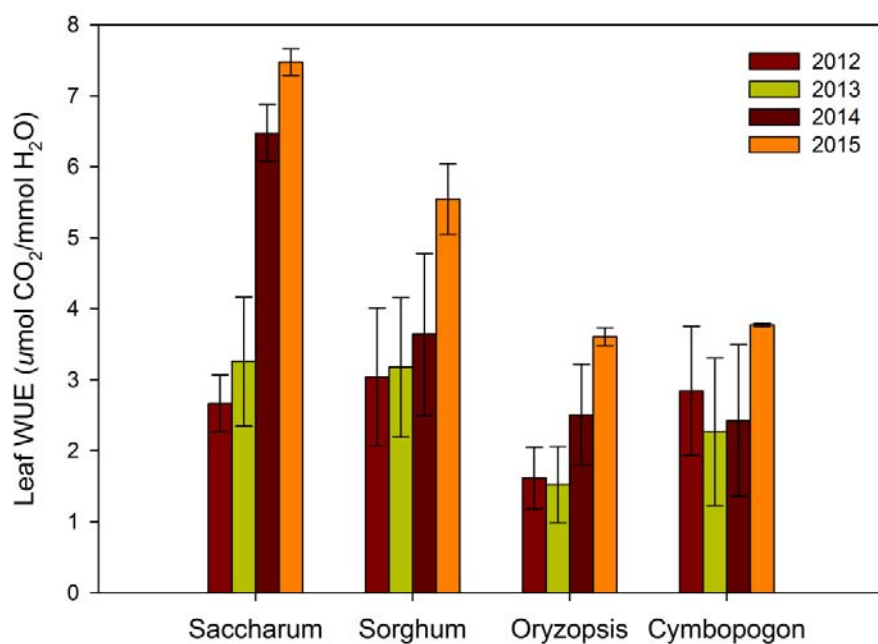
**Figure 3 - UNICT.** Transpiration rate ( $E$ ,  $\text{mmol H}_2\text{O m}^{-2} \text{s}^{-1}$ ) of the four endemic grasses in 2012, 2013, 2014 and 2015 growing seasons.



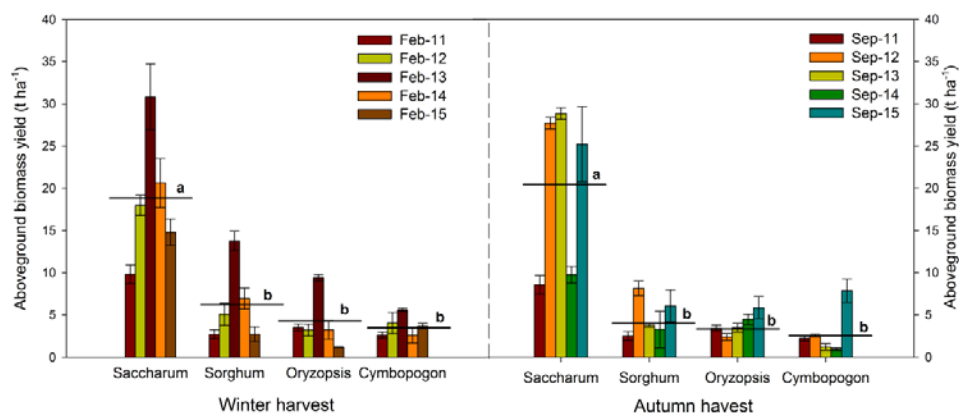


**Figure 4 - UNICT.** Stomatal conductance (GS,  $\text{mol m}^{-2} \text{s}^{-1}$ ) of the four endemic grasses in 2012, 2013, 2014 and 2015 growing seasons.



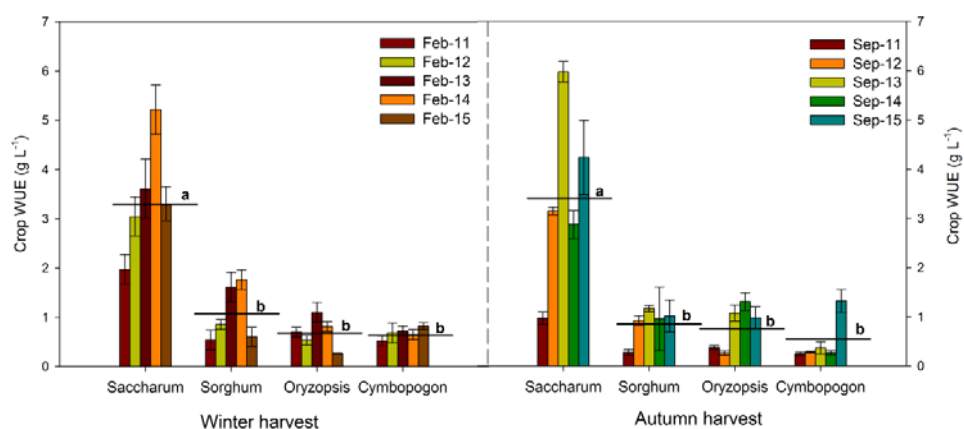


**Figure 5 - UNICT.** Leaf Water Use Efficiency ( $\mu\text{mol CO}_2/\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) of the four endemic grasses in 2012, 2013, 2014 and 2015 growing seasons.

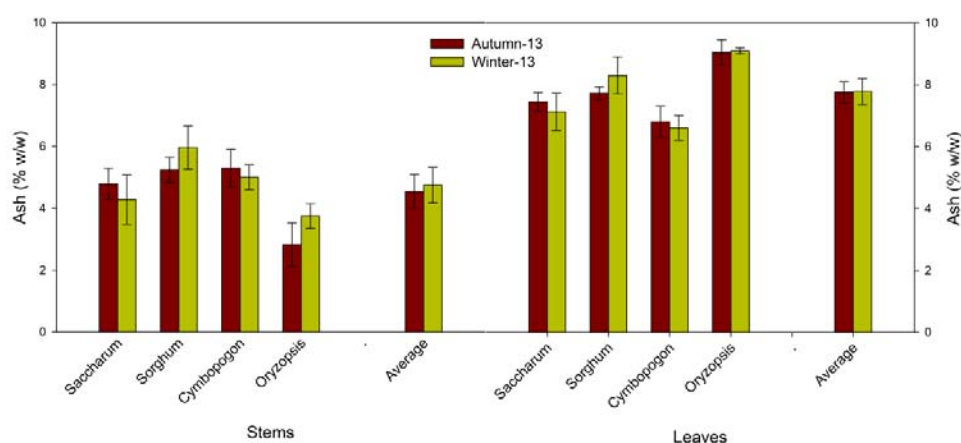


**Figure 6 - UNICT.** Aboveground dry biomass yield of wild grasses (*O. miliacea*, *C. hirtus*, *S. halepense* and *S. spontaneum* L. ssp. *aegyptiacum*) in different years and harvest dates. Horizontal lines represent the mean within the species. Different letters indicate significant mean according to the LSD test at  $p \leq 0.05$ .





**Figure 7 - UNICT.** Water Use Efficiency (g L<sup>-1</sup>) of wild grasses (*O. miliacea*, *C. hirtus*, *S. halepense* and *S. spontaneum* L. ssp. *aegyptiacum*) in different years and harvest dates. Horizontal lines represent the mean within the species. Different letters indicate significant mean according to the LSD test at  $p \leq 0.05$ .

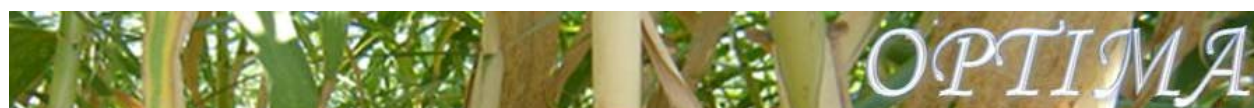


**Figure 8 - UNICT.** Ash content of wild grasses (*O. miliacea*, *C. hirtus*, *S. halepense* and *S. spontaneum*) in different harvest dates and biomass part (stems and leaves).



**Table 1 - UNICT.** Raw material composition of *O. miliacea*, *C. hirtus*, *S. halepense* and *S. spontaneum* in different harvest dates (autumn 2013 and winter 2014) and biomass part (stems and leaves).

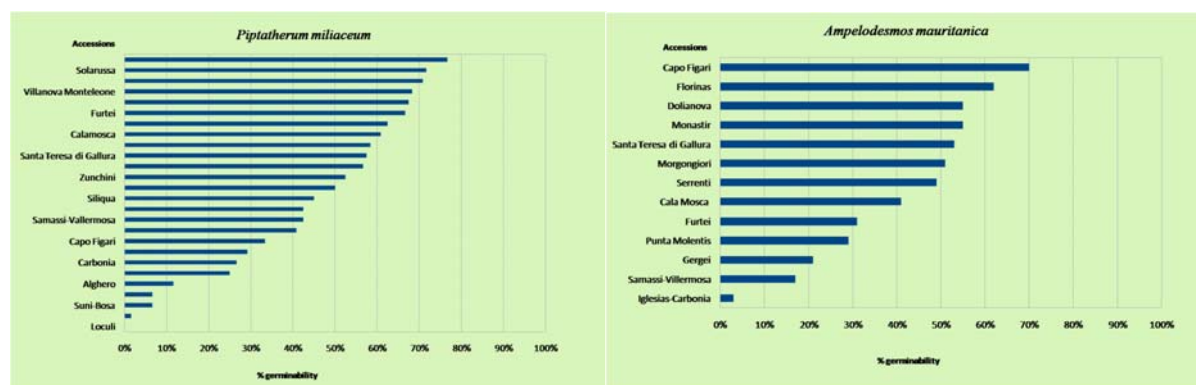
| Species           | Year | Harvest | Part | NDS               | Hemicellulose     | Cellulose         | Lignin            |
|-------------------|------|---------|------|-------------------|-------------------|-------------------|-------------------|
| <i>Saccharum</i>  | 2014 | Winter  | Stem | 20.82 ±0.51       | 21.16±3.95        | 35.62±4.30        | 22.23±3.53        |
| <i>Sorghum</i>    | 2014 | Winter  | Stem | 13.04±2.21        | 27.68±4.33        | 39.13±2.19        | 16.87±0.84        |
| <i>Cymbopogon</i> | 2014 | Winter  | Stem | 21.08±2.94        | 11.27±0.66        | 43.27±0.89        | 23.48±2.09        |
| <i>Oryzopsis</i>  | 2014 | Winter  | Stem | 21.70±1.08        | 20.83±4.42        | 34.06±5.52        | 17.94±0.45        |
| <b>Average</b>    |      |         |      | <b>19.16±3.54</b> | <b>20.23±5.85</b> | <b>38.02±3.54</b> | <b>20.13±2.78</b> |
| <i>Saccharum</i>  | 2014 | Winter  | Leaf | 19.65±0.58        | 27.99±1.68        | 30.79±1.30        | 15.31±1.97        |
| <i>Sorghum</i>    | 2014 | Winter  | Leaf | 19.43±3.23        | 22.06±2.63        | 35.29±2.40        | 16.26±1.18        |
| <i>Cymbopogon</i> | 2014 | Winter  | Leaf | 18.27±4.66        | 29.70±4.39        | 28.90±1.58        | 16.39±1.13        |
| <i>Oryzopsis</i>  | 2014 | Winter  | Leaf | 23.01±2.15        | 23.81±4.42        | 26.73±4.56        | 19.69±1.33        |
| <b>Average</b>    |      |         |      | <b>20.07±1.76</b> | <b>25.89±3.07</b> | <b>30.42±3.15</b> | <b>16.91±1.65</b> |
| <i>Saccharum</i>  | 2013 | Autumn  | Stem | 13.00±2.24        | 22.15±2.95        | 36.72±1.18        | 17.46±2.59        |
| <i>Sorghum</i>    | 2013 | Autumn  | Stem | 25.64±1.57        | 15.80±2.67        | 31.69±1.10        | 14.78±2.73        |
| <i>Cymbopogon</i> | 2013 | Autumn  | Stem | 13.88±0.98        | 25.47±1.97        | 35.64±3.11        | 19.10±1.14        |
| <i>Oryzopsis</i>  | 2013 | Autumn  | Stem | 19.92±2.22        | 27.30±0.99        | 23.82±3.31        | 29.69±2.98        |
| <b>Average</b>    |      |         |      | <b>18.11±5.09</b> | <b>22.68±4.38</b> | <b>31.96±5.06</b> | <b>20.25±5.65</b> |
| <i>Saccharum</i>  | 2013 | Autumn  | Leaf | 13.89±2.42        | 28.13±0.39        | 36.65±2.26        | 19.07±2.35        |
| <i>Sorghum</i>    | 2013 | Autumn  | Leaf | 22.85±2.60        | 16.36±2.09        | 32.22±2.60        | 15.38±2.30        |
| <i>Cymbopogon</i> | 2013 | Autumn  | Leaf | 22.86±2.49        | 27.29±0.92        | 27.19±3.53        | 17.66±2.60        |
| <i>Oryzopsis</i>  | 2013 | Autumn  | Leaf | 17.84±1.84        | 27.10±1.14        | 26.89±2.51        | 21.74±2.29        |
| <b>Average</b>    |      |         |      | <b>19.36±3.76</b> | <b>24.72±4.84</b> | <b>30.73±4.01</b> | <b>18.46±2.30</b> |



## Istituto per il Sistema Produzione Animale in Ambiente Mediterraneo – CNR-IPAAM (n. 8)

**Table 1 – CNR-IPAAM.** Total number of accessions (TotA) and accessions of Mediterranean (MedA) origin found in the main world repositories freely searchable on the Internet. The search regarded four high-yielding forage species and four native species collected in the project OPTIMA for their potential interest as bioenergy plants for marginal environments.

| Species                | Repository    |               |               |               |               |               |
|------------------------|---------------|---------------|---------------|---------------|---------------|---------------|
|                        | GRIN          |               | EURISCO       |               | Margot Ford   |               |
|                        | TotA<br>(no.) | MedA<br>(no.) | TotA<br>(no.) | MedA<br>(no.) | TotA<br>(no.) | MedA<br>(no.) |
| <i>D. glomerata</i>    | 1491          | 755           | 13569         | 759           | 1135          | 421           |
| <i>F. arundinacea</i>  | 940           | 572           | 2230          | 235           | 941           | 289           |
| <i>L. perenne</i>      | 645           | 212           | 10581         | 380           | 3681          | 880           |
| <i>P. aquatica</i>     | 371           | 265           | 57            | 26            | 431           | 147           |
| <i>S. spontaneum</i>   | 620*          | 1**           | 2             | 0             | 0             | 0             |
| <i>A. mauritanicus</i> | 0             | 0             | 1             | 1             | 0             | 0             |
| <i>P. miliaceum</i>    | 111           | 105           | 6             | 6             | 1             | 0             |
| <i>H. hirta</i>        | 34            | 21            | 3             | 3             | 0             | 0             |



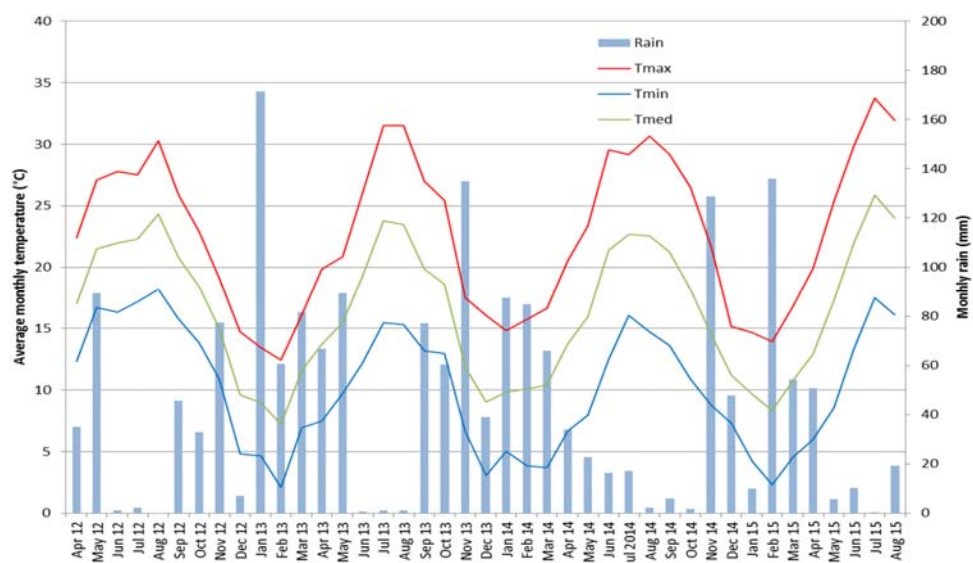
**Table 2 – CNR-ISPAAAM.** Survival rates of native populations of *P. miliaceum*, *A. mauritanicus* and *H. hirta*. The survival rate was recorded after two months from transplant of plantlets.

| Species                | Number of accessions | Survival rate after transplant (%) |          | Survival rate after summer (%) |          |
|------------------------|----------------------|------------------------------------|----------|--------------------------------|----------|
|                        |                      | mean                               | range    | mean                           | range    |
| <i>A. mauritanicus</i> | 12                   | 29.8                               | 0-58.3   | 23.5                           | 0-54.2   |
| <i>P. miliaceum</i>    | 12                   | 84.0                               | 33.3-100 | 81.3                           | 16.7-100 |
| <i>L. perenne</i>      | 1                    | 95.8                               | -        | 50.0                           | -        |
| <i>P. aquatica</i>     | 1                    | 50.0                               | -        | 16.7                           | -        |
| <i>D. glomerata</i>    | 2                    | 39.6                               | 29.2-50  | 39.6                           | 29.2-50  |
| <i>F. arundinacea</i>  | 4                    | 97.9                               | 91.7-100 | 95.8                           | 87.5-100 |
| <i>H. hirta</i>        | 14                   | 30.0                               | 0.0-100  |                                |          |

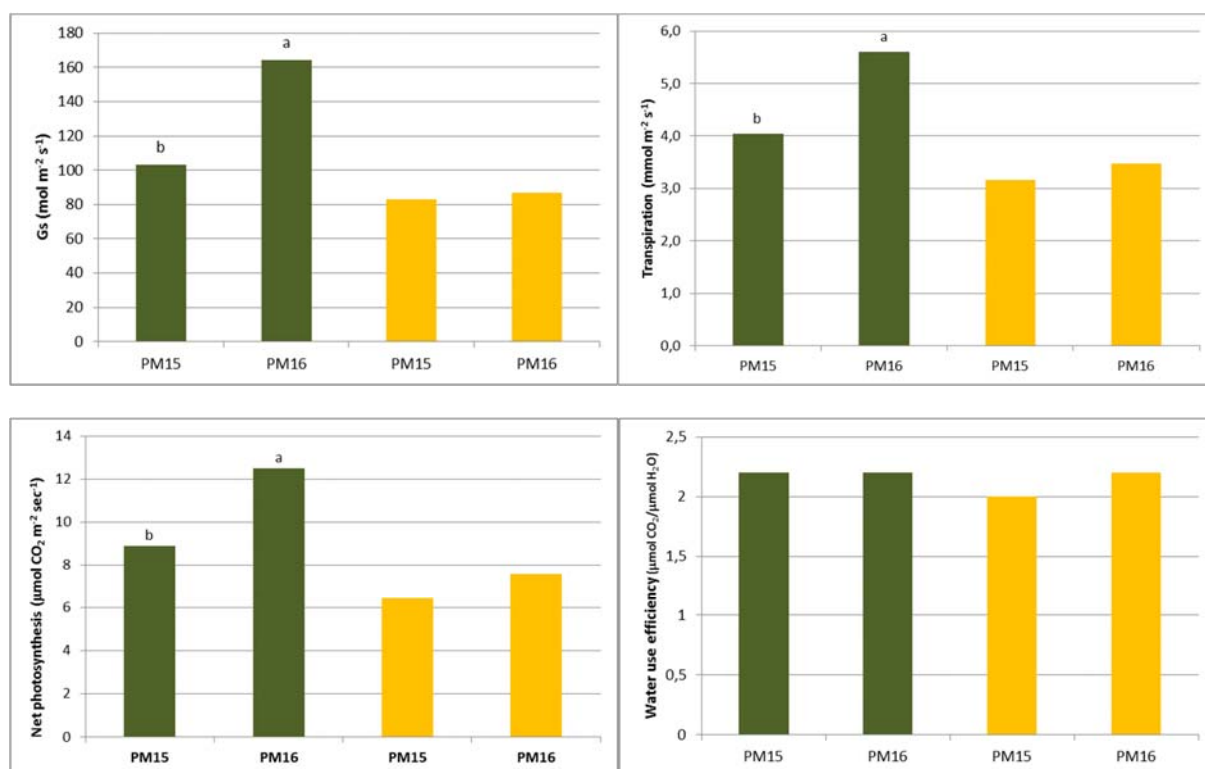
**Table 3 – CNR-ISPAAAM.** Average air temperature and intercepted PAR

| Date       | PAR  | Max air temperature (°C) |
|------------|------|--------------------------|
| 06.06.2014 | 1666 | 28.4                     |
| 07.07.2014 | 1560 | 30.6                     |
| 23.04.2015 | 1874 | 23.6                     |
| 13.05.2015 | 1954 | 29.7                     |
| 29.05.2015 | 2261 | 26.2                     |





**Figure 2 – CNR-ISPAAAM.** The weather pattern of the period from January 2012 to August 2015.



**Figure 3 – CNR-ISPAAAM.** Stomatal conductance, transpiration, net photosynthesis and leaf water use efficiency in two accessions of *P. miliaceum*, measured on June 6<sup>th</sup> 2014 (green bars) and on July 7<sup>th</sup> 2014 (yellow bars).



**Table 4 - CNR-ISPAAM.** Stomatal conductance (Gs), instantaneous transpiration (E), instantaneous photosynthesis (PN) and photosynthetic water use efficiency (WUEi) of *P. miliaceum*, *A. mauritanicus* and *F. arundinacea* under rainfed conditions in Mediterranean environments. Different letters in the column indicate significant statistical differences among values (LSD test,  $p < 0.05$ ).

| Species/<br>accession | Gs<br>(mol m <sup>-2</sup> s <sup>-1</sup> ) |       |        | E<br>(mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> ) |       |        | PN<br>(μmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> ) |       |        | WUEi<br>(μmol CO <sub>2</sub> /mmol<br>H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> ) |       |       |
|-----------------------|----------------------------------------------|-------|--------|---------------------------------------------------------------|-------|--------|---------------------------------------------------------------|-------|--------|-------------------------------------------------------------------------------------------|-------|-------|
|                       | 23.04                                        | 13.05 | 29.05  | 23.04                                                         | 13.05 | 29.05  | 23.04                                                         | 13.05 | 29.05  | 23.04                                                                                     | 13.05 | 29.05 |
| AM4                   | 1.69a                                        | 1.63a | 1.63a  | 0.90a                                                         | 0.74a | 0.73a  | 2.59a                                                         | 2.05a | 2.08a  | 2.91b                                                                                     | 2.88a | 2.86a |
| PM15                  | 1.66a                                        | 1.57b | 1.56b  | 0.75a                                                         | 0.53b | 0.50b  | 2.85a                                                         | 1.39b | 1.49b  | 3.90ab                                                                                    | 2.53a | 3.01a |
| PM16                  | 1.68a                                        | 1.63a | 1.60ab | 0.92a                                                         | 0.76a | 0.59ab | 2.95a                                                         | 2.03a | 1.63ab | 3.35ab                                                                                    | 2.76a | 2.81a |
| FE28                  | 1.66a                                        | 1.54b | 1.57b  | 0.82a                                                         | 0.46b | 0.51b  | 3.10a                                                         | 1.09b | 1.15b  | 4.09a                                                                                     | 2.35a | 2.21b |

**Table 5 - CNR-ISPAAM.** Differences induced by mild (T50) water stress on ecophysiological performances of two accessions of *P. miliaceum*. Different letters in each column indicates significant different values for  $p < 0.05$  (LSD test).

| Accession |  | Treatment | Measurement date                                              |        |       | Measurement date                                                                       |       |       |
|-----------|--|-----------|---------------------------------------------------------------|--------|-------|----------------------------------------------------------------------------------------|-------|-------|
|           |  |           | 12.06                                                         | 24.06  | 01.07 | 12.06                                                                                  | 24.06 | 01.07 |
|           |  |           | Gs<br>(mol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> ) |        |       | E<br>(mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> )                          |       |       |
| PM15      |  | T100      | 109.7a                                                        | 124.6a | 79.4a | 4.2a                                                                                   | 4.2a  | 2.8a  |
| PM15      |  | T50       | 21.1bc                                                        | 3.9c   | -     | 1.3c                                                                                   | 0.9c  | -     |
| PM16      |  | T100      | 41.2b                                                         | 36.7b  | 6.0b  | 2.9b                                                                                   | 2.7b  | 0.9b  |
| PM16      |  | T50       | 9.1c                                                          | 5.4c   | -     | 0.6c                                                                                   | 0.8c  | -     |
|           |  |           | PN<br>(μmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> ) |        |       | WUEi<br>(μmol CO <sub>2</sub> /mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> ) |       |       |
|           |  |           | 12.06                                                         | 24.06  | 01.07 | 12.06                                                                                  | 24.06 | 01.07 |
|           |  |           | Gs<br>(mol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> ) |        |       | E<br>(mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> )                          |       |       |
| PM15      |  | T100      | 7.6a                                                          | 7.4a   | 7.6a  | 7.4a                                                                                   | 7.6a  | 7.4a  |
| PM15      |  | T50       | 1.8c                                                          | 1.8c   | 1.8c  | 1.8c                                                                                   | 1.8c  | 1.8c  |
| PM16      |  | T100      | 5.2b                                                          | 4.4b   | 5.2b  | 4.4b                                                                                   | 5.2b  | 4.4b  |
| PM16      |  | T50       | 0.9c                                                          | 0.8c   | 0.9c  | 0.8c                                                                                   | 0.9c  | 0.8c  |

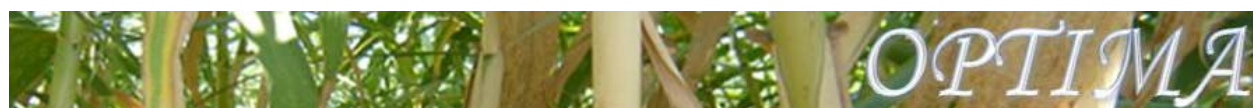


**Table 6 – CNR-ISPAAM.** Relative water content (RWC) of leaves of *P. miliaceum* under four regimes of drought stress

| Accession         | Treatment        | RWC             |
|-------------------|------------------|-----------------|
| P. miliaceum PM15 | T <sub>100</sub> | 92.8 $\pm$ 1.94 |
| P. miliaceum PM15 | T <sub>50</sub>  | 86.0 $\pm$ 6.4  |
| P. miliaceum PM15 | T <sub>25</sub>  | 78.5 $\pm$ 16.0 |
| P. miliaceum PM15 | T <sub>0</sub>   | 66.1 $\pm$ 19.3 |
| P. miliaceum PM16 | T <sub>100</sub> | 91.4 $\pm$ 4.9  |
| P. miliaceum PM16 | T <sub>50</sub>  | 84.1 $\pm$ 6.6  |
| P. miliaceum PM16 | T <sub>25</sub>  | 79.0 $\pm$ 8.5  |
| P. miliaceum PM16 | T <sub>0</sub>   | 62.1 $\pm$ 13.4 |

**Table 7 – CNR-ISPAAM.** Differences induced by mild (T50), severe (T25) and very severe (T0) water stress on some traits of *P. miliaceum*. Values refer to mean values and range (in brackets).

| Accession | Treatment | Tillers height (cm) | N. tillers           | Roots DW (g)       | Tillers DW (g)      | Panicles DW (g)  | Root to shoot ratio |
|-----------|-----------|---------------------|----------------------|--------------------|---------------------|------------------|---------------------|
| PM15      | T100      | 160.0<br>(153-167)  | 26.3<br>(24.7-27.9)  | 11,1<br>(9.3-12.8) | 31.6<br>(28-35.2)   | 4.6<br>(4.4-4.8) | 0.3                 |
| PM15      | T50       | 154.0<br>(147-161)  | 15.7<br>(14.1-17.3)  | 8,5<br>(6.8-10.2)  | 15.1<br>(11.5-18.7) | 1.4<br>(1.0-1.9) | 0.4                 |
| PM15      | T25       | 154.3<br>(148-161)  | 11.0<br>(9.4-12.6)   | 7.3<br>(5.6-9)     | 19.8<br>(16.2-23.4) | 1.4<br>(1.0-1.6) | 0.3                 |
| PM15      | T0        | 116.0<br>(109-123)  | 12.33<br>(10.7-13.9) | 7.6<br>(5.9-9.4)   | 11.5<br>(7.9-15.1)  | 0.8<br>(0.6-1.1) | 0.4                 |
| PM16      | T100      | 161,7<br>(155-168)  | 15.3<br>(13.7-16.9)  | 8.3<br>(6.6-10.1)  | 29.9<br>(26.3-33.5) | 0.8<br>(0.6-1.0) | 0.2                 |
| PM16      | T50       | 155.3<br>(149-162)  | 13.7<br>(12.1-15.2)  | 7.5<br>(5.8-9.2)   | 18.8<br>(15.2-22.4) | 1.1<br>(0.9-1.3) | 0.3                 |
| PM16      | T25       | 136.0<br>(129-143)  | 12.7<br>(11.1-14.3)  | 5.8<br>(4.1-7.5)   | 14.0<br>(10.4-17.6) | 0.2<br>(0.1-0.5) | 0.3                 |
| PM16      | T0        | 110.7<br>(104-117)  | 13.0<br>(11.4-14.6)  | 4.7<br>(3.0-6.5)   | 11.4<br>(7.8-15.0)  | 0.1<br>(0.1-0.3) | 0.3                 |



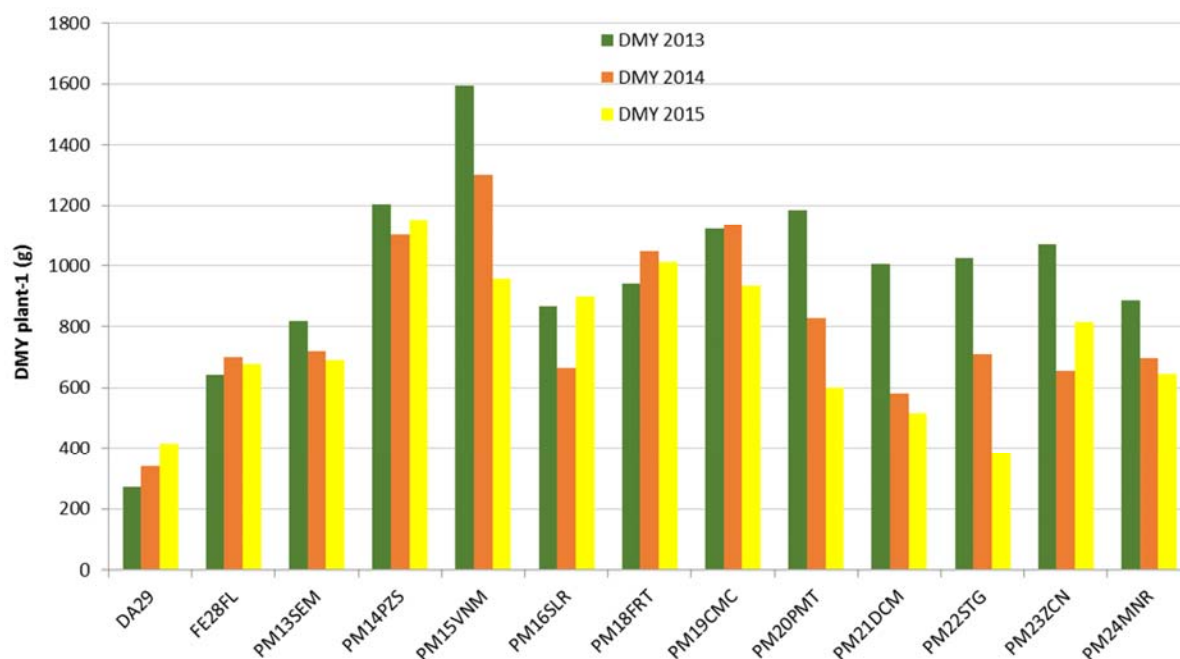
**Table 8 – CNR-ISPAAM.** Biometric traits of tillers, heads and flag leaf of accessions of *P. miliaceum*.

| Accession      | Tiller       |               | Head length<br>(cm) | Flag leaf   |            |
|----------------|--------------|---------------|---------------------|-------------|------------|
|                | Length (cm)  | No internodes |                     | Length (cm) | Width (cm) |
| PM13SEM        | 107.3        | 7.4           | 39.3 ab             | 25.1 c      | 0.7        |
| PM14PZS        | 116.3        | 7.5           | 44.3 a              | 30.9 ab     | 0.8        |
| PM16SLR        | 122.2        | 7.4           | 39.3 ab             | 32.2 a      | 0.8        |
| PM18FRT        | 112.4        | 7.1           | 43.7 ab             | 30.5 ab     | 0.7        |
| PM19CMC        | 119.2        | 7.1           | 39.8 ab             | 28.8 abc    | 0.7        |
| PM20PMT        | 119.2        | 6.7           | 40.8 ab             | 28.5 abc    | 0.6        |
| PM21DCM        | 118.0        | 7.2           | 41.9 ab             | 31.0 ab     | 0.7        |
| PM22STG        | 105.7        | 6.7           | 36.3 b              | 25.8 c      | 0.7        |
| PM23ZCN        | 119.8        | 7.5           | 43.2 ab             | 27.0 bc     | 0.7        |
| PM24MNR        | 113.4        | 7.6           | 39.3 ab             | 27.8 bc     | 0.6        |
| <i>average</i> | <b>115.4</b> | <b>7.2</b>    | <b>40.8</b>         | <b>28.8</b> | <b>0.7</b> |
| <i>minimum</i> | <b>105.7</b> | <b>6.7</b>    | <b>36.3</b>         | <b>25.1</b> | <b>0.6</b> |
| <i>maximum</i> | <b>122.2</b> | <b>7.6</b>    | <b>44.3</b>         | <b>32.2</b> | <b>0.8</b> |

**Table 9 – CNR-ISPAAM.** Dry weight and dry matter content in tillers, heads and flag leaf in accessions of *P. miliaceum*.

| Accession      | Tiller     |             | Head       |             | Flag leaf  |             |
|----------------|------------|-------------|------------|-------------|------------|-------------|
|                | DW (g)     | DM %        | DW (g)     | DM%         | DW (g)     | DM %        |
| PM13SEM        | 3.11       | 52.4        | 0.78       | 69.2        | 1.18 ab    | 82.7        |
| PM14PZS        | 3.92       | 46.7        | 0.87       | 60.1        | 1.38 a     | 73.5        |
| PM16SLR        | 3.57       | 42.5        | 0.71       | 57.1        | 1.23 ab    | 69.9        |
| PM18FRT        | 3.14       | 44.6        | 0.74       | 63.4        | 1.19 ab    | 71.6        |
| PM19CMC        | 3.57       | 41.9        | 0.82       | 54.0        | 1.10 ab    | 69.9        |
| PM20PMT        | 3.70       | 53.8        | 0.90       | 67.5        | 1.06 ab    | 84.7        |
| PM21DCM        | 3.03       | 47.4        | 0.72       | 62.2        | 1.06 ab    | 77.9        |
| PM22STG        | 2.84       | 49.9        | 0.82       | 64.4        | 1.13 ab    | 83.4        |
| PM23ZCN        | 3.90       | 50.5        | 0.94       | 64.7        | 1.06 ab    | 80.3        |
| PM24MNR        | 2.77       | 46.3        | 0.67       | 60.7        | 0.94 b     | 77.6        |
| <i>average</i> | <b>3.4</b> | <b>47.6</b> | <b>0.8</b> | <b>62.3</b> | <b>1.1</b> | <b>77.1</b> |
| <i>minimum</i> | <b>2.8</b> | <b>41.9</b> | <b>0.7</b> | <b>54.0</b> | <b>0.9</b> | <b>69.9</b> |
| <i>maximum</i> | <b>3.9</b> | <b>53.8</b> | <b>0.9</b> | <b>69.2</b> | <b>1.4</b> | <b>84.7</b> |

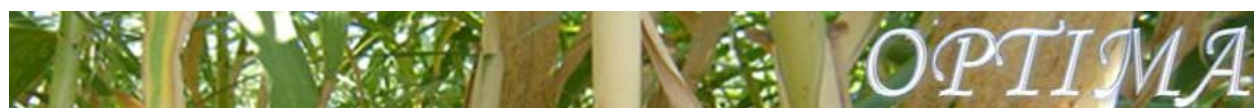




**Figure 4 – CNR-ISPAAAM.** Dry matter yield of native Sardinian accessions of *P. miliaceum* in 2013, 2014, 2015 compared to conventional forage grasses *D. glomerata* and *F. arundinacea*.

**Table 10 – CNR-ISPAAAM.** Number of tillers, water content and phytomass partitioning in *P. miliaceum*.

| Accession | No. of tillers |       |      | Water content (%) |      |      | Leaves ratio (%) |      |      | Tillers ratio (%) |      |      |
|-----------|----------------|-------|------|-------------------|------|------|------------------|------|------|-------------------|------|------|
|           | 2013           | 2014  | 2015 | 2013              | 2014 | 2015 | 2013             | 2014 | 2015 | 2013              | 2014 | 2015 |
| PM13SEM   | 244.2          | 245.0 | 259  | 37.0              | 39.9 | 47.3 | 28.3             | 16.0 | 17.5 | 71.7              | 84.0 | 82.4 |
| PM14PZS   | 292.7          | 319.7 | 324  | 35.1              | 34.5 | 36.5 | 27.1             | 13.2 | 23.8 | 72.9              | 86.8 | 76.2 |
| PM15VNM   | 300.5          | 246.0 | 255  | 29.3              | 36.2 | 40.2 | 21.0             | 18.0 | 25.4 | 79.0              | 82.0 | 74.6 |
| PM16SLR   | 224.7          | 203.3 | 288  | 32.1              | 39.9 | 38.4 | 26.7             | 12.3 | 22.8 | 73.3              | 87.7 | 77.2 |
| PM17SNS   | n.a.           | n.a.  | 306  | n.a.              | n.a. | 36.7 | n.a.             | n.a. | 23.1 | n.a.              | n.a. | 76.9 |
| PM18FRT   | 283.0          | 416.0 | 296  | 36.0              | 42.9 | 41.7 | 29.4             | 15.0 | 25.1 | 70.6              | 85.0 | 74.9 |
| PM19CMC   | 279.7          | 395.7 | 318  | 36.2              | 40.4 | 36.2 | 24.3             | 14.1 | 18.1 | 75.7              | 85.9 | 81.8 |
| PM20PMT   | 316.6          | 328.7 | 277  | 31.0              | 39.3 | 39.1 | 24.0             | 17.4 | 23.3 | 76.0              | 82.6 | 76.6 |
| PM21DCM   | 282.3          | 260.3 | 219  | 36.3              | 38.8 | 39.8 | 25.8             | 17.3 | 23.9 | 74.2              | 82.7 | 76.1 |
| PM22STG   | 296.0          | 273.3 | 213  | 36.1              | 43.0 | 44.6 | 25.8             | 14.3 | 19.4 | 74.2              | 85.7 | 80.6 |
| PM23ZCN   | 288.8          | 260.8 | 277  | 32.2              | 39.5 | 40.5 | 27.0             | 17.8 | 21.0 | 73.0              | 82.2 | 79.0 |
| PM24MNR   | 241.3          | 292.8 | 262  | 27.8              | 39.4 | 36.1 | 26.5             | 12.9 | 21.7 | 73.5              | 87.1 | 78.3 |

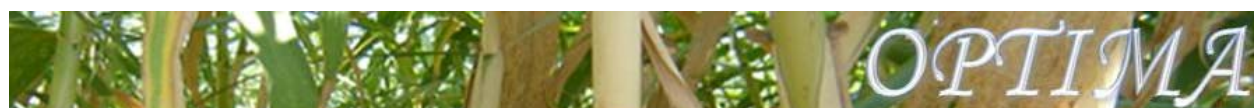


**Table 11 – CNR-ISPAAM.** Number of tillers, Dry matter yield per plant and phytomass partitioning in *A. mauritanicus*.

| Accession | No. of tillers |      | DMY per plant (g) |        | Water content (%) |      | Leaves ratio (%) |       | Tillers ratio (%) |      |
|-----------|----------------|------|-------------------|--------|-------------------|------|------------------|-------|-------------------|------|
|           | 2014           | 2015 | 2014              | 2015   | 2014              | 2015 | 2014             | 2015  | 2014              | 2015 |
| AM1MGR    | 48             | 71   | 251.1             | 261.8  | 46.4              | 36.1 | n.a.             | 100.0 | n.a.              | 0.0  |
| AM2GRG    | 42             | 90   | 179.5             | 453.0  | 45.8              | 40   | n.a.             | 77.5  | n.a.              | 22.5 |
| AM3SRT    | 97             | 156  | 803.6             | 783.8  | 44.6              | 44   | n.a.             | 67.0  | n.a.              | 33   |
| AM4MNR    | 154            | 190  | 1870.2            | 1633.8 | 43.9              | 46.9 | n.a.             | 71.2  | n.a.              | 28.8 |
| AM5DLV    | 7              | 35   | 52.3              | n.a.   | 47.1              | 37.7 | n.a.             | 86.1  | n.a.              | 9.3  |
| AM6CPF    | 174            | 95   | 561.3             | 563.8  | 48.0              | 40.9 | n.a.             | 94.0  | n.a.              | 6.0  |
| AM7SMV    | 163            | 141  | 1037.9            | 666.5  | 42.7              | 39.9 | n.a.             | 56.3  | n.a.              | 43.7 |
| AM8FRT    | 94             | 161  | 421.7             | 643.2  | 42.3              | 38.9 | n.a.             | 73.6  | n.a.              | 26.4 |
| AM9STG    | 146            | 119  | 833.3             | 730.7  | 41.6              | 42.8 | n.a.             | 58.3  | n.a.              | 41.7 |
| AM10FLN   | 167            | 147  | 2028.5            | 1153.1 | 43.6              | 44.2 | n.a.             | 42.2  | n.a.              | 42.8 |
| AM11CMC   | 99             | 117  | 677.2             | 648.1  | 47.5              | 38.4 | n.a.             | 70.0  | n.a.              | 30.0 |
| AM12PMT   | 100            | 176  | 750.7             | 767.4  | 43.0              | 39.3 | n.a.             | 76.1  | n.a.              | 23.9 |

**Table 12 – CNR-ISPAAM.** Dry matter yield per plant, water content, number of tillers and biomass partitioning in accessions of *A. mauritanicus* (AM), *D. glomerata* (DA), *P. aquatica* (FA), *F. arundinacea* (FE), *L. perenne* (LO), and *P. miliaceum* (PM).

| Accession | DMY plant <sup>-1</sup> (g) |     | WC (%) |     | N. tillers |     | Leaves ratio (%) |     | Tillers ratio (%) |     |
|-----------|-----------------------------|-----|--------|-----|------------|-----|------------------|-----|-------------------|-----|
| AM01MGR   | 261.8                       | i-m | 36.1   | a-d | 71         | mn  | 100.0            | a   | 0.0               | i   |
| AM02GRG   | 453.0                       | f-m | 40.0   | a-d | 90         | l-n | 77.5             | bc  | 22.5              | gh  |
| AM03SRT   | 783.8                       | b-h | 44.0   | a-c | 156        | f-n | 67.0             | b-d | 33                | e-g |
| AM04MNR   | 1633.8                      | a   | 46.9   | ab  | 190        | d-l | 71.2             | bc  | 28.8              | gh  |
| AM05DLV   | NA                          | -   | 37.7   | g   | 35         | n   | 86.1             | b   | 9.3               | hi  |
| AM06CPF   | 563.8                       | d-m | 40.9   | a-c | 95         | l-n | 94.0             | b-d | 6.0               | f-h |
| AM07SMV   | 666.5                       | c-i | 39.9   | a-d | 141        | h-n | 56.3             | c-e | 43.7              | d-g |
| AM08FRT   | 643.2                       | d-m | 38.9   | a-d | 161        | f-n | 73.6             | bc  | 26.4              | gh  |
| AM09STG   | 730.7                       | b-i | 42.8   | a-c | 119        | h-n | 58.3             | b-d | 41.7              | e-g |
| AM10FLN   | 1153.1                      | a-c | 44.2   | a-c | 147        | g-n | 42.2             | hi  | 42.8              | d-g |
| AM11CMC   | 648.1                       | d-l | 38.4   | a-d | 117        | h-n | 70.0             | b-d | 30.0              | e-g |
| AM12PMT   | 767.4                       | b-h | 39.3   | a-d | 176        | e-m | 76.1             | bc  | 23.9              | gh  |
| DA29JA    | 415.0                       | g-m | 27.5   | de  | 145        | g-n | 32.7             | f-h | 67.2              | b-d |
| DA30OT    | 159.8                       | lm  | 21.8   | ef  | 101        | i-n | 35.1             | f-h | 64.9              | b-d |
| FA31AU    | 468.8                       | e-m | 41.2   | a-c | 161        | f-n | 30.2             | f-h | 69.9              | b-d |
| FE25TA    | 598.6                       | d-m | 32.4   | c-e | 249        | a-g | 55.8             | c-g | 44.2              | c-g |
| FE26SI    | 568.1                       | d-m | 37.2   | a-d | 268        | a-e | 30.4             | f-h | 69.6              | b-d |
| FE27CE    | 445.9                       | f-m | 42.6   | a-c | 205        | c-i | 41.4             | e-h | 58.5              | b-e |
| FE28FL    | 678.2                       | c-i | 33.3   | b-e | 202        | c-i | 42.3             | d-h | 57.6              | b-f |
| LO32AZ    | 30.0                        | m   | 13.4   | f   | 114        | h-n | 8.2              | i   | 91.8              | a   |
| PM13SEM   | 690.7                       | c-i | 47.3   | a   | 259        | a-f | 17.5             | hi  | 82.4              | ab  |
| PM14PZS   | 1152.8                      | a-c | 36.5   | a-d | 324        | a   | 23.8             | hi  | 76.2              | ab  |
| PM15VNM   | 956.4                       | b-e | 40.2   | a-d | 255        | a-f | 25.4             | g-i | 74.6              | a-c |
| PM16SLR   | 899.7                       | b-g | 38.4   | a-d | 288        | a-d | 22.8             | hi  | 77.2              | ab  |



|         |        |     |      |     |     |     |      |     |      |    |
|---------|--------|-----|------|-----|-----|-----|------|-----|------|----|
| PM17SNS | 1200.8 | ab  | 36.7 | a-d | 306 | a-c | 23.1 | hi  | 76.9 | ab |
| PM18FRT | 1013.2 | b-d | 41.7 | a-c | 296 | a-d | 25.1 | g-i | 74.9 | ab |
| PM19CMC | 934.6  | b-f | 36.2 | a-d | 318 | ab  | 18.1 | hi  | 81.8 | ab |
| PM20PMT | 599.9  | d-m | 39.1 | a-d | 277 | a-e | 23.3 | hi  | 76.6 | ab |
| PM21DCM | 516.4  | d-m | 39.8 | a-d | 219 | a-h | 23.9 | hi  | 76.1 | ab |
| PM22STG | 382.6  | h-m | 44.6 | a-c | 213 | b-h | 19.4 | hi  | 80.6 | ab |
| PM23ZCN | 813.4  | b-h | 40.5 | a-d | 277 | a-e | 21.0 | hi  | 79.0 | ab |
| PM24MNR | 644.8  | d-m | 36.1 | a-d | 262 | a-f | 21.7 | hi  | 78.3 | ab |

| Accession | date  |       |       |       |       |       |       |       |       |       |       |       |       |
|-----------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
|           | 05.04 | 12.04 | 19.04 | 26.04 | 03.05 | 10.05 | 17.05 | 24.05 | 31.05 | 07.06 | 14.06 | 21.06 | 28.06 |
| AM10FLN   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM11CMC   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM12PMT*  |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM1MGR*   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM2GRG    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM3SRT*   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM4MNR*   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM5DLV    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM6CPF    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM7SMV*   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM8FRT*   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| AM9STG*   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| DA29JA    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| DA30OT    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| FA31AU    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| FE25TA    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| FE26SI    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| FE27CE    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| FE28FL    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| LO32AZ    |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM13SEM   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM14PZS   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM15VNM   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM16SLR   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM17SNS   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM18FTR   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM19CMC   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM20PMT   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM21DCM   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM22STG   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM23ZCN   |       |       |       |       |       |       |       |       |       |       |       |       |       |
| PM24MNR   |       |       |       |       |       |       |       |       |       |       |       |       |       |

**Figure 5 – CNR-ISPAAAM.** Phenological stages of accessions growing in the field nursery of OPTIMA project in Sardinia. Stars indicates that only a negligible number of plants reached the flowering phase in each plot, the others remaining in a vegetative stage. The phenological stages are shown with different



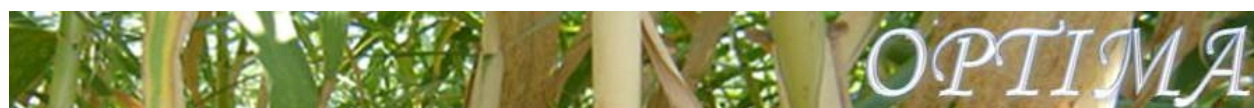
colours: in green the vegetative stage, in yellow the heading stage, in red the full flowering stage, in pink the end of flowering, in brown the development and ripening of seeds. *AM* indicates *A. mauritanicus*, *DA* *D. glomerata*, *FA* *P. aquatica*, *FE* *F. arundinacea*, *LO* *L. perenne*, *PM* *P. miliaceum* accessions.

**Table 13 – CNR-ISPAAM.** Content of cellulose, hemicellulose and lignin in *P. miliaceum*, at harvest time, in July 2013. Average values are the average of ten values (ten accessions).

| Plant organ   | Hemicellulose (%) |           | Cellulose (%) |           | Lignin (%) |           |
|---------------|-------------------|-----------|---------------|-----------|------------|-----------|
|               | mean              | range     | mean          | range     | mean       | range     |
| <b>Tiller</b> | 28.8              | 26.6-30.1 | 38.1          | 36.0-40.7 | 13.4       | 12.0-15.0 |
| <b>Leaf</b>   | 25.4              | 24.1-27.3 | 39.0          | 37.6-40.9 | 10.0       | 7.9-11.7  |
| <b>Spike</b>  | 34.5              | 32.8-36.2 | 30.2          | 26.0-33.2 | 12.8       | 10.7-15.7 |

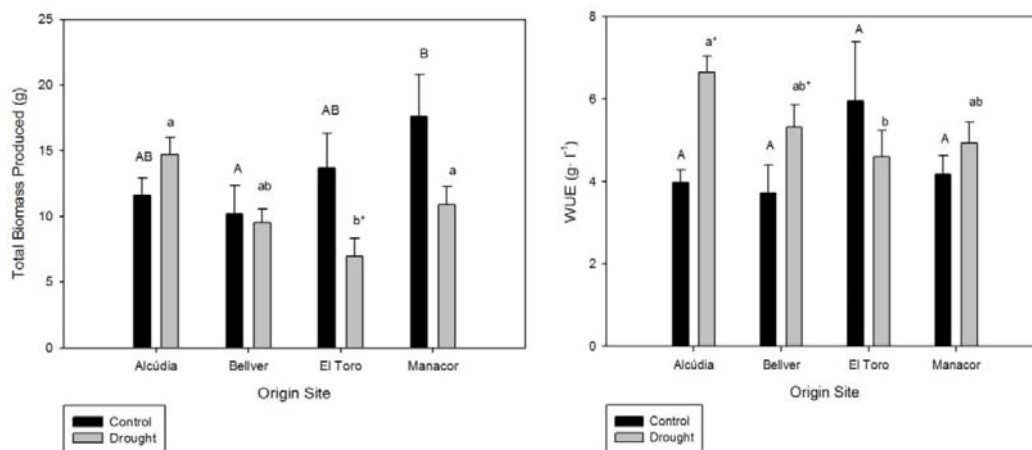
**Table 14 – CNR-ISPAAM.** Proximate and ultimate analysis of plant components in *P. miliaceum* vs *miscanthus*

| Species             | Calorific value (MJ kg <sup>-1</sup> ) | Volatile matter (%) | Fixed carbon (%) | Ash content (%) | C (%) | H (%) | N (%) | S (%) | Cl (%) | O (%) |
|---------------------|----------------------------------------|---------------------|------------------|-----------------|-------|-------|-------|-------|--------|-------|
| <i>Leaves</i>       |                                        |                     |                  |                 |       |       |       |       |        |       |
| <i>P. miliaceum</i> | 16.0                                   | 70.0                | 5.0              | 13.0            | 49.0  | 6.1   | 3.6   | <0.8  | 1.24   | 41.3  |
| <i>M. x</i>         | 16.0                                   | 73.0                | 1.7              | 9.5             | 49.0  | 6.3   | 2.7   | <0.3  | <0.03  | 42.0  |
| <i>Stalks</i>       |                                        |                     |                  |                 |       |       |       |       |        |       |
| <i>P. miliaceum</i> | 17.0                                   | 77.0                | 11.0             | 5.0             | 49.0  | 6.5   | 1.4   | <0.3  | 1.08   | 43.1  |
| <i>M. x</i>         | 17.0                                   | 77.0                | 3.6              | 6.0             | 50.0  | 6.3   | 1.7   | <0.3  | <0.03  | 42.0  |
| <i>Heads</i>        |                                        |                     |                  |                 |       |       |       |       |        |       |
| <i>P. miliaceum</i> | 16.0                                   | 69.0                | 1.5              | 12.0            | 48.0  | 6.0   | 2.6   | <0.8  | 0.98   | 43.4  |



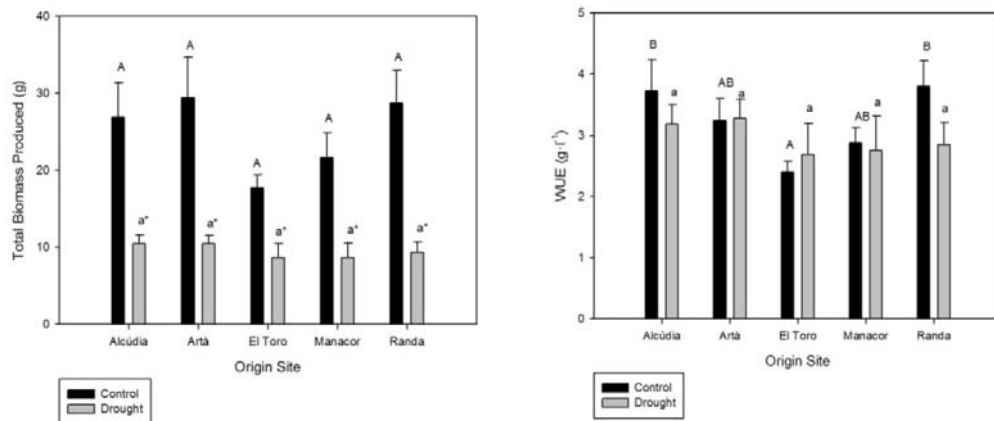


**Figure 1 – UIB.** Localization of the selected populations in the Island of Mallorca. *D. glomerata*: El Toro, Bellver, Alcúdia and Manacor; *P. miliaceum*: El Toro, Randa, Alcúdia, Artà and Manacor; *A. mauritanicus*: Bunyola, Randa, Alcúdia, Formentor, Cúber and Manacor.

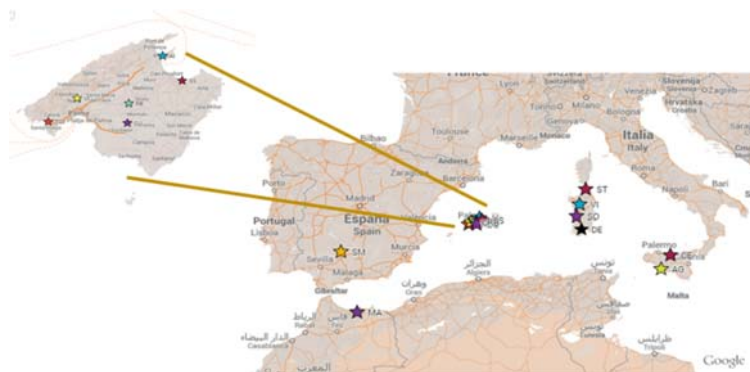


**Figure 2 – UIB.** Total biomass production and plant water use efficiency of four populations of *D. glomerata* under well-watered (black bars) and water-limited (grey-bars) conditions. Values are means of six replicates  $\pm$  standard error. Different letters show significant differences between populations within each treatment at  $p < 0.05$ . \*Shows significant differences between treatments within each population at  $p < 0.05$ .



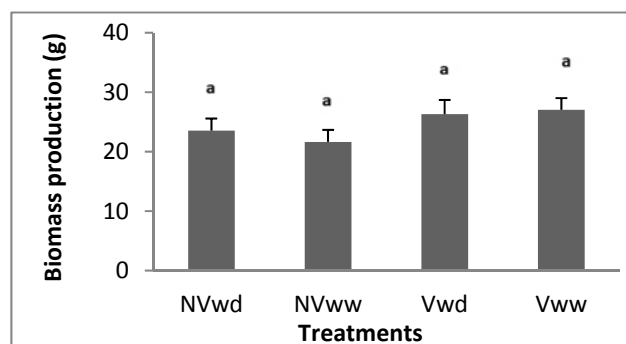


**Figure 3 – UIB.** Total biomass production and plant water use efficiency of four populations of *P. miliaceum* under well-watered (black bars) and water-limited (grey-bars) conditions. Values are means of six replicates  $\pm$  standard error. Different letters show significant differences between populations within each treatment at  $p < 0.05$ . \*Shows significant differences between treatments within each population at  $p < 0.05$ .

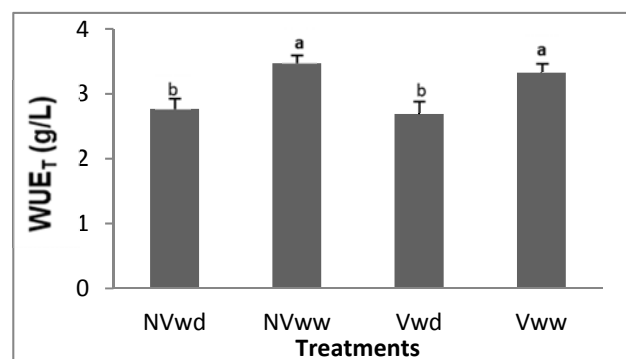


**Figure 4 – UIB.** Distribution of the sampled populations along the Western Mediterranean basin: Majorca (Toro, UIB, Alcúdia, Son Serra, Cura and Ruberts), Sicily (Cefalú and Agrigento), Sardinia (Santa Teresa, Solaruso, Vilanova and Décimo), Iberian Peninsula (Sierra Morena) and Morocco.

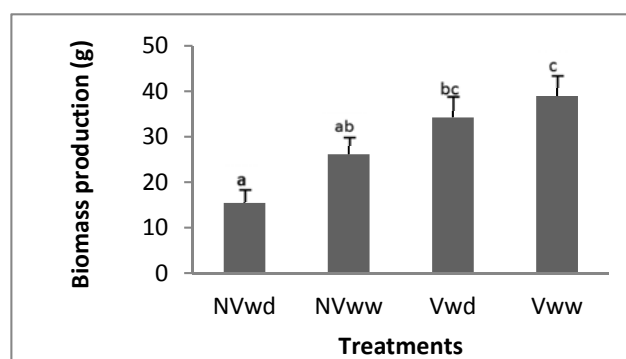




**Figure 5 – UIB:** Plant biomass production (g) of *D. glomerata* under four treatments: water-deficit plants without mycorrhizae (NVwd), well-watered plants without mycorrhizae (NVww), water-deficit plants with mycorrhizae (Vwd) and well-watered plants with mycorrhizae (Vww).

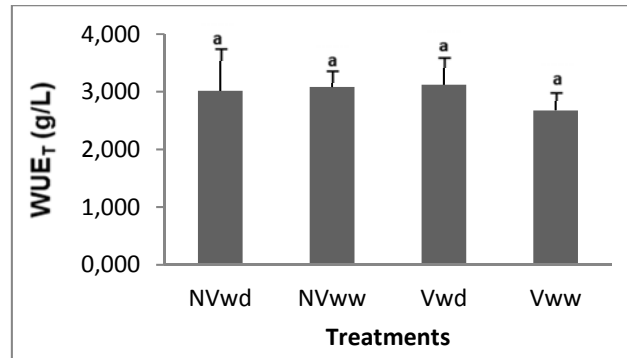


**Figure 6 – UIB:** Plant water use efficiency (WUE<sub>T</sub> calculated as total produced biomass per consumed water) (g/L) of *D. glomerata* under four treatments: water-deficit plants without mycorrhizae (NVwd), well-watered plants without mycorrhizae (NVww), water-deficit plants with mycorrhizae (Vwd) and well-watered plants with mycorrhizae (Vww).

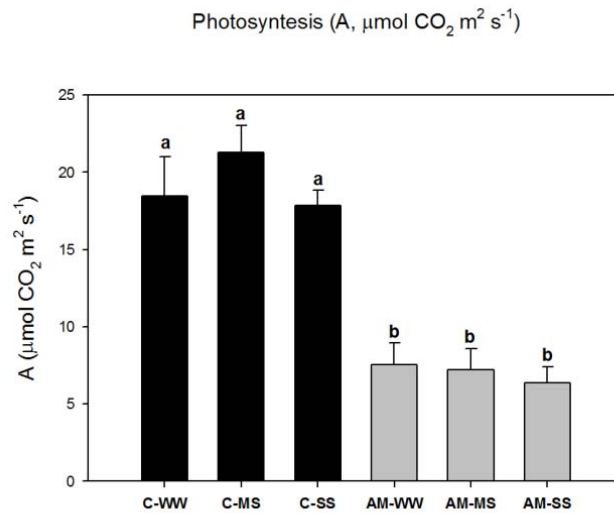


**Figure 7 – UIB:** Plant biomass production (g) of *P. miliaceum* under four treatments: water-deficit plants without mycorrhizae (NVwd), well-watered plants without mycorrhizae (NVww), water-deficit plants with mycorrhizae (Vwd) and well-watered plants with mycorrhizae (Vww).



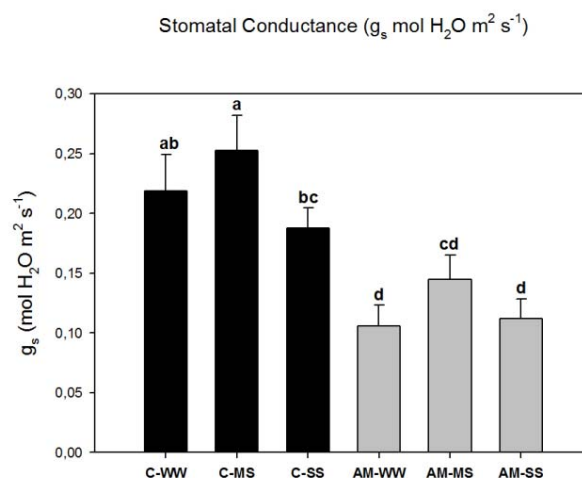


**Figure 8 – UIB:** Plant water use efficiency (WUE<sub>T</sub> calculated as total produced biomass per consumed water) (g/L) of *P. miliaceum* under four treatments: water-deficit plants without mycorrhizae (NVwd), well-watered plants without mycorrhizae (NVww), water-deficit plants with mycorrhizae (Vwd) and well-watered plants with mycorrhizae (Vww).



**Figure 9 – UIB.** Net photosynthesis of inoculated (AM) and non-inoculated (C) *Ampelodesmos mauritanicus* under different water regimes (VW: plants under 100% field capacity; MS: plants under 40% field capacity; SS: plants under 20% field capacity). Values are means of six replicates  $\pm$  s.e.





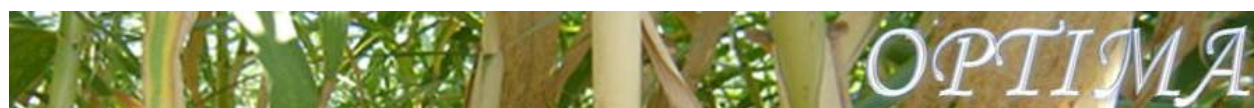
**Figure 10 – UIB.** Stomatal conductance of inoculated (AM) and non-inoculated (C) *Ampelodesmos mauritanicus* under different water regimes (WW: plants under 100% field capacity; MS: plants under 40% field capacity; SS: plants under 20% field capacity). Values are means of six replicates  $\pm$  s.e.

**Table 1 – UIB.** Biomass production (g m<sup>-2</sup>) of each species and population. Biomass was determined in one cut in 2013 and in two cuts in 2014. Different letters indicate statistical significant differences within each cut period at  $p < 0.05$ .

| Species               | Genotype | Dry Matter Yield (g m <sup>-2</sup> ) |                              |                                |
|-----------------------|----------|---------------------------------------|------------------------------|--------------------------------|
|                       |          | Summer 2013                           | Spring 2014                  | Summer 2014                    |
| <i>D. glomerata</i>   | Alcudia  | 236,9 $\pm$ 19,47 <sup>c</sup>        | 64.5 $\pm$ 2.93 <sup>b</sup> | 132.6 $\pm$ 1.67 <sup>b</sup>  |
|                       | Jana     | 189,1 $\pm$ 19,47 <sup>bc</sup>       | 66.0 $\pm$ 2.04 <sup>b</sup> | 129.3 $\pm$ 1.35 <sup>b</sup>  |
|                       | Manacor  | 218.4 $\pm$ 19,47 <sup>c</sup>        | 66.2 $\pm$ 3.15 <sup>b</sup> | 131.6 $\pm$ 2.14 <sup>b</sup>  |
| <i>F. arundinacea</i> | Flecha   | 69,3 $\pm$ 19,47 <sup>a</sup>         | 65.3 $\pm$ 3.51 <sup>b</sup> | 104.6 $\pm$ 16.60 <sup>a</sup> |
| <i>P. tomassi</i>     | Alcudia  | 151,7 $\pm$ 19,47 <sup>b</sup>        | 68.2 $\pm$ 3.23 <sup>b</sup> | 112.9 $\pm$ 1.24 <sup>ab</sup> |
| <i>P. miliaceum</i>   | Toro     | 236,1 $\pm$ 19,47 <sup>c</sup>        | 67.2 $\pm$ 3.79 <sup>b</sup> | 100.6 $\pm$ 2.01 <sup>a</sup>  |

**Table 2 – UIB.** Number of tillers per plant of each species and population in 2013 and 2014. Different letters indicate statistical significant differences within each period at  $p < 0.05$ .

| Species               | Genotype | N° tillers                    |                               |                               |                              |
|-----------------------|----------|-------------------------------|-------------------------------|-------------------------------|------------------------------|
|                       |          | July 2013                     | October 2013                  | July 2014                     | October 2014                 |
| <i>D. glomerata</i>   | Alcudia  | 22,4 $\pm$ 1,74 <sup>c*</sup> | 16,7 $\pm$ 2,07 <sup>b</sup>  | 70.2 $\pm$ 5.25 <sup>c</sup>  | 34.9 $\pm$ 3.64 <sup>b</sup> |
|                       | Jana     | 11,1 $\pm$ 1,74 <sup>a</sup>  | 25,1 $\pm$ 2,07 <sup>c</sup>  | 59.8 $\pm$ 8.80 <sup>bc</sup> | 30.5 $\pm$ 4.23 <sup>b</sup> |
|                       | Manacor  | 17,1 $\pm$ 1,74 <sup>b</sup>  | 15,0 $\pm$ 2,07 <sup>ab</sup> | 59.7 $\pm$ 6.25 <sup>bc</sup> | 40.4 $\pm$ 6.15 <sup>b</sup> |
| <i>F. arundinacea</i> | Flecha   | 16,5 $\pm$ 1,74 <sup>b</sup>  | 19,6 $\pm$ 2,07 <sup>bc</sup> | 66.2 $\pm$ 5.92 <sup>c</sup>  | 30.5 $\pm$ 5.84 <sup>b</sup> |
| <i>P. tomassi</i>     | Alcudia  | 35,9 $\pm$ 1,74 <sup>e</sup>  | 20,5 $\pm$ 2,07 <sup>bc</sup> | 40.8 $\pm$ 4.25 <sup>a</sup>  | 14.6 $\pm$ 3.63 <sup>a</sup> |
| <i>P. miliaceum</i>   | Toro     | 30,7 $\pm$ 1,74 <sup>d</sup>  | 9,8 $\pm$ 2,07 <sup>a</sup>   | 45.9 $\pm$ 2.79 <sup>ab</sup> | 5.8 $\pm$ 1.61 <sup>a</sup>  |



**Table 3 – UIB.** Biomass quality parameters (all parameters expressed in %). Values are means  $\pm$  standard error of four replicates. Different letters indicate statistical significant differences at  $p < 0.05$

| Species               | Genotype | Dry matter (%)                | Ash content (%)              | Cellulose (%)                | Hemicellulose (%)             | Lignin (%)                   |
|-----------------------|----------|-------------------------------|------------------------------|------------------------------|-------------------------------|------------------------------|
| <i>D. glomerata</i>   | Alcudia  | 50.8 $\pm$ 0.15 <sup>a</sup>  | 8.8 $\pm$ 0.04 <sup>cd</sup> | 36.7 $\pm$ 0.20 <sup>b</sup> | 21.2 $\pm$ 0.43 <sup>ab</sup> | 6.5 $\pm$ 0.12 <sup>cd</sup> |
|                       | Jana     | 51.6 $\pm$ 0.22 <sup>a</sup>  | 9.2 $\pm$ 0.18 <sup>cd</sup> | 37.1 $\pm$ 0.36 <sup>b</sup> | 20.5 $\pm$ 0.49 <sup>a</sup>  | 5.0 $\pm$ 0.18 <sup>b</sup>  |
|                       | Manacor  | 50.8 $\pm$ 0.05 <sup>a</sup>  | 8.6 $\pm$ 0.01 <sup>c</sup>  | 36.4 $\pm$ 0.38 <sup>b</sup> | 22.2 $\pm$ 0.34 <sup>bc</sup> | 5.8 $\pm$ 0.39 <sup>c</sup>  |
| <i>F. arundinacea</i> | Flecha   | 54.5 $\pm$ 1.17 <sup>b</sup>  | 9.2 $\pm$ 0.01 <sup>cd</sup> | 37.6 $\pm$ 0.40 <sup>b</sup> | 23.3 $\pm$ 0.45 <sup>c</sup>  | 3.3 $\pm$ 0.09 <sup>a</sup>  |
| <i>P. tomassi</i>     | Alcudia  | 56.2 $\pm$ 0.42 <sup>bc</sup> | 6.8 $\pm$ 0.13 <sup>a</sup>  | 36.5 $\pm$ 0.54 <sup>b</sup> | 27.0 $\pm$ 0.45 <sup>d</sup>  | 6.8 $\pm$ 0.15 <sup>d</sup>  |
| <i>P. miliaceum</i>   | Toro     | 57.4 $\pm$ 0.57 <sup>c</sup>  | 7.5 $\pm$ 0.06 <sup>b</sup>  | 35.0 $\pm$ 0.34 <sup>a</sup> | 29.3 $\pm$ 0.36 <sup>e</sup>  | 6.0 $\pm$ 0.13 <sup>c</sup>  |

**Table 4 – UIB.** Biomass production of *A. donax*, *A. mauritanicus* and *P. virgatum*. Different letters indicate statistical significant differences at  $p < 0.05$ .

| Species                | Fresh Biomass (g m <sup>-2</sup> ) | Dry Biomass (g m <sup>-2</sup> ) |
|------------------------|------------------------------------|----------------------------------|
| <i>A. donax</i>        | 1883.3 $\pm$ 114.93 <sup>b</sup>   | 1154.8 $\pm$ 77.91 <sup>b</sup>  |
| <i>A. mauritanicus</i> | 1398.9 $\pm$ 126.3 <sup>a</sup>    | 649.7 $\pm$ 63.7 <sup>a</sup>    |
| <i>P. virgatum</i>     | 1078.3 $\pm$ 86.07 <sup>a</sup>    | 535.9 $\pm$ 48.9 <sup>a</sup>    |

**Table 5 – UIB.** Biomass quality parameters of *A. donax*, *A. mauritanicus* and *P. virgatum*. (all parameters expressed in %). Values are means  $\pm$  standard error of four replicates. Different letters indicate statistical significant differences at  $p < 0.05$

| Species                | Dry matter                   | Ash content                 | Cellulose                    | Hemicellulose                | Lignin                      |
|------------------------|------------------------------|-----------------------------|------------------------------|------------------------------|-----------------------------|
| <i>A. donax</i>        | 42.5 $\pm$ 0.91 <sup>a</sup> | 6.1 $\pm$ 0.44 <sup>b</sup> | 44.5 $\pm$ 0.35 <sup>b</sup> | 20.9 $\pm$ 0.38 <sup>a</sup> | 9.8 $\pm$ 0.13 <sup>c</sup> |
| <i>A. mauritanicus</i> | 56.6 $\pm$ 1.01 <sup>c</sup> | 6.7 $\pm$ 0.02 <sup>c</sup> | 39.3 $\pm$ 0.36 <sup>a</sup> | 28.3 $\pm$ 1.08 <sup>b</sup> | 5.3 $\pm$ 0.15 <sup>a</sup> |
| <i>P. virgatum</i>     | 53.8 $\pm$ 0.73 <sup>b</sup> | 5.0 $\pm$ 0.09 <sup>a</sup> | 47.4 $\pm$ 0.09 <sup>c</sup> | 28.9 $\pm$ 0.56 <sup>b</sup> | 8.5 $\pm$ 0.09 <sup>b</sup> |



## Center for Renewable Energy Sources and Energy Saving – CRES (n. 3)



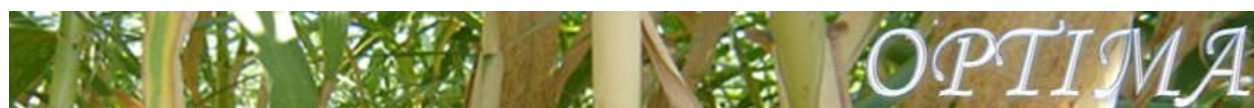
**Figure 1 – CRES.** Experimental layout of the screening trial (eight perennial grasses in one replication)



**Figure 2 –CRES.** View of the perennial grasses that were compared in sub-task 3.1.3 in Greece.

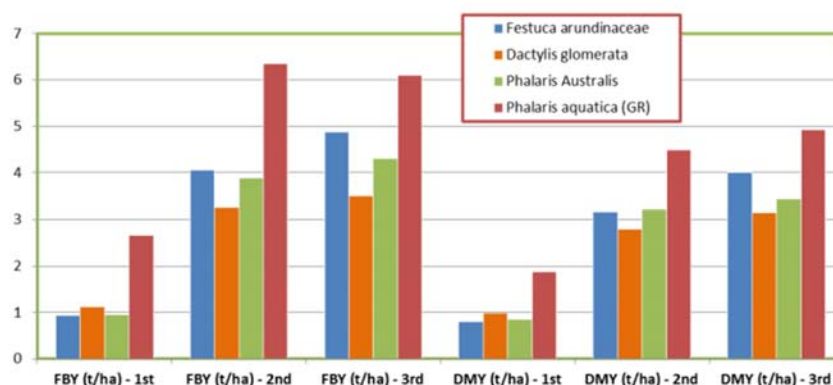
**Table 2 – CRES.** Growth (height) and yields data (fresh and dry matter yields) for the four endemic grasses for three subsequent years (2013-5).

| Endemic grasses               | Height (cm) |      |      | Fresh biomass yields (t/ha) |      |      | Dry matter yields (t/ha) |      |      |
|-------------------------------|-------------|------|------|-----------------------------|------|------|--------------------------|------|------|
|                               | 2013        | 2014 | 2015 | 2013                        | 2014 | 2015 | 2013                     | 2014 | 2015 |
| <i>Festuca arundinacea</i>    | 150         | 150  | 150  | 0.94                        | 4.05 | 4.88 | 0.80                     | 3.16 | 4.00 |
| <i>Dactylis glomerata</i>     | 30          | 50   | 50   | 1.12                        | 3.25 | 3.50 | 0.99                     | 2.80 | 3.15 |
| <i>Phalaris Australis</i>     | 140         | 150  | 140  | 0.95                        | 3.88 | 4.30 | 0.85                     | 3.22 | 3.44 |
| <i>Phalaris aquatica</i> (GR) | 108         | 140  | 143  | 2.67                        | 6.35 | 6.10 | 1.87                     | 4.49 | 4.93 |

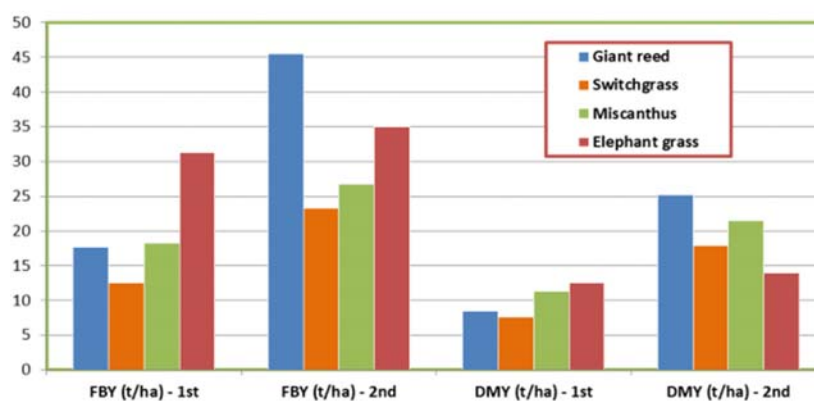


**Table 3 – CRES.** Growth (height and tiller density) and yields data (fresh and dry matter yields) for the four known perennial grasses for two growing periods (2013-4 & 2014-5).

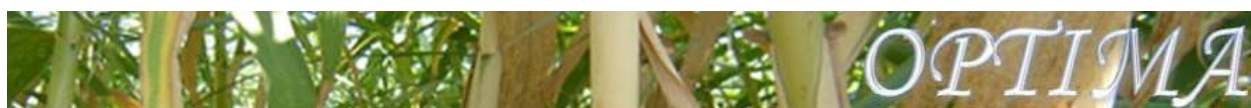
| Known perennial grasses | Height (cm) |      | Tiller density (tillers/m <sup>2</sup> ) |        | Fresh biomass yields (t/ha) |       | Dry matter yields (t/ha) |       |
|-------------------------|-------------|------|------------------------------------------|--------|-----------------------------|-------|--------------------------|-------|
|                         | 2014        | 2015 | 2014                                     | 2015   | 2014                        | 2015  | 2014                     | 2015  |
| Giant reed              | 415         | 616  | 7.96                                     | 17.00  | 17.70                       | 45.50 | 8.50                     | 25.16 |
| Switchgrass             | 168         | 294  | 410.00                                   | 387.50 | 12.50                       | 23.25 | 7.63                     | 17.90 |
| Miscanthus              | 312         | 360  | 55.44                                    | 58.00  | 18.30                       | 26.80 | 11.30                    | 21.50 |
| Elephant grass          | 318         | 414  | 9.50                                     | 23.50  | 31.25                       | 35.00 | 12.50                    | 13.93 |



**Figure 3 – CRES.** Yields (t/ha, as harvested and oven-dried) for four endemic grasses from 2013-5.

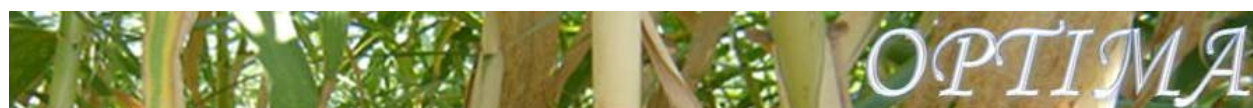


**Figure 4 – CRES.** Yields (t/ha, as harvested and oven-dried) for four known perennial grasses for two subsequent growing seasons (2013-4 & 2014-5).



**Table 4 – CRES.** Results from the calorific value (GCY, NCY) and proximate analysis (volatiles, ash and fixed carbon) on samples that were collected at the end of the second growing period.

| Perennial grasses                     | Calorific value<br>(kcal/kg) |                      | Proximate analysis<br>(%) |                       |                         | Elemental analysis (%)  |                      |                      |
|---------------------------------------|------------------------------|----------------------|---------------------------|-----------------------|-------------------------|-------------------------|----------------------|----------------------|
|                                       | GCV                          | NCV                  | Volatiles                 | Ash                   | Fixed C                 | C                       | H                    | N                    |
| Phalaris aquatica                     | 4167                         | 3897                 | 70.88                     | 12.24                 | 15.77                   | 41.72                   | 5.36                 | 0.71                 |
| Dactylis glomerata                    | 4181                         | 3878                 | 70.67                     | 13.05                 | 16.08                   | 40.86                   | 5.55                 | 0.68                 |
| Festuca arundinaceae                  | 4180                         | 3901                 | 72.05                     | 12.14                 | 15.77                   | 40.53                   | 5.62                 | 0.61                 |
| Phalaris aquatica (GR)                | 4133                         | 3834                 | 70.67                     | 11.20                 | 18.13                   | 40.91                   | 5.93                 | 0.76                 |
| Elephant grass (stems, leaves, whole) | 4378<br>41.48<br>4355        | 4069<br>3840<br>4046 | 74.12<br>72.41<br>73.95   | 7.39<br>12.08<br>7.86 | 18.50<br>15.52<br>18.20 | 44.23<br>41.28<br>43.94 | 6.10<br>6.09<br>6.10 | 0.75<br>1.62<br>0.84 |
| Switchgrass (stems, leaves, whole)    | 4471<br>4325<br>4413         | 4165<br>4029<br>4111 | 80.22<br>78.56<br>79.56   | 1.85<br>7.86<br>4.25  | 17.94<br>13.59<br>16.20 | 47.72<br>43.26<br>45.94 | 6.06<br>5.86<br>5.98 | 0.28<br>0.64<br>0.42 |
| Giant reed (stems, leaves, whole)     | 4484<br>4325<br>4452         | 4176<br>4029<br>4147 | 77.04<br>73.52<br>76.34   | 3.56<br>9.95<br>4.84  | 19.41<br>16.53<br>18.83 | 46.56<br>43.04<br>45.86 | 6.10<br>5.83<br>6.05 | 0.54<br>1.42<br>0.72 |
| Miscanthus (stems, leaves, whole)     | 4575                         | 4255                 | 81.08                     | 1.78                  | 11.13                   | 47.63                   | 6.34                 | 0.19                 |



### Task 3.2 - Good agricultural practices for perennial grasses

University of Bologna – UNIBO (n. 2)

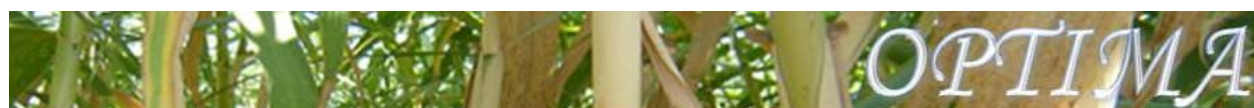


**Figure 1 - UNIBO.** How a fully established switchgrass crop (Alamo) appeared at two row distances: 20 cm (left), 80 cm (right).



**Table 1 - UNIBO.** Main results from switchgrass density trial established in Cadriano in 2002.

| Year | Inter row<br>(m) | Aboveground biomass<br>FM (Mg ha <sup>-1</sup> ) | Moisture % | Aboveground biomass DM<br>(Mg ha <sup>-1</sup> ) |
|------|------------------|--------------------------------------------------|------------|--------------------------------------------------|
| 2009 | 0.20             | 60.1                                             | 56.5       | 26.1                                             |
|      | 0.80             | 57.4                                             | 57.4       | 24.7                                             |
| 2010 | 0.20             | 53.6                                             | 57.9       | 22.8                                             |
|      | 0.80             | 52.6                                             | 56.9       | 22.4                                             |
| 2011 | 0.20             | 45.1                                             | 55         | 20.2                                             |
|      | 0.80             | 45.3                                             | 54.2       | 20.7                                             |
| 2012 | 0.20             | 37.2                                             | 57.5       | 15.8                                             |
|      | 0.80             | 42.0                                             | 57.1       | 18.0                                             |
| 2013 | 0.20             | 35.5                                             | 56.5       | 15.4                                             |
|      | 0.80             | 34.1                                             | 57.7       | 14.4                                             |
| 2014 | 0.20             | 62.08                                            | 59.38      | 25.26                                            |
|      | 0.80             | 54.90                                            | 58.68      | 22.59                                            |
| 2015 | 0.20             | 37.3                                             | 59.5       | 13.6                                             |
|      | 0.80             | 31.6                                             | 57.4       | 15.4                                             |



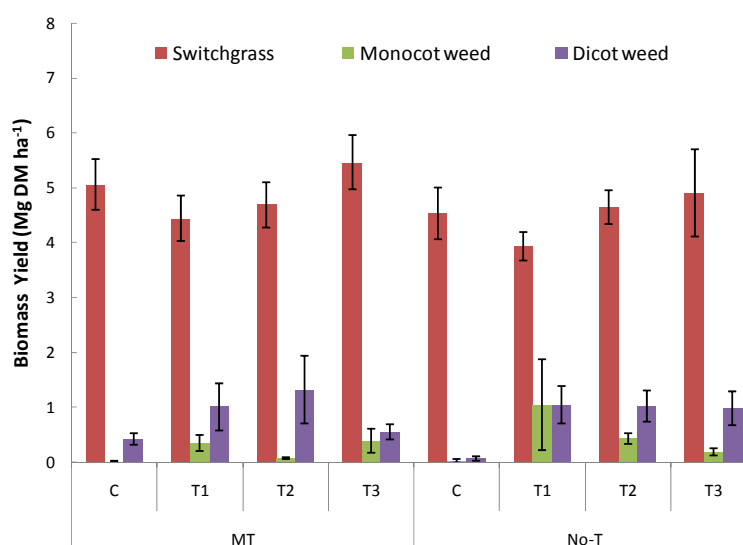
**Table 2 - UNIBO.** Hydro-seeding mixtures used in 2014 trial in Cadriano (Bologna, Italy).

| Hydro-seeding type | Name/Composition*                                                                 | Rate                                       |
|--------------------|-----------------------------------------------------------------------------------|--------------------------------------------|
| T1                 | Envitotal (complete mixture of mulch, fertilizer, adhesive)                       | 45 g m <sup>-2</sup>                       |
| T2                 |                                                                                   | 75 g m <sup>-2</sup>                       |
| T3                 | Cellugrün (cellulose fiber mulch) + soil control (adhesive hydrocolloid compound) | 60 g m <sup>-2</sup> + 1 g m <sup>-2</sup> |

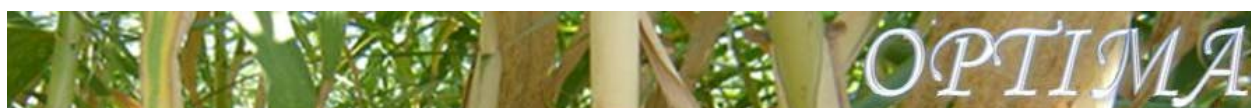
\*All the material for the hydro-seeding trials was supplied by Biasion S.p.A. (Bolzen, Italy) a company specialized in hydro-seeding of ski slopers and degraded lands (e.g., abandoned quarries).

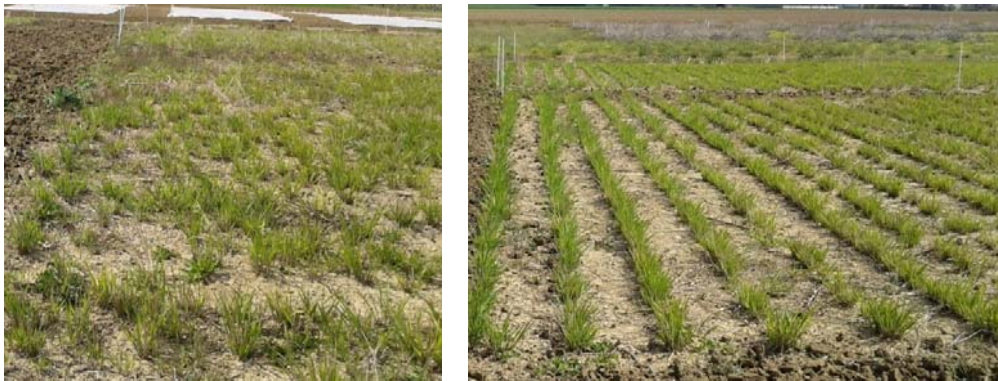


**Figure 2 - UNIBO.** Mixing of switchgrass seeds with the hydro-seeding mulch (left). Equipment used for the hydro-seeding of switchgrass (right).

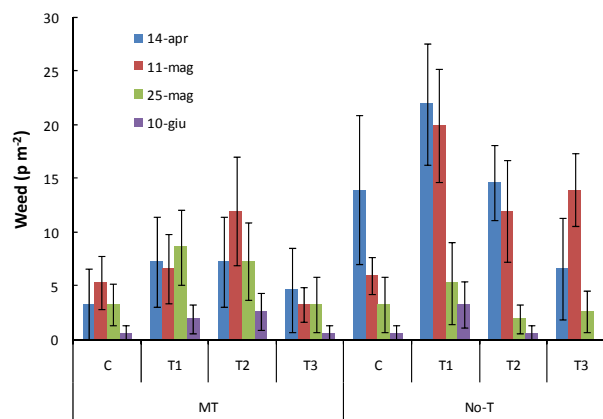


**Figure 3 - UNIBO.** Biomass production (switchgrass, monocot and dicot weeds) in the first year after establishment. Vertical bars: standard deviations.





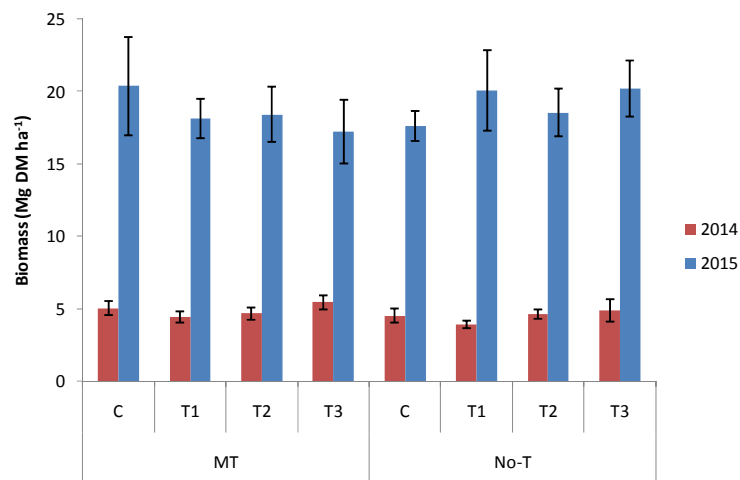
**Figure 4 - UNIBO.** View of the hydro-seeded (left) and conventional seeded (right) plots of switchgrass in the second year (early spring 2015).



**Figure 5 - UNIBO.** Weed presence ( $p\ m^{-2}$ ) in 2015 surveyed at different dates until switchgrass canopy closure. Vertical bars: standard deviations.



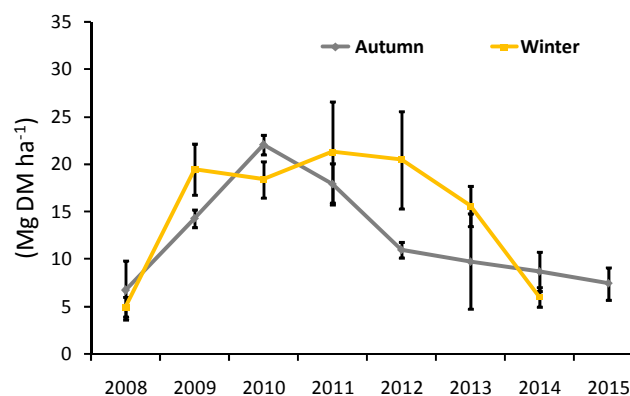
**Figure 6 - UNIBO.** View of the hydro-seeded (left) and conventional (right) seeded plots of switchgrass in the second year in June 2015.



**Figure 7 - UNIBO.** Switchgrass biomass production in the second year after establishment (blue bars) compared to that reached in the first year (red bars). Vertical bars: standard deviations.

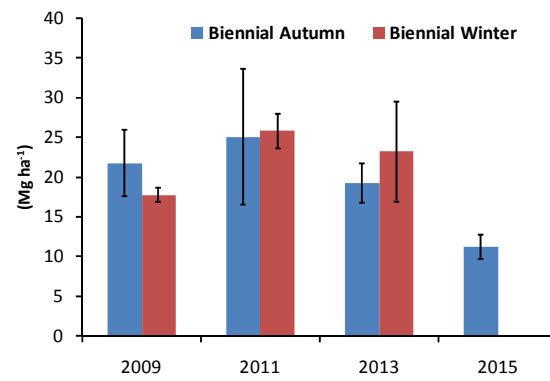


**Figure 8 - UNIBO.** Annual (left) and biennial (right) biomass of switchgrass plots established in 2008 in Cadriano (Bologna).



**Figure 9 - UNIBO.** Effect of harvest time (autumn vs. winter) on switchgrass productivity in Cadriano (Bologna) between 2008 and 2015. Vertical bars: standard deviation.





**Figure 10 - UNIBO.** Effect of harvest time (annual vs. biennial) on switchgrass productivity in Cadriano (Bologna) between 2008 and 2015. Vertical bars: standard deviation.



## Università di Catania – UNICT (n. 1)

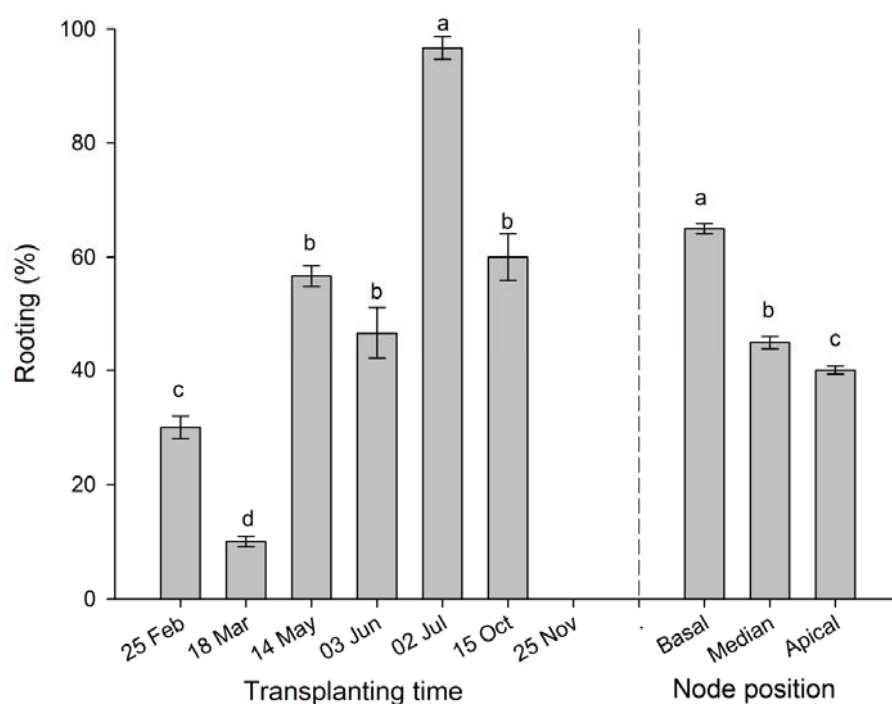


**Figure 1 - UNICT.** Basal, median and apical single node stem cutting of giant reed (*Arundo donax* L.) transplanted in pots.

**Table 1 - UNICT.** Average maximum and minimum air temperatures (°C) during the eight weeks after each transplanting date of giant reed (*Arundo donax* L.) single node stem cuttings at the University of Catania, Italy.

| Transplanting time | Tmax | Tmin |
|--------------------|------|------|
| February 25        | 17.6 | 11.5 |
| March 18           | 19.7 | 12.4 |
| May 14             | 22.6 | 17.3 |
| June 3             | 28.2 | 20.9 |
| July, 2            | 28.0 | 23.8 |
| October 15         | 20.5 | 14.3 |
| November 25        | 15.2 | 9.5  |





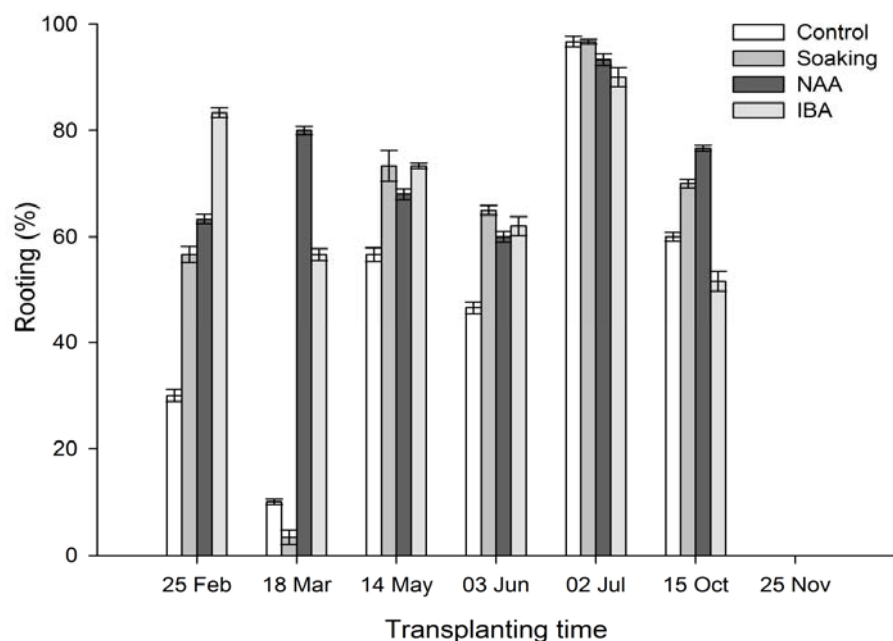
**Figure 2 - UNICT.** Rooting rate of giant reed (*Arundo donax* L.) in relation to the transplanting time and to the node position. Different letters represent statistically different means ( $P \leq 0.05$ ).

**Table 2 - UNICT.** Correlation coefficients ( $r$ ) between air temperatures and rooting rate in relation to node position on stem of giant reed (*Arundo donax* L.) at the University of Catania, Italy.

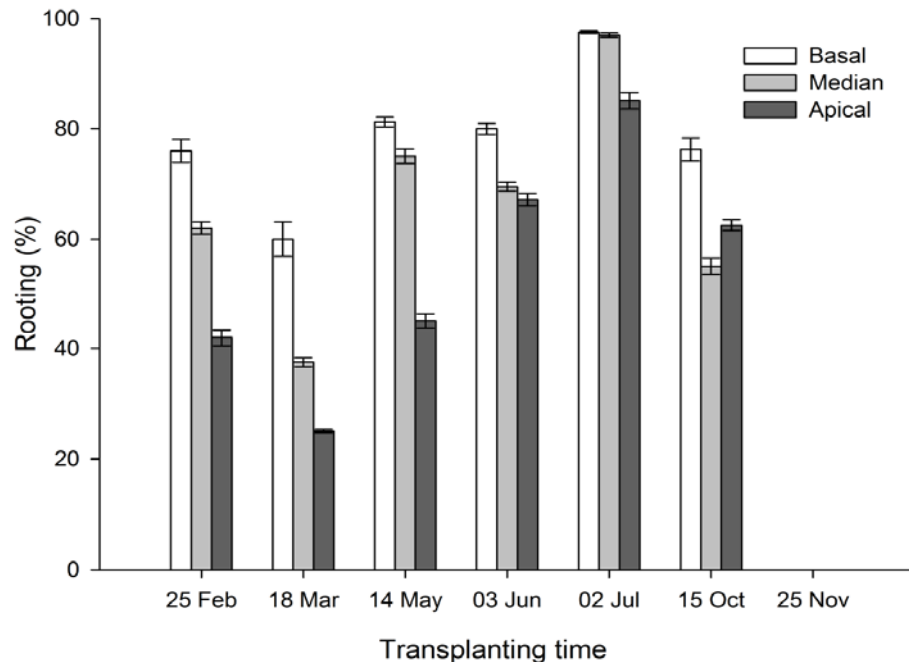
| Node position | Tmax (°C) | Tmin (°C) |
|---------------|-----------|-----------|
| Basal         | 0.70      | 0.75      |
| Median        | 0.74      | 0.84*     |
| Apical        | 0.79*     | 0.87**    |

\*Significant for  $P \leq 0.05$ ; \*\* Significant for  $P \leq 0.01$



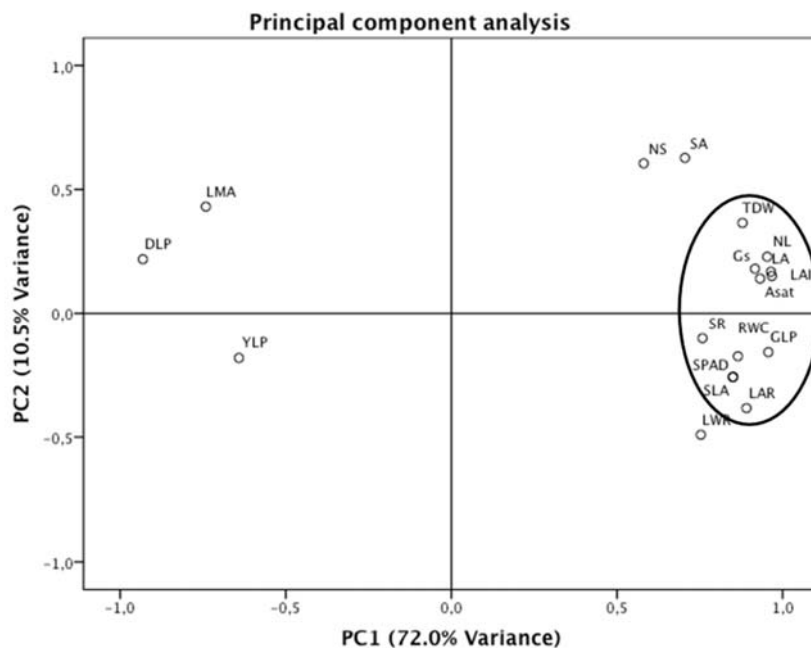


**Figure 3 - UNICT.** Rooting rate of giant reed (*Arundo donax* L.) in relation to the growth regulator pretreatments at different transplanting times, averaged across node position. Fisher's LSD for main effects (transplanting time and pretreatment) and interaction "transplanting time x pretreatment" per  $P \leq 0.05 = 12.69$ .

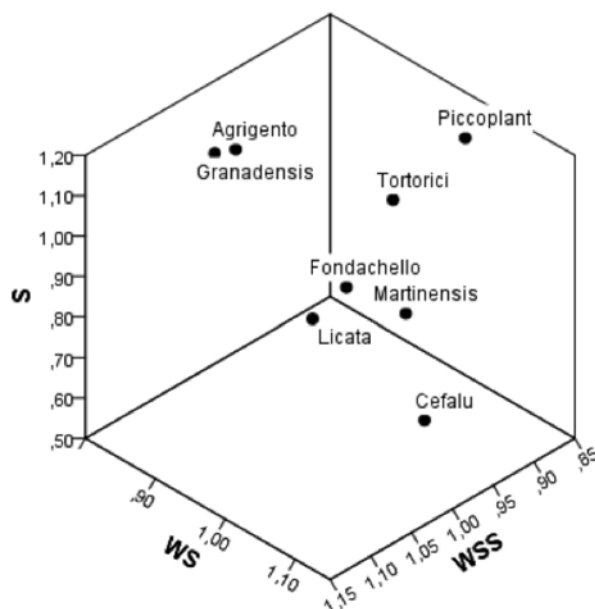


**Figure 4 - UNICT.** Rooting rate of giant reed (*Arundo donax* L.) in relation to node position at different transplanting times, averaged across growth regulator pretreatments. Fisher's LSD for main effects (transplanting time and node position) and interaction "transplanting time x node position" per  $P \leq 0.05 = 10.99$ .



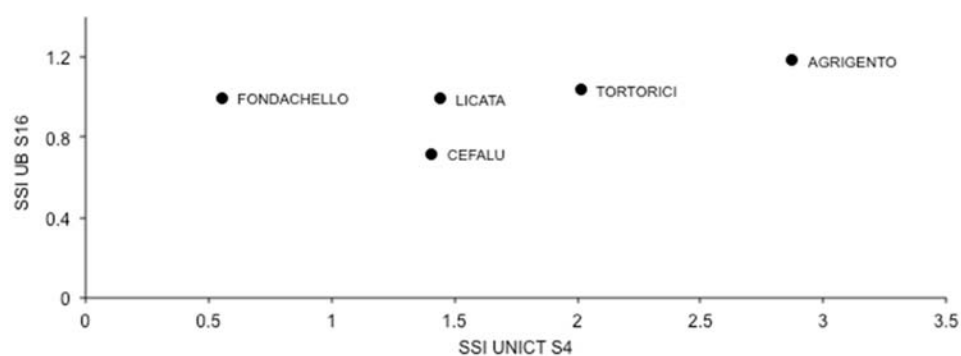


**Figure 5 - UNICT.** Principal component analysis (PCA) and variance (%) of each component. The most significant and positive parameters of component 1 (above 0.750) were selected: Green Leaf Area (gLA), % Green Leaves (GLP), Number of leaves (NL),  $A_{sat}$ , % Dry Leaves (DLP),  $g_s$ , Leaf Area Ratio (LAR), Total Dry Weight (TDW), Relative Water Content (RWC), Specific Leaf Area (SLA), SPAD and Shoot/Root (SR). Non-selected parameters were % Dry Leaves (DLP), Leaf Mass Area (LMA), % Yellow Leaves (YLP), Number of Stems (NS), Stem Area (SA) and Leaf Weight Ratio (LWR).

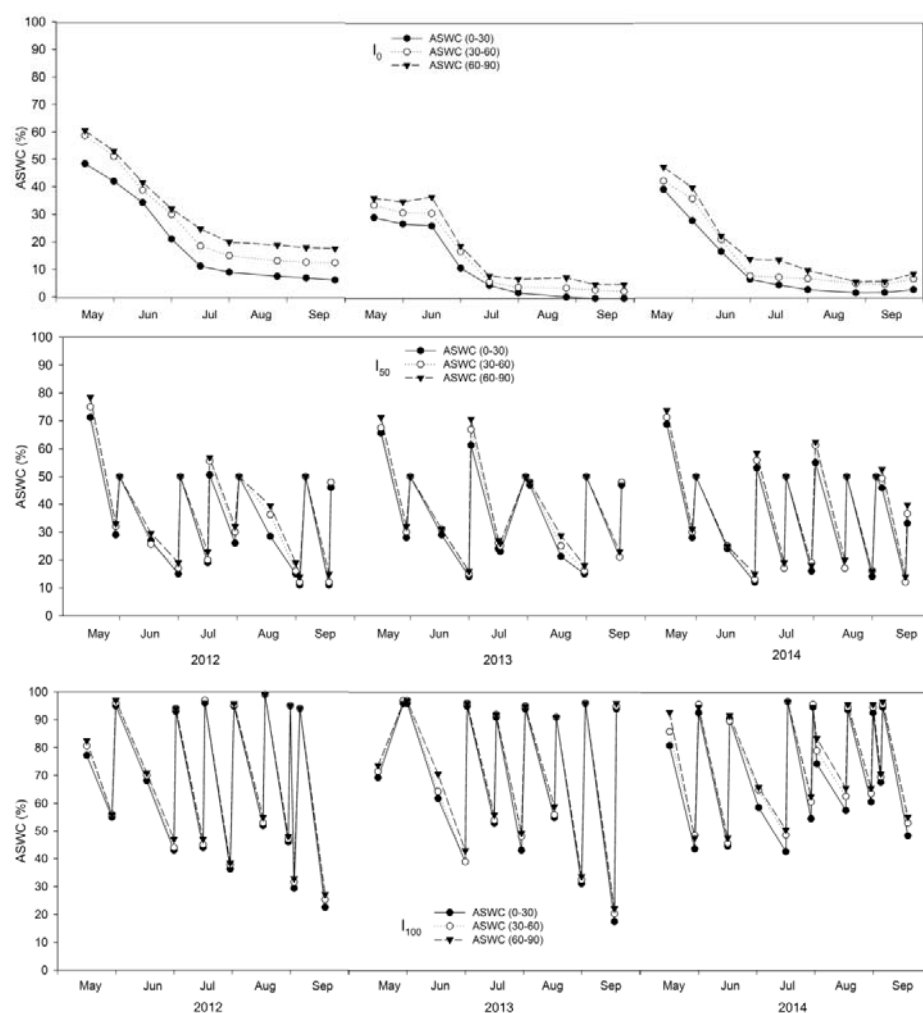


**Figure 6 - UNICT.** 3-D graphic representing stress susceptibility index (SSI) of each clone according to water stress (WS; X axis), salinity (S; Y axis) and double stress (WSS; Z axis). Higher SSI values indicate lower tolerance to stress.





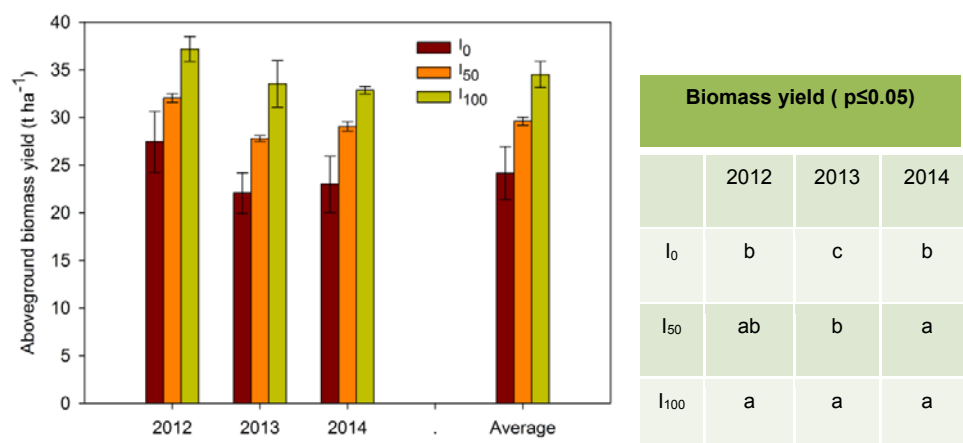
**Figure 7 - UNICT.** 2-D graphic representing stress susceptibility index (SSI) of each clone according to UNICT S4 (X axis) and UB S16 (Y axis). Higher SSI values indicate lower tolerance to stress.



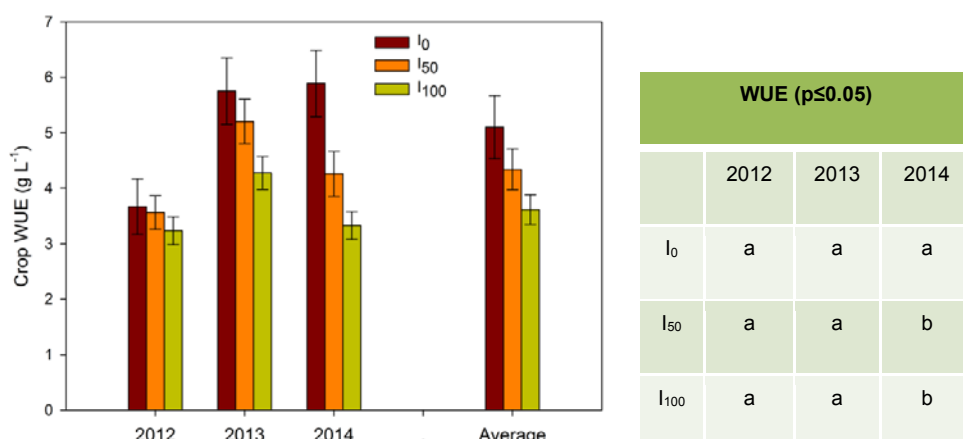
**Figure 8 - UNICT.** Available soil water content (ASWC, %) at three different depths (0–30, 30–60, and 60–90 cm) along the three growing seasons (2011, 2012, and 2013) of *Saccharum spontaneum* L. spp.



*aegyptiacum* (Willd.) Hack. at the Experimental farm of Catania University (10 m a.s.l., 37°25'N lat., 15°03'E long.) in I<sub>0</sub>, I<sub>50</sub>, and I<sub>100</sub> treatment.

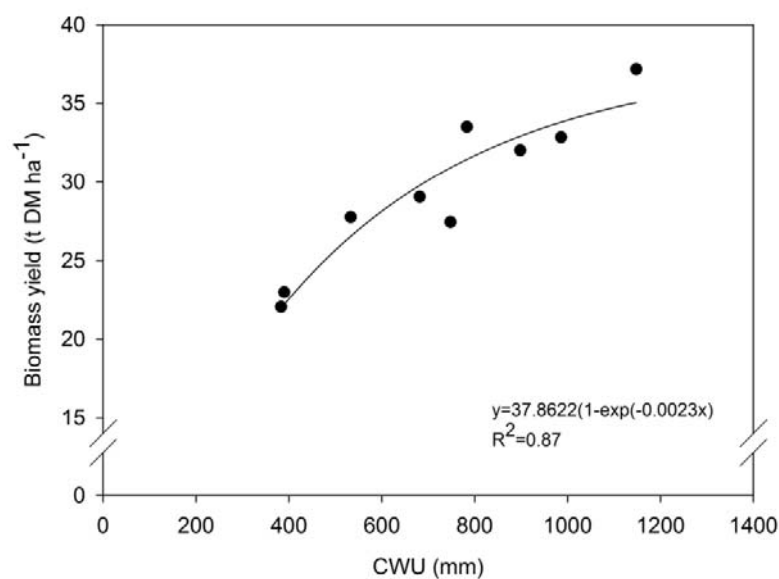


**Figure 9 - UNICT.** Aboveground biomass yield (Mg DM ha<sup>-1</sup>) of *Saccharum spontaneum* L. spp. *aegyptiacum* (Willd.) Hack. in relation to the studied treatment.

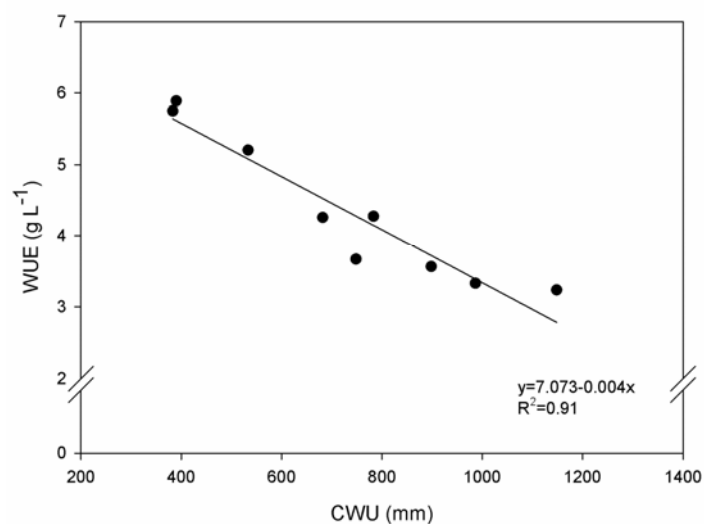


**Figure 10 - UNICT.** Crop water use (g L<sup>-1</sup>) of *Saccharum spontaneum* L. spp. *aegyptiacum* (Willd.) Hack. in relation to the studied treatment.





**Figure 11 - UNICT.** Relation between crop water use (CWU, mm) and aboveground dry biomass yield (Mg ha<sup>-1</sup>) of *Saccharum spontaneum* L. spp. *aegyptiacum* (Willd.) Hack.



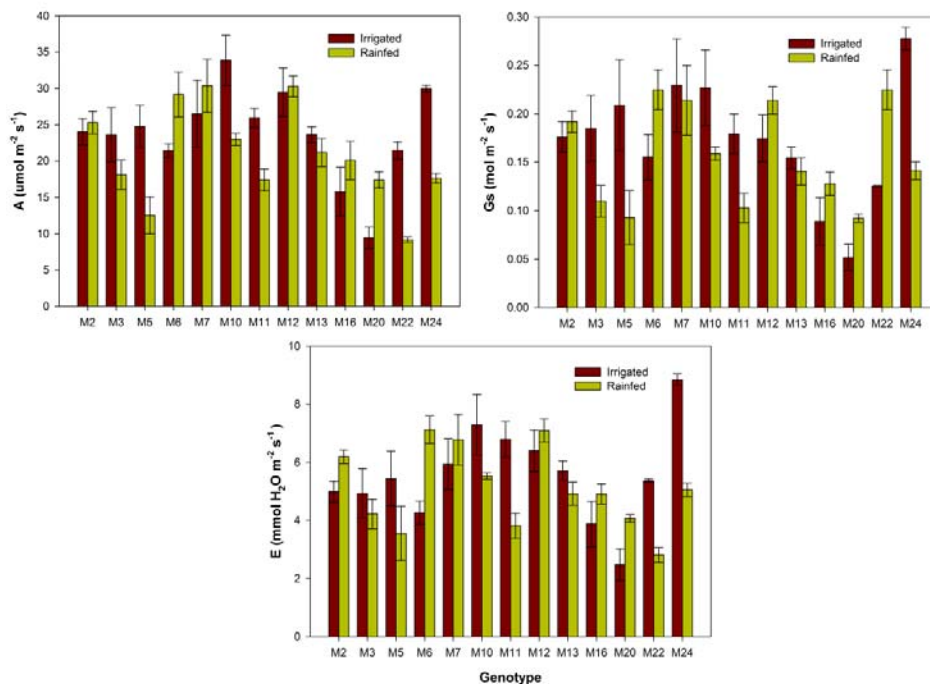
**Figure 12 - UNICT.** Relations between crop water use (CWU, mm) and water use efficiency (WUE, g L<sup>-1</sup>) of *Saccharum spontaneum* L. spp. *aegyptiacum* (Willd.) Hack.



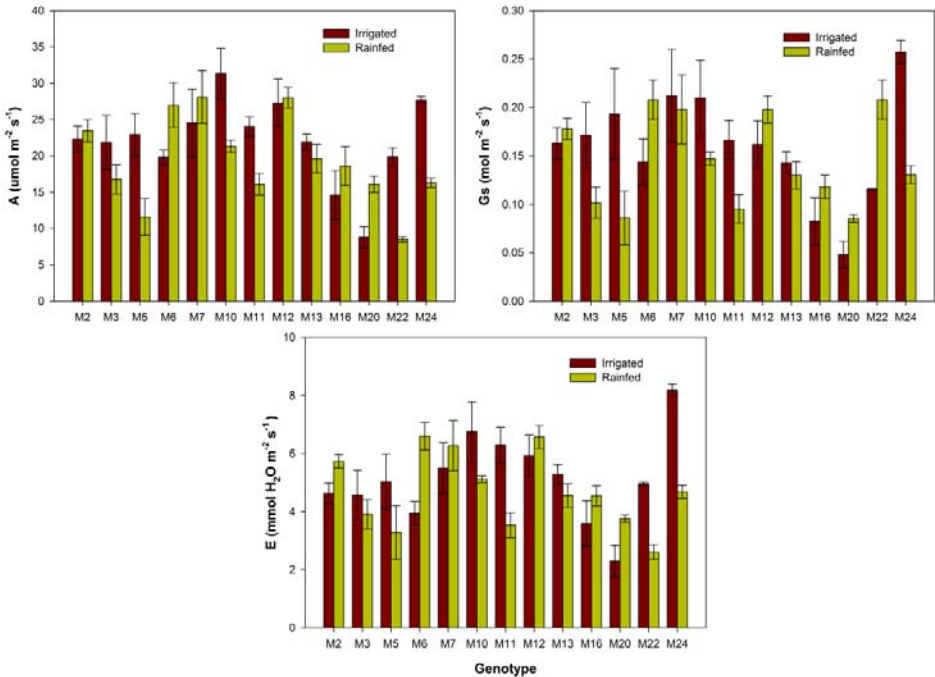
|                     |                     |                     |                     |                     |                     |                     |                     |                     |                     |
|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| Mb 1093#44<br>26279 | Mb 961#28<br>22618  | Mb 311 GIG          | Mb 1123#25<br>8564  | Mb 933#24<br>23009  | Mb 1023#21<br>22661 | Mb 99 GOL           | Mb 969#25<br>22645  | Mb 1131#25<br>8634  | Mb 967#24<br>23059  |
| Mb 1122#25<br>8584  | Mb 931#23<br>22998  | Mb 1168#42<br>26157 | Mb 914#22<br>22937  | Mb 969#25<br>22645  | Mb 933#24<br>23009  | Mb 1122#26<br>8585  | Mb 910#25<br>22930  | Mb 1122#25<br>8584  | Mb 1168#42<br>26157 |
| Mb 910#28<br>22933  | Mb 1025#22<br>22672 | Mb 967#24<br>23059  | Mb 1131#25<br>8634  | Mb 1023#23<br>22663 | Mb 931#28<br>23003  | Mb 925#22<br>22967  | Mb 961#28<br>22618  | Mb 1123#25<br>8564  | Mb 836#23<br>22758  |
| Mb 99 GOL           | Mb 925#22<br>22967  | Mb 1131#24<br>8633  | Mb 931#21<br>22996  | Mb 1023#21<br>22661 | Mb 914#22<br>22937  | Mb 931#23<br>22998  | Mb 1023#23<br>22663 | Mb 893#21<br>22876  | Mb 910#28<br>22933  |
| Mb 910#25<br>22930  | Mb 836#23<br>22758  | Mb 1122#26<br>8585  | Mb 893#21<br>22876  | Mb 931#28<br>23003  | Mb 931#21<br>22996  | Mb 1093#44<br>26279 | Mb 311 GIG          | Mb 1025#22<br>22672 | Mb 1131#24<br>8633  |
| Mb 1122#25<br>8584  | Mb 969#25<br>22645  | Mb 910#28<br>22933  | Mb 1023#23<br>22663 | Mb 967#24<br>23059  | Mb 933#24<br>23009  | Mb 1168#42<br>26157 | Mb 969#25<br>22645  | Mb 931#21<br>22996  | Mb 1131#24<br>8633  |
| Mb 914#22<br>22937  | Mb 925#22<br>22967  | Mb 1168#42<br>26157 | Mb 1122#26<br>8585  | Mb 931#23<br>22998  | Mb 893#21<br>22876  | Mb 1093#44<br>26279 | Mb 910#25<br>22930  | Mb 1025#22<br>22672 | Mb 1131#25<br>8564  |
| Mb 933#24<br>23009  | Mb 931#23<br>22998  | Mb 1131#24<br>8633  | Mb 931#28<br>23003  | Mb 931#21<br>22996  | Mb 910#28<br>22933  | Mb 311 GIG          | Mb 931#28<br>23003  | Mb 99 GOL           | Mb 1123#25<br>8564  |
| Mb 1025#22<br>22672 | Mb 910#25<br>22930  | Mb 1131#25<br>8634  | Mb 99 GOL           | Mb 1093#44<br>26279 | Mb 967#24<br>23059  | Mb 1023#21<br>22661 | Mb 961#28<br>22618  | Mb 914#22<br>22937  | Mb 925#22<br>22967  |
| Mb 961#28<br>22618  | Mb 1023#21<br>22661 | Mb 836#23<br>22758  | Mb 1123#25<br>8564  | Mb 893#21<br>22876  | Mb 1023#23<br>22663 | Mb 1131#25<br>8634  | Mb 1122#26<br>8585  | Mb 1122#25<br>8584  | Mb 836#23<br>22758  |

|                     |                     |                     |                     |                     |                     |                     |                    |                     |                     |
|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|--------------------|---------------------|---------------------|
| Mb 893#21<br>22876  | Mb 1168#42<br>26157 | Mb 836#23<br>22758  | Mb 1122#25<br>8584  | Mb 910#25<br>22930  | Mb 925#22<br>22967  | Mb 1123#25<br>8564  | Mb 931#28<br>23003 | Mb 967#24<br>23059  | Mb 931#23<br>22998  |
| Mb 925#22<br>22967  | Mb 1023#23<br>22663 | Mb 931#21<br>22996  | Mb 931#23<br>22998  | Mb 961#28<br>22618  | Mb 1131#25<br>8634  | Mb 931#21<br>22996  | Mb 1122#25<br>8584 | Mb 933#24<br>23009  | Mb 1168#42<br>26157 |
| Mb 931#28<br>23003  | Mb 311 GIG          | Mb 1131#24<br>8633  | Mb 1025#22<br>22672 | Mb 1131#25<br>8634  | Mb 1023#21<br>22661 | Mb 1023#23<br>22663 | Mb 99 GOL          | Mb 311 GIG          | Mb 893#21<br>22876  |
| Mb 1123#25<br>8564  | Mb 910#28<br>22933  | Mb 967#24<br>23059  | Mb 969#25<br>22645  | Mb 1023#21<br>22661 | Mb 1093#44<br>26279 | Mb 1122#26<br>8585  | Mb 910#25<br>22930 | Mb 1025#22<br>22672 | Mb 961#28<br>22618  |
| Mb 914#22<br>22937  | Mb 933#24<br>23009  | Mb 1093#44<br>26279 | Mb 1122#26<br>8585  | Mb 910#25<br>22930  | Mb 931#28<br>23003  | Mb 1131#24<br>8633  | Mb 910#28<br>22933 | Mb 836#23<br>22758  | Mb 1023#21<br>22661 |
| Mb 1025#22<br>22672 | Mb 1122#26<br>8585  | Mb 1131#24<br>8633  | Mb 1093#44<br>26279 | Mb 910#25<br>22930  | Mb 931#28<br>23003  | Mb 1131#24<br>8633  | Mb 910#28<br>22933 | Mb 836#23<br>22758  | Mb 1023#21<br>22661 |
| Mb 961#28<br>22618  | Mb 1123#25<br>8564  | Mb 311 GIG          | Mb 931#23<br>22998  | Mb 969#25<br>22645  | Mb 910#25<br>22930  | Mb 961#28<br>22618  | Mb 969#25<br>22645 | Mb 1025#22<br>22672 | Mb 933#24<br>23009  |
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| Mb 914#22<br>22937  | Mb 925#22<br>22967  | Mb 99 GOL           | Mb 933#24<br>23009  | Mb 931#21<br>22996  | Mb 1023#23<br>22663 | Mb 925#22<br>22967  | Mb 311 GIG         | Mb 931#21<br>22996  | Mb 1122#25<br>8584  |
| Mb 1023#21<br>22661 | Mb 931#28<br>23003  | Mb 836#23<br>22758  | Mb 910#28<br>22933  | Mb 1023#23<br>22663 | Mb 1122#26<br>8585  | Mb 1168#42<br>26157 | Mb 1123#25<br>8564 | Mb 1093#44<br>26279 | Mb 931#23<br>22998  |

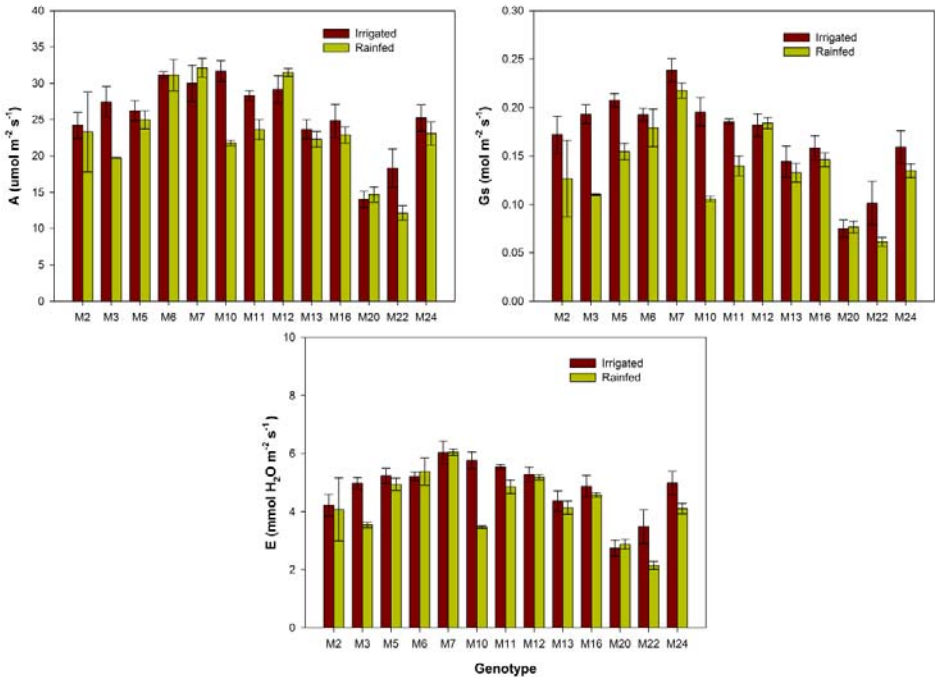
**Figure 13 – UNICT.** Experimental design overview showing the two blocks (rainfed and well watered) and the completely randomized 25 *Miscanthus* accessions replicated four times.



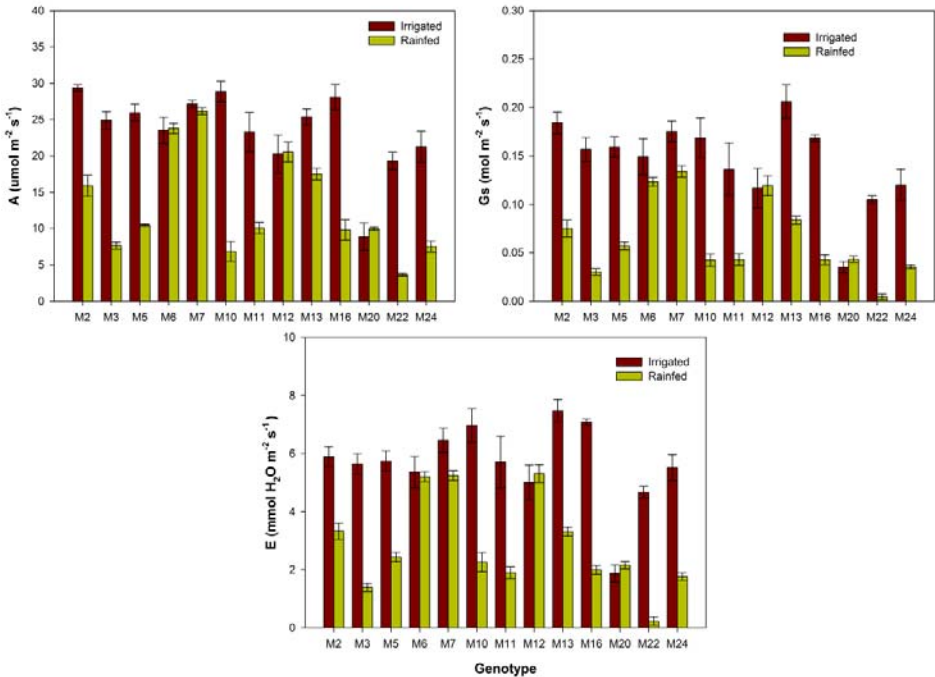
**Figure 14 – UNICT.** Gas exchange in June 20 of 25 Miscanthus accessions grown in drought stress (rainfed) and well watered conditions.



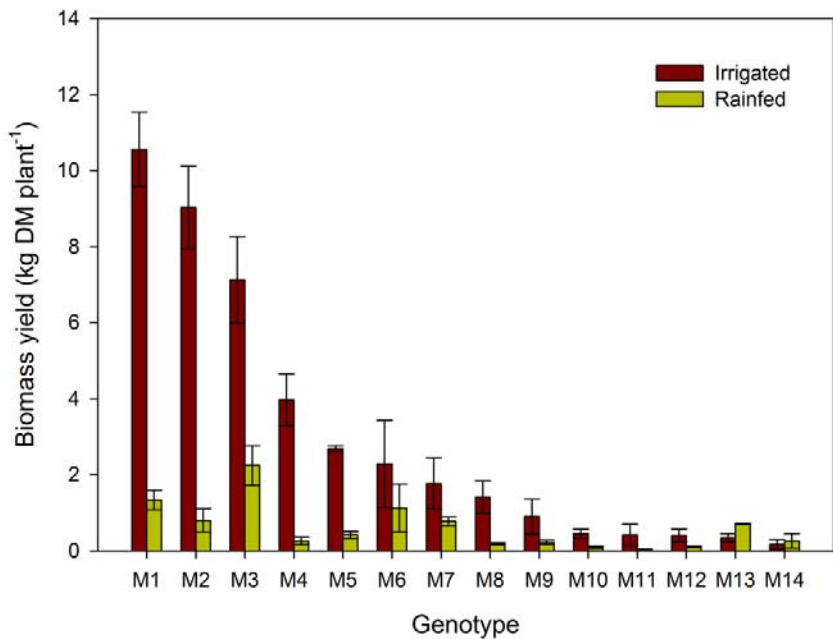
**Figure 15– UNICT.** Gas exchange in July 20 of 25 Miscanthus accessions grown in drought stress (rainfed) and well watered conditions.



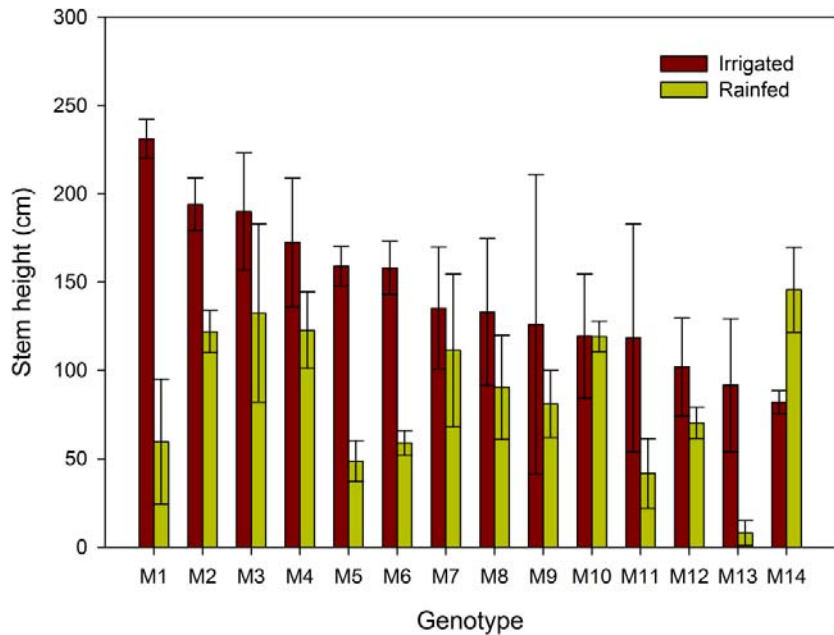
**Figure 16 – UNICT.** Gas exchange in August of 25 Miscanthus accessions grown in drought stress (rainfed) and well watered conditions.



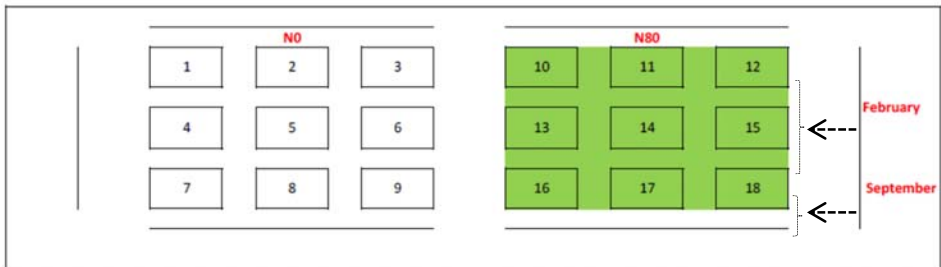
**Figure 17 - UNICT.** Gas exchange in September 10 of 25 Miscanthus accessions grown in drought stress (rainfed) and well watered conditions.



**Figure 18 - UNICT.** Biomass yield of Miscanthus accessions grown in drought stress (rainfed) and well watered conditions at UNICT.



**Figure 19 – UNICT.** Stem height of Miscanthus accessions grown in drought stress (rainfed) and well watered conditions at UNICT.



**Figure 20 - UNICT.** Experimental layout of the long term plantation at Experimental farm of UNICT (Catania plain, 10 m a.s.l., 37°27' N, 15° 03' E).



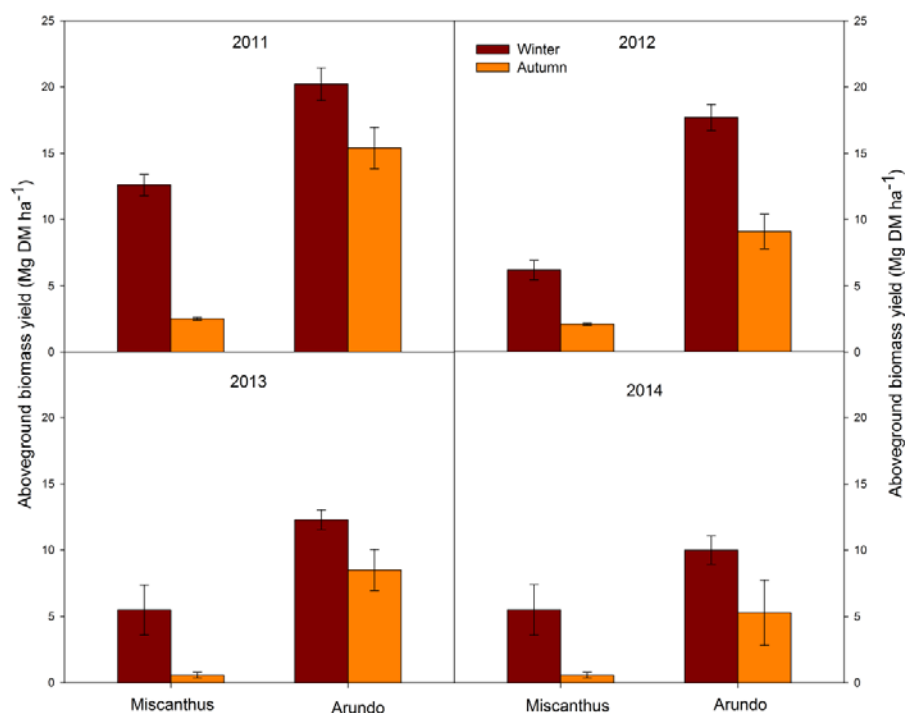


**Figure 21 - UNICT.** February 2014 harvest of long-term giant reed (up-left), *Miscanthus x giganteus* (up-right) and September 2014 harvest of giant reed (down-left) and *Miscanthus x giganteus* (down-right).

**Table 3 - UNICT.** ANOVA for biomass yield and moisture content of *Arundo donax* and *Miscanthus x giganteus* in relation to harvest time (autumn and winter). LSD test was carried out for mean separations.

|                   | Yield (p-value) | Moisture (p-value) |
|-------------------|-----------------|--------------------|
| <b>2011</b>       |                 |                    |
| Species           | ***             | ***                |
| Harvest           | ***             | ***                |
| Species x Harvest | *               | ***                |
| <b>2012</b>       |                 |                    |
| Species           | ***             | ***                |
| Harvest           | ***             | ***                |
| Species x Harvest | **              | ***                |
| <b>2013</b>       |                 |                    |
| Species           | ***             | ***                |
| Harvest           | ***             | ***                |
| Species x Harvest | ns              | ***                |
| <b>2014</b>       |                 |                    |
| Species           | **              | ***                |
| Harvest           | **              | ***                |
| Species x Harvest | ns              | ***                |



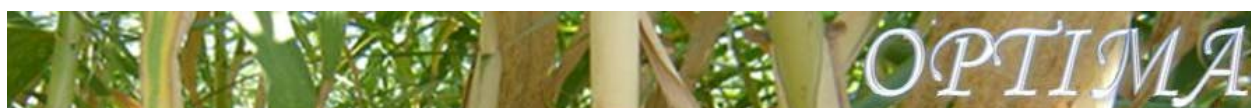


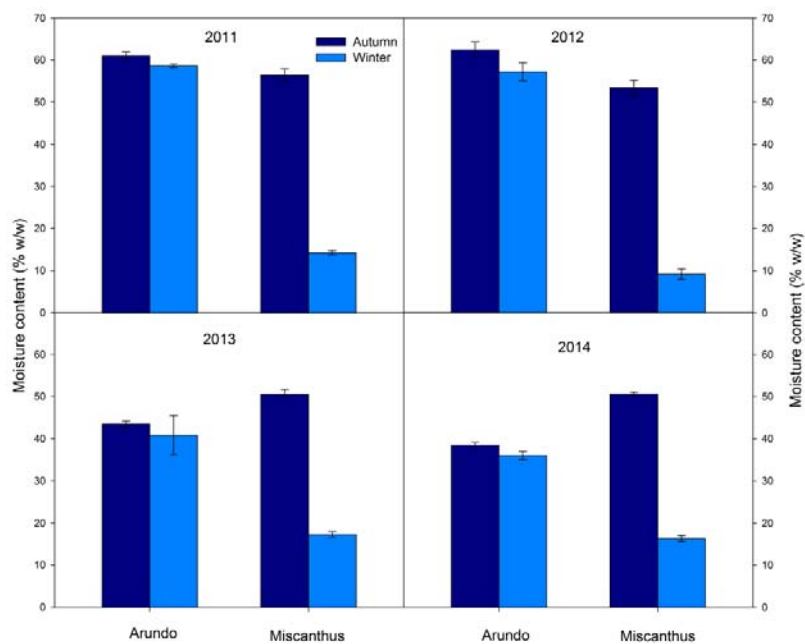
**Figure 22 - UNICT.** Aboveground biomass yield (Mg DM ha<sup>-1</sup>) of giant reed and miscanthus harvested in autumn and winter, in 2011, 2012, 2013 and 2014.

**Table 4 - UNICT.** ANOVA for biomass quality of *Arundo donax* and *Miscanthus x giganteus* in relation to harvest time (autumn and winter) and year (2011, 2012 and 2013 growing seasons) in Catania trial. LSD test was carried out for mean separations.

| Statistics (p values)         | NDS | NDF | ADF | ADL | ASH |
|-------------------------------|-----|-----|-----|-----|-----|
| Harvest time                  | *** | *** | *** | ns  | *** |
| Species                       | *** | *** | *   | *** | *** |
| Year                          | *   | *** | *** | *** | *** |
| Harvest time × Species        | *** | *** | *** | ns  | ns  |
| Harvest time × Year           | *** | ns  | *   | ns  | ns  |
| Species × Year                | ns  | *** | *** | **  | ns  |
| Harvest time × Species × Year | **  | *** | *** | *** | ns  |

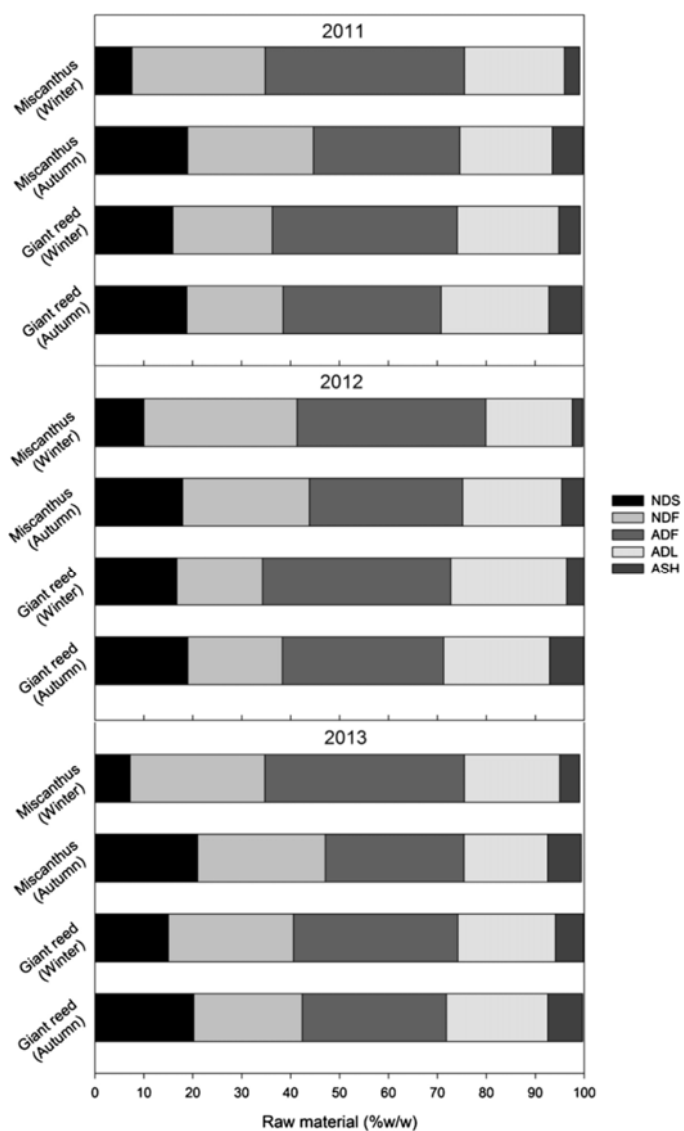
\*significant per  $p \leq 0.05$ ; \*\* significant per  $p \leq 0.01$ ; \*\*\* significant per  $p \leq 0.001$ ; ns (non-significant)





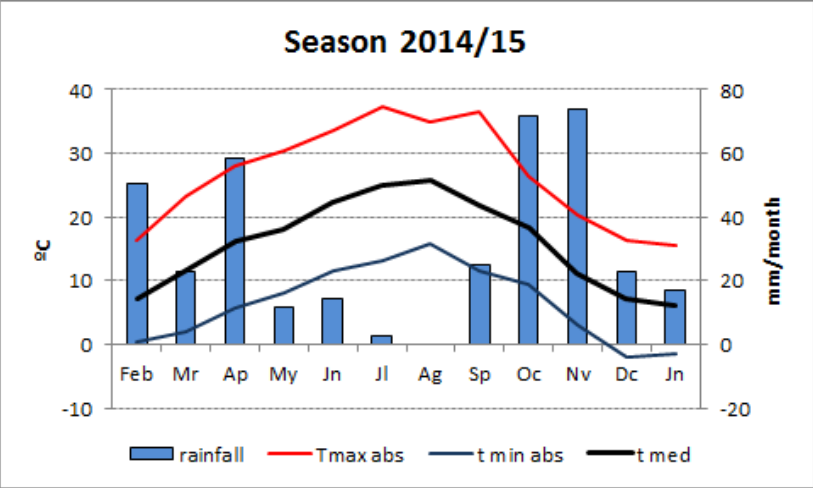
**Figure 23 - UNICT.** Moisture content (% w/w) of giant reed and miscanthus harvested in autumn and winter, in 2011, 2012, 2013 and 2014.



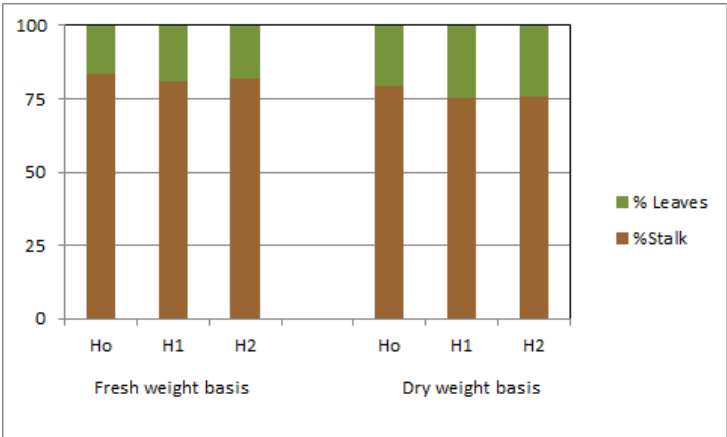


**Figure 24 - UNICT.** Qualitative traits of giant reed and miscanthus on whole biomass (raw material) harvested in autumn and winter, in 2011, 2012 and 2013. NDS: soluble compounds, NDF: hemicellulose, ADF: cellulose, ADL: lignin, ASH: ashes.

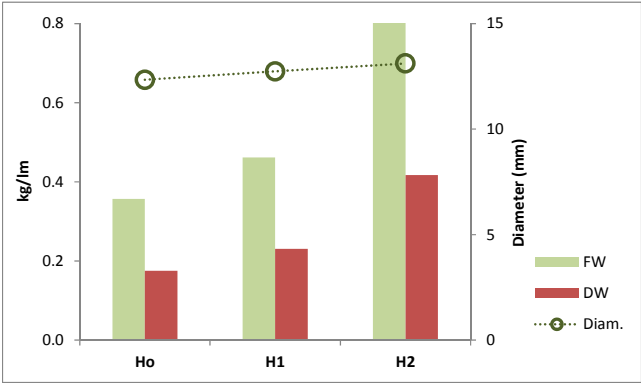




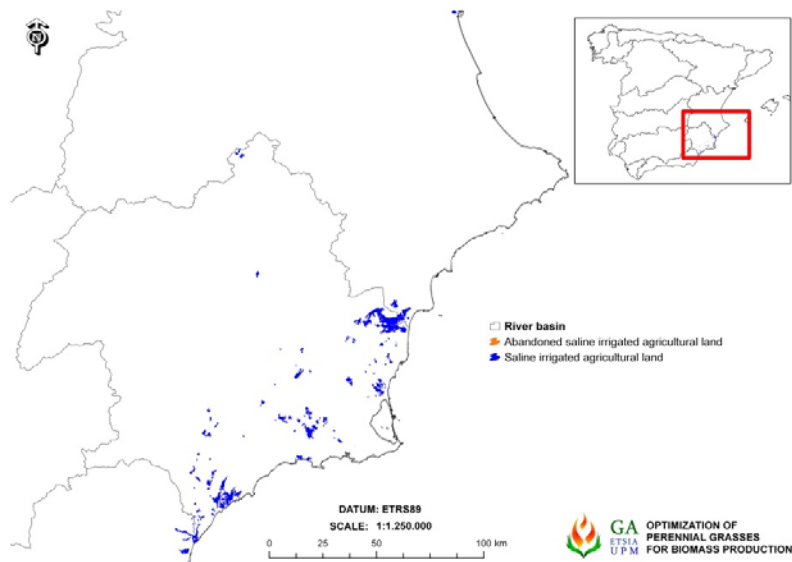
**Figure 1 - UPM.** Records of temperature and rainfall in the season 2014/15. Data from AEMET, Meteorological Station of Madrid-Retiro, Longitude 34041 W, Latitude 402443, Altitude 667 m.a.s.l.



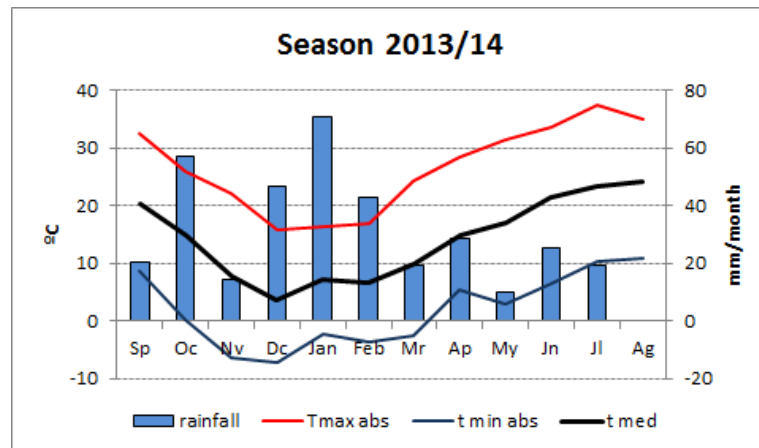
**Figure 2 - UPM.** Mean biomass partitioning of Arundo biomass under limiting water and soil conditions, 2015' harvest.



**Figure 3 - UPM.** Mean biomass production and mean cane diameter versus hydric regime, in the 2014' harvest of a crop of Arundo grown under limiting water and soil conditions. H0=dry farming; H1= target water regime of 450 mm; H2= target water regime of 650 mm.

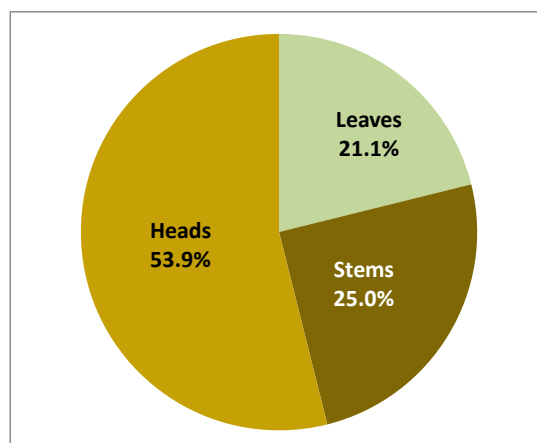


**Figure 4 - UPM.** Saline and prone-saline agricultural areas suitable for growing giant reed.

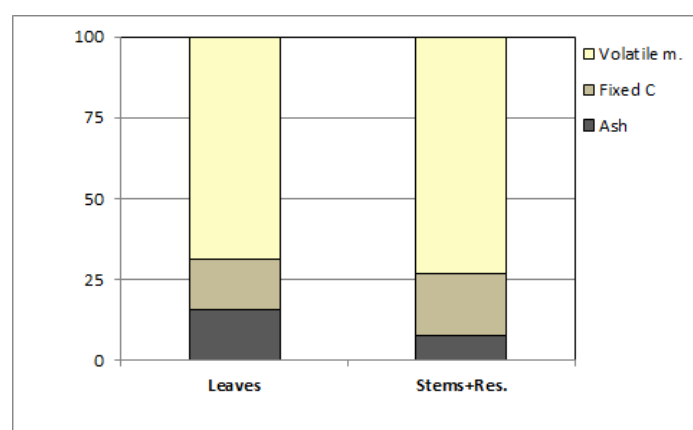


**Figure 5 - UPM.** Temperature and rainfall in 2013/2014 season. Data from the Meteorological Station of San Fernando de Henares. UTM X: 457867, UTM Y: 4473610; Huse 30; altitude 604 m.a.s.l.

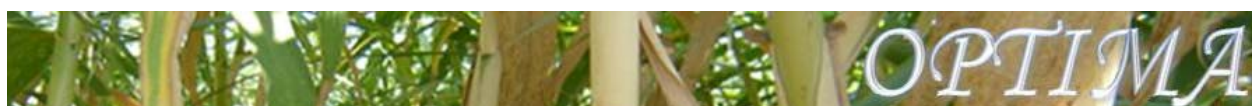


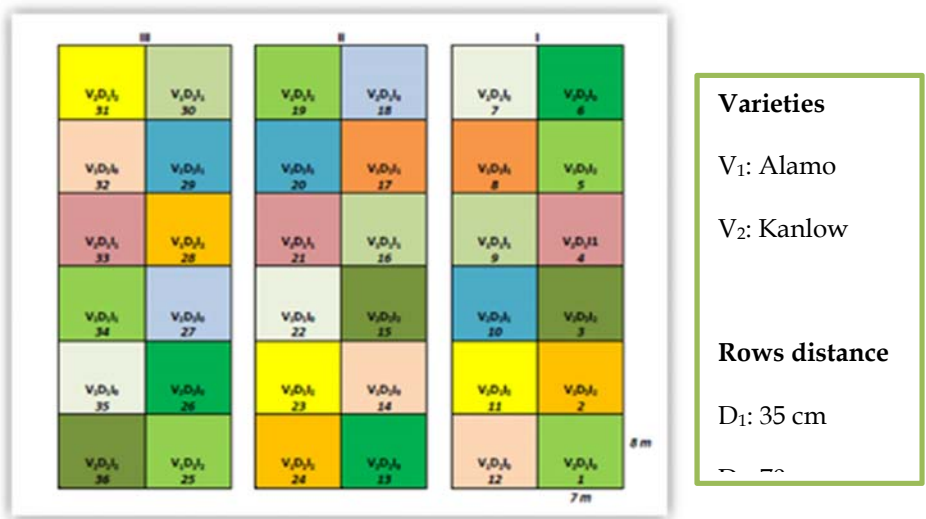


**Figure 6 - UPM.** Mean biomass partitioning (w/w). Cynara grown at demonstrative scale. Daganzo (Spain), 2014 harvest. Annual rainfall (San Fernando de Henares, 2013/2014): 356 mm.



**Figure 7 - UPM.** Proximate analysis of cynara biomass as harvested (no biomass pre-treatments, no washing). Cynara crop grown at demonstrative scale. Daganzo (Spain), 2014 harvest. Annual rainfall (San Fernando de Henares, 2013-14): 356 mm.





**Figure 1 – CRES.** View of the experimental layout of switchgrass trial in central Greece (tested factors: variety, density and irrigation rates).



**Figure 2 – CRES.** View of the switchgrass trial at the establishment (25/6/12); D<sub>1</sub>: 35 cm between the rows on the left and D<sub>2</sub>: 70 cm on the right.



**Figure 3 – CRES.** View of the switchgrass trial with lodging problems (in September on the left and in November 2012 on the right).

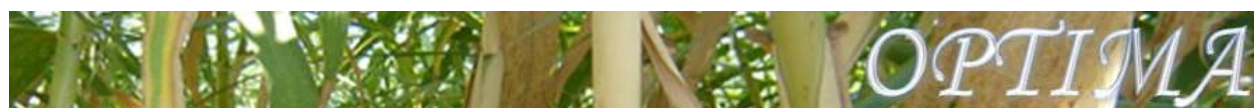


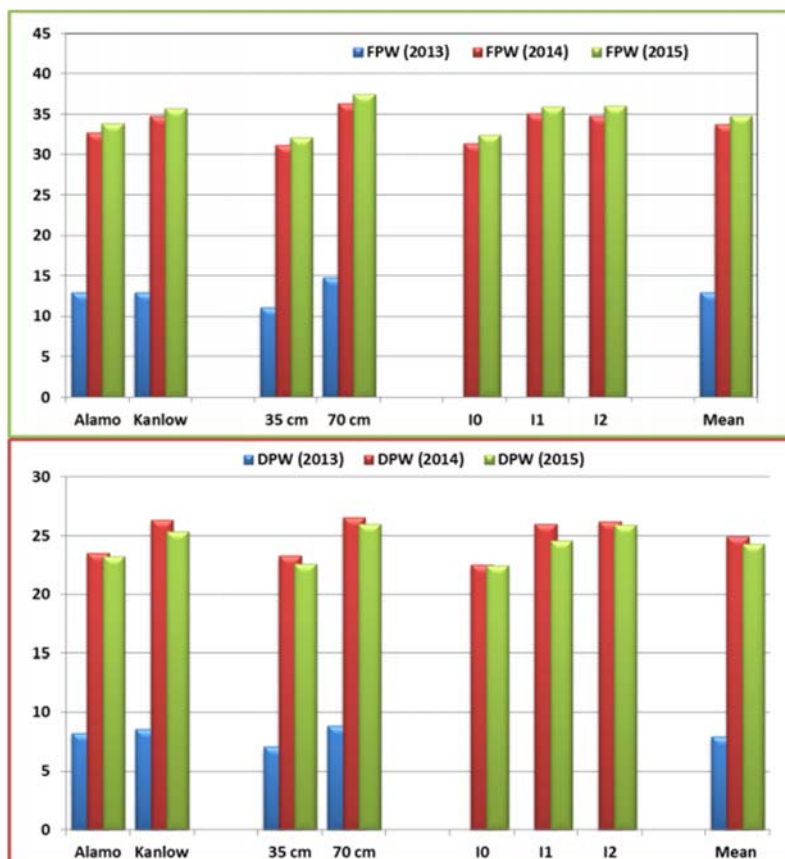
**Table 1 – CRES.** Growth characteristics (plant height, number of tillers/m<sup>2</sup> and tiller density) for three subsequent growing periods.

| Years                             | Plant height (cm) |      |      | Number of tillers/m <sup>2</sup> |      |      | Tiller diameter (mm) |      |      |
|-----------------------------------|-------------------|------|------|----------------------------------|------|------|----------------------|------|------|
|                                   | 2013              | 2014 | 2015 | 2013                             | 2014 | 2015 | 2013                 | 2014 | 2015 |
| <b>Varieties</b>                  |                   |      |      |                                  |      |      |                      |      |      |
| <b>Alamo</b>                      | 163               | 226  | 208  | 384                              | 462  | 471  | 3.55                 | 4.41 | 4.48 |
| <b>Kanlow</b>                     | 156               | 227  | 198  | 467                              | 613  | 603  | 3.37                 | 4.18 | 4.22 |
| <b>Distances between the rows</b> |                   |      |      |                                  |      |      |                      |      |      |
| <b>D1: 35 cm</b>                  | 144               | 221  | 196  | 404                              | 555  | 546  | 3.06                 | 3.97 | 4.06 |
| <b>D1: 70 cm</b>                  | 157               | 231  | 210  | 406                              | 510  | 528  | 3.51                 | 4.62 | 4.56 |
| <b>Irrigation rates</b>           |                   |      |      |                                  |      |      |                      |      |      |
| <b>I0</b>                         | -                 | 224  | 201  | -                                | 491  | 465  | -                    | 4.02 | 4.37 |
| <b>I1</b>                         | -                 | 226  | 202  | -                                | 580  | 552  | -                    | 4.48 | 4.10 |
| <b>I2</b>                         | -                 | 228  | 206  | -                                | 527  | 597  | -                    | 4.38 | 4.46 |
| <b>Mean</b>                       | 151               | 226  | 203  | 405                              | 533  | 537  | 3.29                 | 4.29 | 4.31 |

**Table 2 – CRES.** Yields results (fresh and dry matter yields, % stems on dry basis) from three subsequent growing seasons.

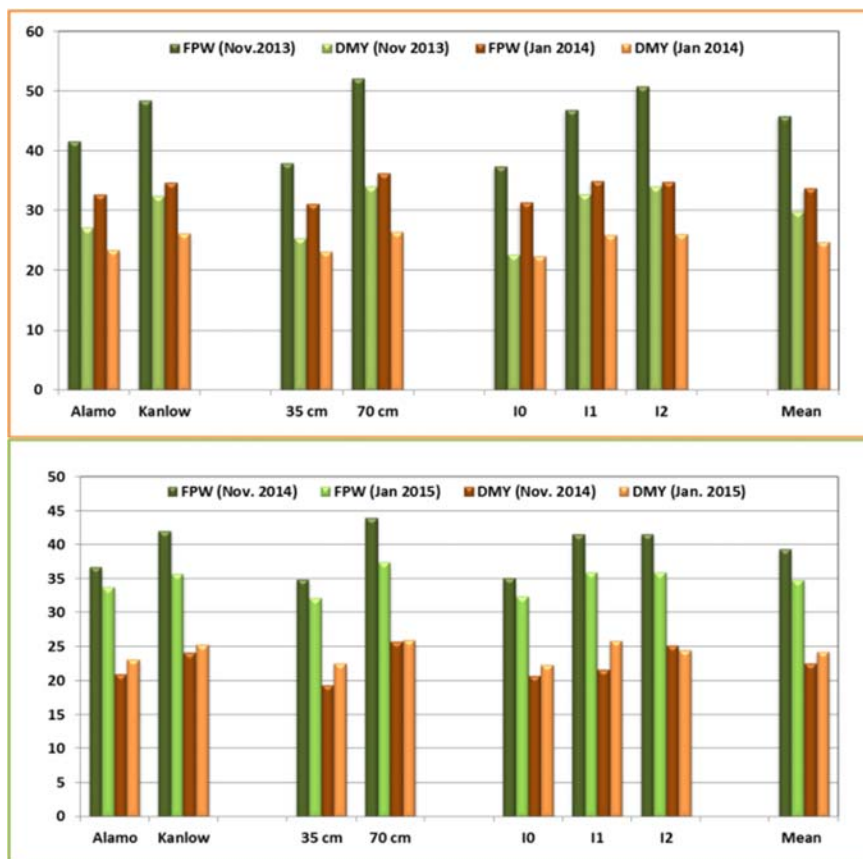
| Years                             | Fresh biomass yields (t/ha) |              |              | Dry matter yields (t/ha) |              |              | % stems on the biomass (oven-dried) |              |              |
|-----------------------------------|-----------------------------|--------------|--------------|--------------------------|--------------|--------------|-------------------------------------|--------------|--------------|
|                                   | 2013                        | 2014         | 2015         | 2013                     | 2014         | 2015         | 2013                                | 2014         | 2015         |
| <b>Varieties</b>                  |                             |              |              |                          |              |              |                                     |              |              |
| <b>Alamo</b>                      | 12.01                       | 32.74        | 33.83        | 7.62                     | 23.52        | 23.23        | 60.63                               | 67.89        | 65.69        |
| <b>Kanlow</b>                     | 12.92                       | 34.79        | 35.75        | 8.57                     | 26.31        | 25.35        | 61.37                               | 66.63        | 64.73        |
| <b>Distances between the rows</b> |                             |              |              |                          |              |              |                                     |              |              |
| <b>D1: 35 cm</b>                  | 11.10                       | 31.18        | 32.14        | 7.07                     | 23.26        | 22.85        | 60.82                               | 64.53        | 63.64        |
| <b>D1: 70 cm</b>                  | 13.83                       | 36.34        | 37.43        | 8.18                     | 26.57        | 27.49        | 61.25                               | 67.78        | 66.54        |
| <b>Irrigation rates</b>           |                             |              |              |                          |              |              |                                     |              |              |
| <b>I0</b>                         | -                           | 31.41        | 32.42        | -                        | 22.53        | 22.43        | -                                   | 63.83        | 65.27        |
| <b>I1</b>                         | -                           | 35.06        | 35.96        | -                        | 26.01        | 24.55        | -                                   | 67.62        | 64.85        |
| <b>I2</b>                         | -                           | 34.81        | 35.98        | -                        | 26.22        | 25.88        | -                                   | 67.39        | 65.46        |
| <b>Mean</b>                       | <b>12.47</b>                | <b>33.76</b> | <b>34.79</b> | <b>7.63</b>              | <b>24.92</b> | <b>24.29</b> | <b>61.07</b>                        | <b>66.41</b> | <b>64.80</b> |





**Figure 4 – CRES.** Comparison in terms of fresh and dry matter yields (t/ha) among the three growing periods (2012-3, 2013-4 & 2014-5) for the tested factors (varieties, row distances and irrigation rates)





**Figure 5 – CRES.** Comparison in term of fresh and dry matter yields (t/ha) between the autumn and winter harvest in the second and the third growing period.

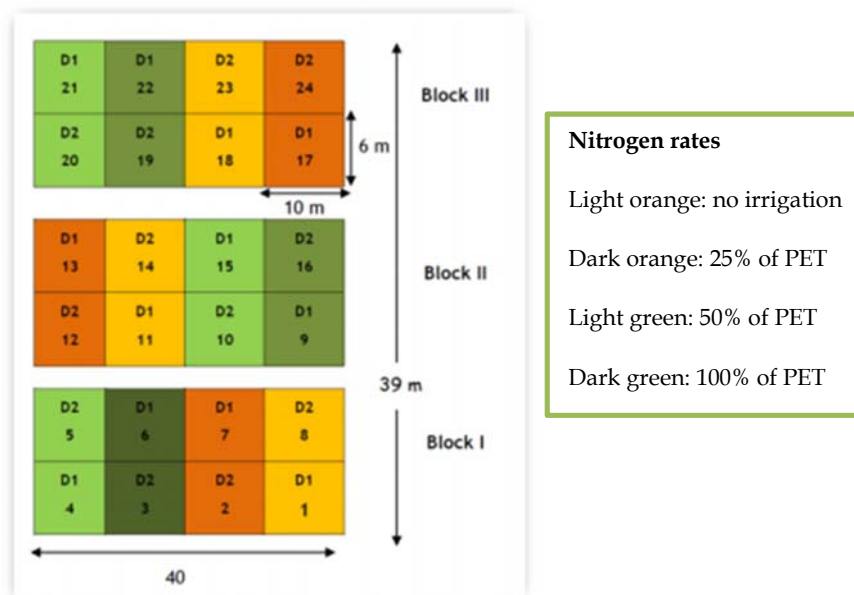


**Figure 6 – CRES.** View of the switchgrass (at the front) and giant reed (at the back) in the beginning of November.

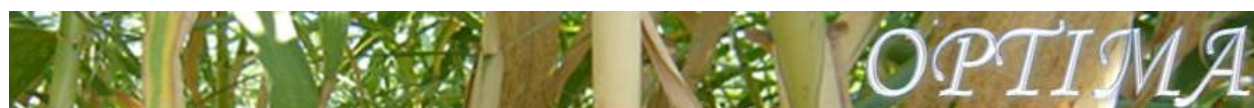


**Table 3 – CRES.** Results from the calorific value (GCV, NCV) and proximate analysis (volatiles, ash and fixed carbon) on samples that were collected at the end of the second growing period.

|                 | Calorific value (Kcal/kg) |             | Proximate analysis (%) |             |              |
|-----------------|---------------------------|-------------|------------------------|-------------|--------------|
|                 | GCV                       | NCV         | Volatiles              | Ash         | Fixed carbon |
| Alamo (stems)   | 4624                      | 4321        | 79.77                  | 1.86        | 14.38        |
| Alamo (leaves)  | 4471                      | 4168        | 78.59                  | 7.31        | 14.12        |
| Alamo           | <b>4563</b>               | <b>4260</b> | <b>79.30</b>           | <b>4.01</b> | <b>16.70</b> |
| Kanlow (stems)  | 4644                      | 4341        | 80.06                  | 1.65        | 18.30        |
| Kanlow (leaves) | 4499                      | 4196        | 78.43                  | 7.45        | 14.13        |
| Kanlow          | <b>4588</b>               | <b>4285</b> | <b>79.43</b>           | <b>3.89</b> | <b>16.69</b> |
| D1 (stems)      | 4637                      | 4334        | 80.21                  | 1.73        | 18.07        |
| D1 (stems)      | 4492                      | 4189        | 78.62                  | 7.55        | 13.85        |
| D1              | <b>4580</b>               | <b>4277</b> | <b>79.58</b>           | <b>4.02</b> | <b>16.41</b> |
| D2 (stems)      | 4631                      | 4328        | 79.62                  | 1.78        | 18.62        |
| D2 (leaves)     | 4477                      | 4174        | 78.41                  | 7.22        | 14.39        |
| D2              | <b>4571</b>               | <b>4268</b> | <b>79.15</b>           | <b>3.88</b> | <b>16.98</b> |
| Mean            | <b>4576</b>               | <b>4273</b> | <b>79.37</b>           | <b>3.95</b> | <b>16.69</b> |



**Figure 7 – CRES.** View of the experimental layout of miscanthus trial (tested factors: nitrogen rates and plant densities in three blocks)





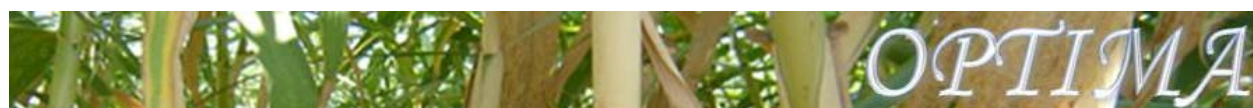
**Figure 8 – CRES.** View of the experimental field of miscanthus at the end of June 2012 (young plants) and at the end of July 2012.

**Table 4 – CRES.** Growth characteristics (plant height, number of tillers/m<sup>2</sup> and tiller density) for three subsequent growing periods.

| Years                             | Plant height (cm) |            |            | Number of tillers/m <sup>2</sup> |            |            | Tiller diameter (mm) |             |             |
|-----------------------------------|-------------------|------------|------------|----------------------------------|------------|------------|----------------------|-------------|-------------|
|                                   | 2013              | 2014       | 2015       | 2013                             | 2014       | 2015       | 2013                 | 2014        | 2015        |
| <b>Distances between the rows</b> |                   |            |            |                                  |            |            |                      |             |             |
| <b>D1: 50 cm</b>                  | 242               | 308        | 326        | 230                              | 228        | 227        | 7.60                 | 8.65        | 8.83        |
| <b>D2:100 cm</b>                  | 261               | 311        | 300        | 214                              | 235        | 242        | 7.69                 | 8.90        | 9.08        |
| <b>Irrigation rates</b>           |                   |            |            |                                  |            |            |                      |             |             |
| <b>I0</b>                         | -                 | 266        | 278        | -                                | 166        | 192        | -                    | 8.40        | 8.85        |
| <b>I1</b>                         | -                 | 312        | 296        | -                                | 202        | 212        | -                    | 8.67        | 8.90        |
| <b>I2</b>                         | -                 | 320        | 308        | -                                | 269        | 262        | -                    | 8.70        | 8.98        |
| <b>I3</b>                         | -                 | 341        | 369        | -                                | 289        | 272        | -                    | 9.37        | 9.09        |
| <b>Mean</b>                       | <b>252</b>        | <b>309</b> | <b>313</b> | <b>222</b>                       | <b>231</b> | <b>234</b> | <b>7.65</b>          | <b>8.78</b> | <b>8.95</b> |

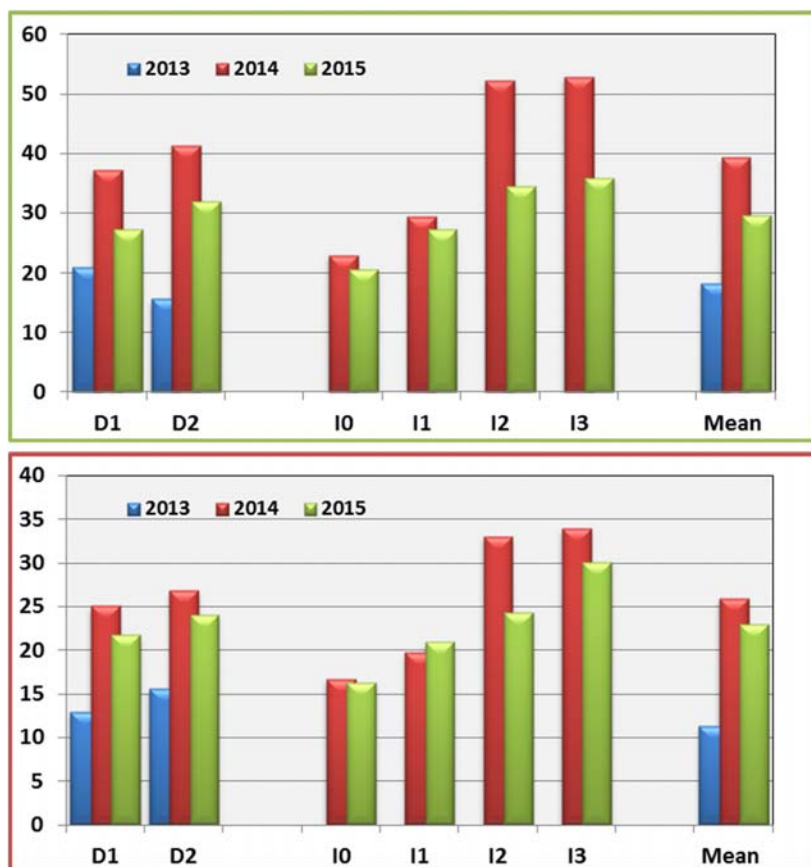
**Table 5 – CRES.** Yields results (fresh and dry matter yields, % stems on dry basis) from three subsequent growing seasons.

| Years                             | Fresh biomass yields (t/ha) |              |              | Dry matter yields (t/ha) |              |              | % stems on the biomass (oven-dried) |              |              |
|-----------------------------------|-----------------------------|--------------|--------------|--------------------------|--------------|--------------|-------------------------------------|--------------|--------------|
|                                   | 2013                        | 2014         | 2015         | 2013                     | 2014         | 2015         | 2013                                | 2014         | 2015         |
| <b>Distances between the rows</b> |                             |              |              |                          |              |              |                                     |              |              |
| <b>D1: 50 cm</b>                  | 20.92                       | 37.17        | 27.13        | 12.92                    | 25.04        | 17.22        | 52.50                               | 72.01        | 60.40        |
| <b>D2:100 cm</b>                  | 15.64                       | 41.35        | 31.93        | 9.64                     | 26.78        | 18.70        | 49.70                               | 70.75        | 67.86        |
| <b>Irrigation rates</b>           |                             |              |              |                          |              |              |                                     |              |              |
| <b>I0</b>                         | -                           | 22.92        | 20.50        | -                        | 16.96        | 12.19        | -                                   | 69.53        | 58.29        |
| <b>I1</b>                         | -                           | 29.29        | 27.25        | -                        | 19.70        | 16.70        | -                                   | 67.94        | 55.60        |
| <b>I2</b>                         | -                           | 52.54        | 34.45        | -                        | 33.02        | 19.51        | -                                   | 70.62        | 70.65        |
| <b>I3</b>                         | -                           | 52.29        | 35.91        | -                        | 33.96        | 23.45        | -                                   | 77.74        | 72.53        |
| <b>Mean</b>                       | <b>18.27</b>                | <b>39.26</b> | <b>29.53</b> | <b>11.27</b>             | <b>25.91</b> | <b>17.96</b> | <b>51.10</b>                        | <b>71.38</b> | <b>64.27</b> |



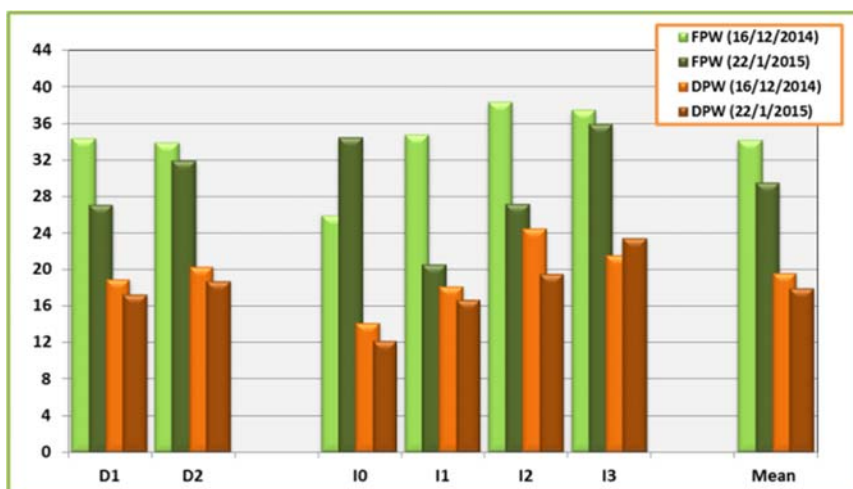


**Figure 9 – CRES.** Effect of irrigation rate on the miscanthus growth.



**Figure 10 – CRES.** Comparison of fresh and dry matter yields (t/ha) among the three growing seasons (Jan 2013/Jan 2014/Jan 2015).

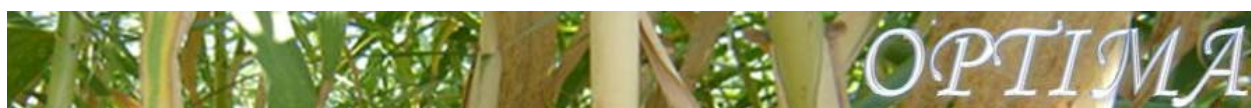




**Figure 11 – CRES.** Comparison of fresh and dry matter yields (t/ha) between the two harvest times (Dec 2014 and Jan. 2015)

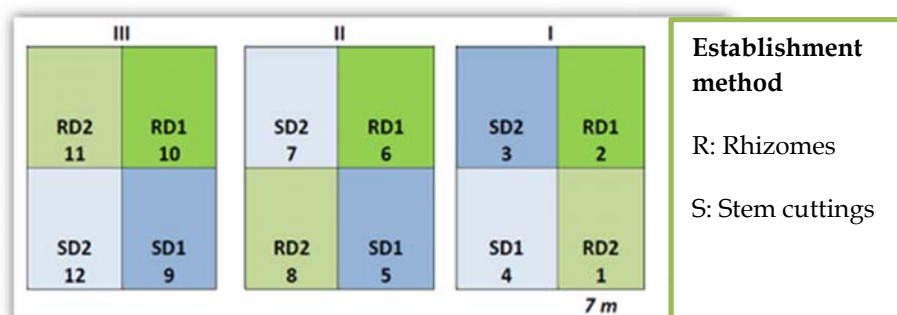
**Table 6 – CRES.** Results from the calorific value (GCV, NCV) and proximate analysis on samples that were collected at the end of the second growing period

| Perennial grasses | Calorific value (kcal/kg) |      | Proximate analysis (%) |       |              |
|-------------------|---------------------------|------|------------------------|-------|--------------|
|                   | GCV                       | NCV  | Volatiles              | Ash   | Fixed carbon |
| I0D1 (stems)      | 4651                      | 4348 | 80.53                  | 1.68  | 17.79        |
| (leaves)          | 4307                      | 4004 | 73.00                  | 11.84 | 15.16        |
| I0D2 (stems)      | 4621                      | 4318 | 80.99                  | 1.72  | 17.30        |
| (leaves)          | 4325                      | 4022 | 73.72                  | 11.29 | 15.00        |
| I1D1 (stems)      | 4701                      | 4398 | 80.82                  | 1.80  | 17.39        |
| (leaves)          | 4230                      | 3927 | 72.95                  | 12.59 | 14.46        |
| I1D2 (stems)      | 4635                      | 4332 | 80.52                  | 1.70  | 17.79        |
| (leaves)          | 4309                      | 4006 | 73.78                  | 11.67 | 14.57        |
| I2D1 (stems)      | 4644                      | 4341 | 80.70                  | 1.57  | 17.74        |
| (leaves)          | 4451                      | 4118 | 76.44                  | 8.63  | 14.94        |
| I2D2 (stems)      | 4613                      | 4310 | 80.81                  | 1.55  | 17.65        |
| (leaves)          | 4369                      | 4066 | 75.61                  | 9.42  | 14.98        |
| I3D1 (stems)      | 4637                      | 4334 | 80.94                  | 1.79  | 17.28        |
| (leaves)          | 4326                      | 4023 | 74.90                  | 10.37 | 14.47        |
| I3D2 (stems)      | 4682                      | 4337 | 80.90                  | 1.67  | 17.44        |
| (leaves)          | 4328                      | 4025 | 75.25                  | 10.21 | 14.56        |



**Table 7 – CRES.** Results from the calorific value (GCV, NCV) and proximate analysis (volatiles, ash and fixed carbon) on samples that were collected at the end of the second growing period

| Perennial grasses | Calorific value (kcal/kg) |      | Proximate analysis (%) |       |              |
|-------------------|---------------------------|------|------------------------|-------|--------------|
|                   | GCV                       | NCV  | Volatiles              | Ash   | Fixed carbon |
| I0D1 stems        | 4660                      | 4357 | 81.46                  | 1.59  | 16.96        |
| I0D1 leaves       | 4389                      | 4086 | 75.89                  | 9.31  | 14.81        |
| I0D2              | 4685                      | 4382 | 81.14                  | 1.69  | 17.17        |
|                   | 4331                      | 4028 | 74.21                  | 10.86 | 14.94        |
| I1D1              | 4678                      | 4375 | 80.95                  | 1.79  | 17.26        |
|                   | 4362                      | 4060 | 75.24                  | 10.13 | 14.64        |
| I1D2              | 4660                      | 4357 | 80.88                  | 1.81  | 17.32        |
|                   | 4309                      | 4006 | 74.46                  | 11.28 | 14.27        |
| I2D1              | 4706                      | 4403 | 81.46                  | 1.44  | 17.11        |
|                   | 4443                      | 4140 | 75.47                  | 9.27  | 15.26        |
| I2D2              | 4670                      | 4367 | 81.08                  | 1.64  | 17.28        |
|                   | 4436                      | 4133 | 76.25                  | 8.68  | 15.07        |
| I3D1              | 4650                      | 4347 | 80.76                  | 1.76  | 17.49        |
|                   | 4364                      | 4061 | 75.62                  | 10.03 | 14.36        |
| I3D2              | 4681                      | 4378 | 81.28                  | 1.59  | 17.14        |
|                   | 4352                      | 4049 | 75.73                  | 10.09 | 14.19        |



**Figure 12 – CRES.** Experimental layout of giant reed established in Aliartos in 2012.

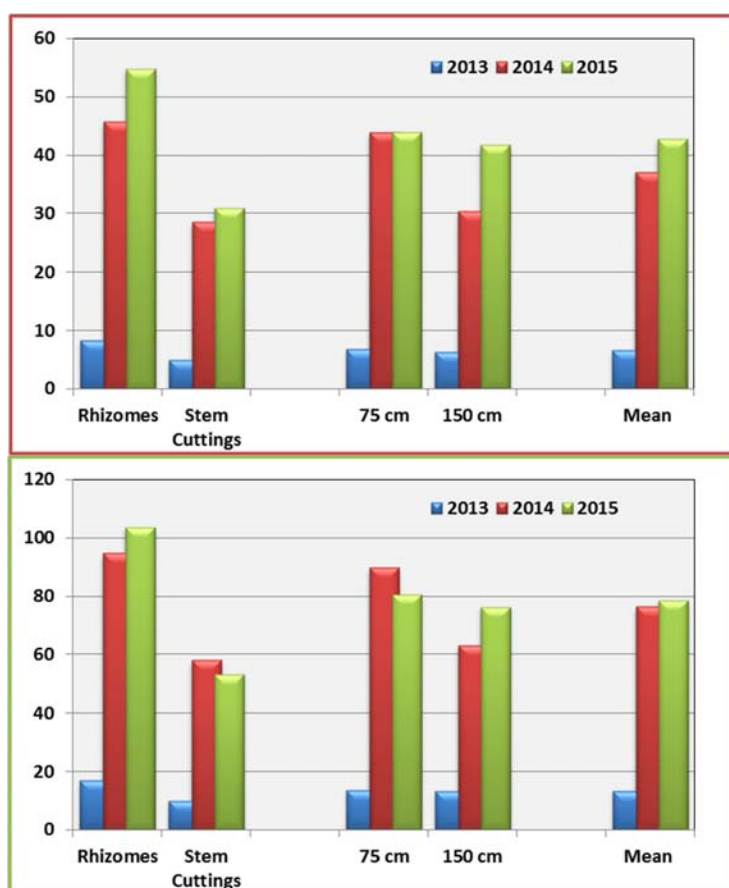
**Table 8 – CRES.** Effect of way of establishment (rhizomes or stem cuttings) and plant density on growth (plant height, tiller density per square meter and tiller diameter) for three subsequent growing periods.

| Years                            | Plant height (cm) |      |      | Number of tillers/m <sup>2</sup> |      |      | Tiller diameter (mm) |       |       |
|----------------------------------|-------------------|------|------|----------------------------------|------|------|----------------------|-------|-------|
|                                  | 2013              | 2014 | 2015 | 2013                             | 2014 | 2015 | 2013                 | 2014  | 2015  |
| <b>Propagation methods</b>       |                   |      |      |                                  |      |      |                      |       |       |
| Rhizomes                         | 414               | 603  | 625  | 8.0                              | 14.3 | 16.0 | -                    | 24.04 | 24.82 |
| S. cuttings                      | 244               | 601  | 610  | 4.6                              | 10.6 | 18.2 | -                    | 25.40 | 24.64 |
| <b>Densities within the rows</b> |                   |      |      |                                  |      |      |                      |       |       |
| D1: 75 cm                        | 337               | 599  | 623  | 6.4                              | 10.9 | 14.4 | -                    | 25.18 | 25.43 |
| D2: 150 cm                       | 322               | 605  | 613  | 6.2                              | 13.8 | 17.7 | -                    | 24.27 | 24.03 |
| Mean                             | 329               | 602  | 618  | 6.3                              | 12.4 | 16.0 | -                    | 24.72 | 24.73 |



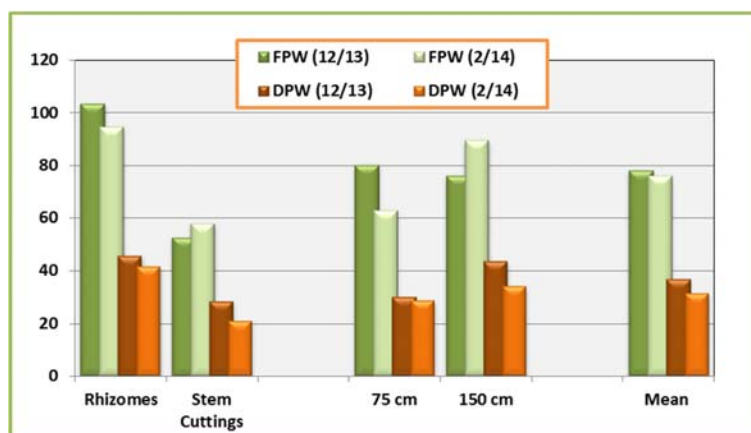
**Table 9 – CRES.** Effect of way of establishment (rhizomes or stem cuttings) and plant density on yields (plant height, tiller density per square meter and tiller diameter) for three subsequent growing periods.

| Years                            | Fresh yields (t/ha) |              |              | Dry matter yields (t/ha) |              |              | Early harvest (2/12/13) |              |
|----------------------------------|---------------------|--------------|--------------|--------------------------|--------------|--------------|-------------------------|--------------|
|                                  | 2013                | 2014         | 2015         | 2013                     | 2014         | 2015         | FFY                     | DMY          |
| <b>Propagation methods</b>       |                     |              |              |                          |              |              |                         |              |
| <b>Rhizomes</b>                  | 16.85               | 94.58        | 103.37       | 8.24                     | 45.63        | 54.67        | 103.38                  | 41.85        |
| <b>S. cuttings</b>               | 9.78                | 57.83        | 53.00        | 4.93                     | 28.49        | 30.83        | 53.00                   | 21.22        |
| <b>Densities within the rows</b> |                     |              |              |                          |              |              |                         |              |
| <b>D1: 75 cm</b>                 | 13.50               | 62.88        | 80.21        | 6.78                     | 30.29        | 43.91        | 80.21                   | 28.86        |
| <b>D2: 150 cm</b>                | 13.13               | 89.54        | 76.17        | 6.38                     | 43.82        | 41.59        | 76.16                   | 34.22        |
| <b>Mean</b>                      | <b>13.31</b>        | <b>76.21</b> | <b>78.18</b> | <b>6.58</b>              | <b>37.06</b> | <b>42.75</b> | <b>78.18</b>            | <b>31.54</b> |



**Figure 13 – CRES.** Comparison of fresh and dry matter yields (t/ha) between the first and the second growing season (Jan 2013 vs. Jan 2014).

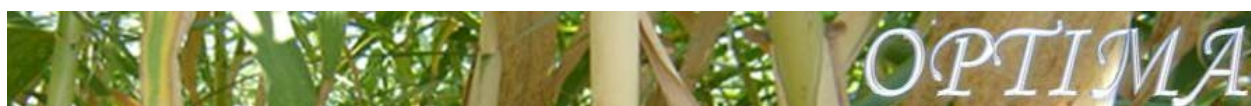




**Figure 14 – CRES.** Fresh and dry matter yields (t/ha) for the two tested factors (ways of establishment and densities within the rows) in two harvest times (Dec. 2013 and Feb. 2014)

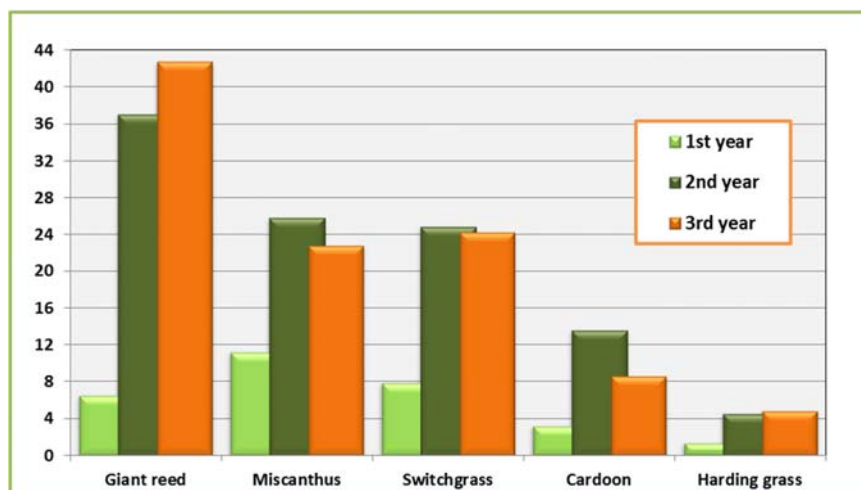
**Table 10 – CRES.** Results from the proximate analysis (volatiles, ash and fixed carbon) on samples that were collected at the end of the second growing period.

|             | Proximate analysis (%) |       |              |
|-------------|------------------------|-------|--------------|
|             | Volatiles              | Ash   | Fixed carbon |
|             | 2/12/13                |       |              |
| RD1 (stems) | 77.86                  | 2.86  | 12.29        |
| (leaves)    | 71.92                  | 12.59 | 15.49        |
| RD2 (stems) | 77.84                  | 2.91  | 12.26        |
| (leaves)    | 65.88                  | 20.12 | 14.00        |
| SD1 (stems) | 78.22                  | 2.86  | 18.93        |
| (leaves)    | 69.09                  | 16.36 | 14.56        |
| SD2 (stems) | 77.82                  | 2.78  | 19.41        |
| (leaves)    | 70.04                  | 15.35 | 14.62        |
|             | 4/2/14                 |       |              |
| RD1 (stems) | 76.96                  | 3.43  | 19.62        |
| (leaves)    | 71.71                  | 13.12 | 15.19        |
| RD2 (stems) | 77.52                  | 3.42  | 19.07        |
| (leaves)    | 71.82                  | 12.75 | 15.46        |
| SD1 (stems) | 77.54                  | 3.07  | 19.39        |
| (leaves)    | 72.50                  | 11.73 | 15.77        |
| SD2 (stems) | 77.56                  | 3.13  | 19.29        |
| (leaves)    | 73.52                  | 9.96  | 16.53        |



**Table 11 – CRES.** Results from the calorific value (GCY, NCY) and elemental analysis (C, H, N) on samples from stems and leaves that were collected in February 2014.

|               | Calorific value (kcal/kg) |                | Elemental analysis (%) |             |             |
|---------------|---------------------------|----------------|------------------------|-------------|-------------|
|               | GCV                       | NCV            | C                      | H           | N           |
| <b>Stems</b>  | <b>4485.16</b>            | <b>4189.23</b> | <b>46.58</b>           | <b>5.86</b> | <b>0.28</b> |
| <b>Leaves</b> | <b>4068.45</b>            | <b>3808.88</b> | <b>38.38</b>           | <b>5.19</b> | <b>0.78</b> |



**Figure 15 – CRES.** Comparison among five perennial crops (switchgrass, giant reed, miscanthus, cardoon and Harding grass) in terms of dry yields for three subsequent years.



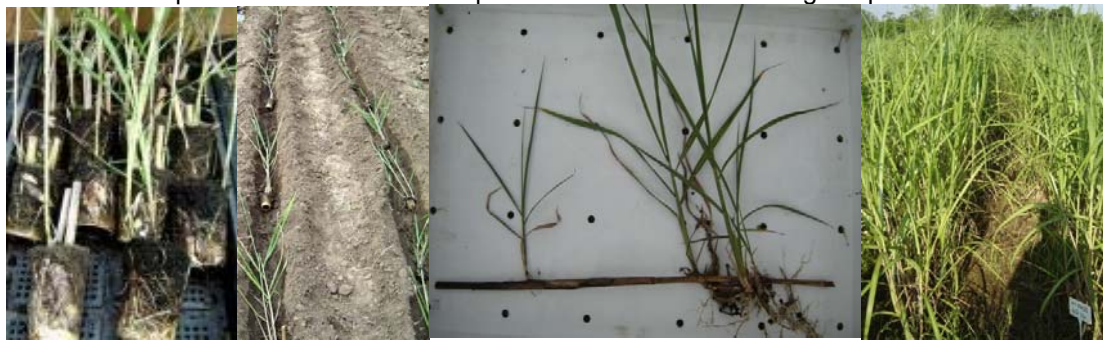
## Primus Ltd. – PRIMUS (n. 20)

**Table 1 - PRIMUS.** Maximum and minimum air temperatures (°C) during transplanting dates (2012) of miscanthus (*Miscanthus x giganteus* Greef et Deuter) stem cuttings at the experimental farm of PRIMUS Ltd., Hungary.

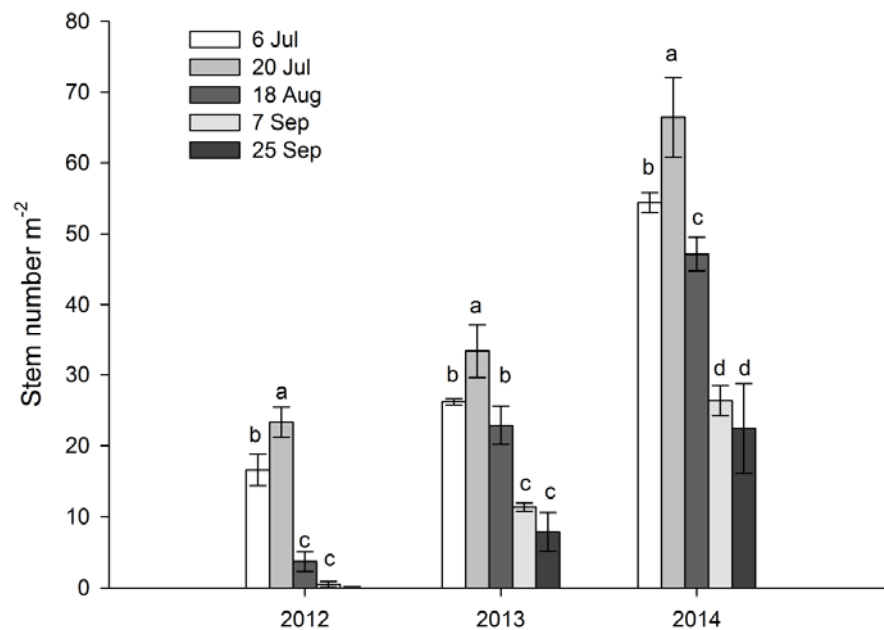
| Transplanting time | Tmax | Tmin |
|--------------------|------|------|
| July 6             | 25   | 15   |
| July 23            | 34   | 21   |
| August 18          | 29   | 14   |
| September 7        | 28   | 5    |
| September 23       | 19   | 4    |



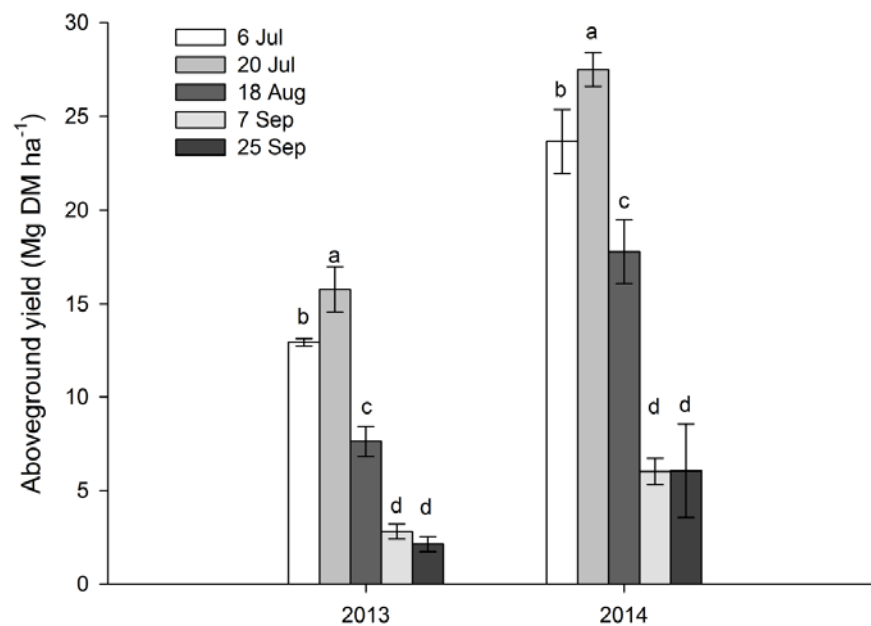
**Figure 1 – PRIMUS.** Pictures of non-sprouting (non-activated) and shooting stems after 3-4-week-growth in 'quasi in vitro' nursing systems (left2). BAP in high concentration induced shoot multiplication on newly developed shoots. High concentration of BAP caused heavy trichome production on leaves also. BAP-induced shoot proliferation and trichome production can be seen on right 2 pictures.



**Figure 2 – PRIMUS.** Usage of new type of propagating materials in plots. Left to right: plantlets derived from activated stem cuttings and its usage as planting material in autumn; shoot development on activated stems was used as direct planting material; plantlets were developed from activated shoots collected in the same year (establishment was in early July, picture made in autumn)



**Figure 3 - PRIMUS.** Stem number in a square meter (no. m<sup>-2</sup>) of *Miscanthus x giganteus* at first, second and third growing season (2012, 2013 and 2014, respectively) at PRIMUS Ltd, Hungary. Different letters within the same year represent statistically different means ( $P \leq 0.05$ ).



**Figure 4 - PRIMUS.** Aboveground dry matter yield (Mg DM ha<sup>-1</sup>) of *Miscanthus x giganteus* at second and third growing season (2013 and 2014, respectively) at PRIMUS Ltd, Hungary. Different letters within the same year represent statistically different means ( $P \leq 0.05$ ).





**Figure 5 - PRIMUS.** Growth of several-year-old *Miscanthus* plants after 3 dry and 2 rainy summers in a sandy hill (Autumn, 2015).

## Task 4.1 Intercropping on marginal land

Center for Renewable Energy Sources and Energy Saving – CRES (n. 3)

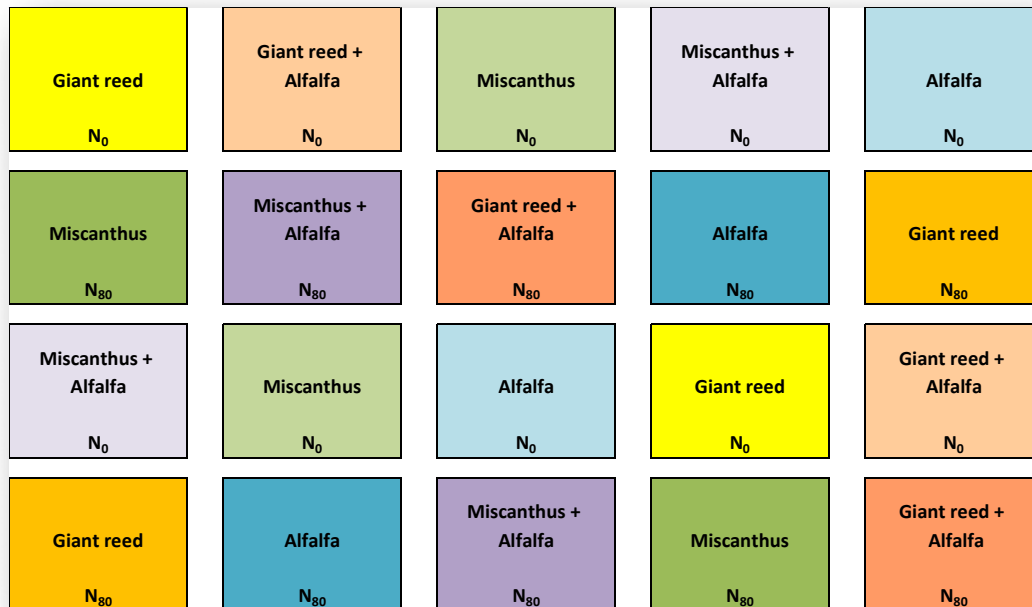


Figure 1 - CRES. Experimental design

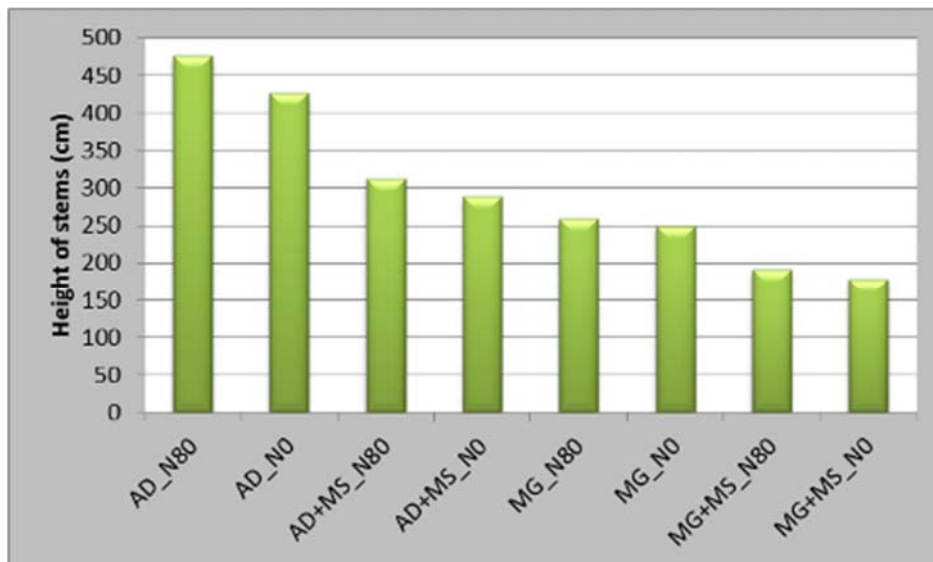
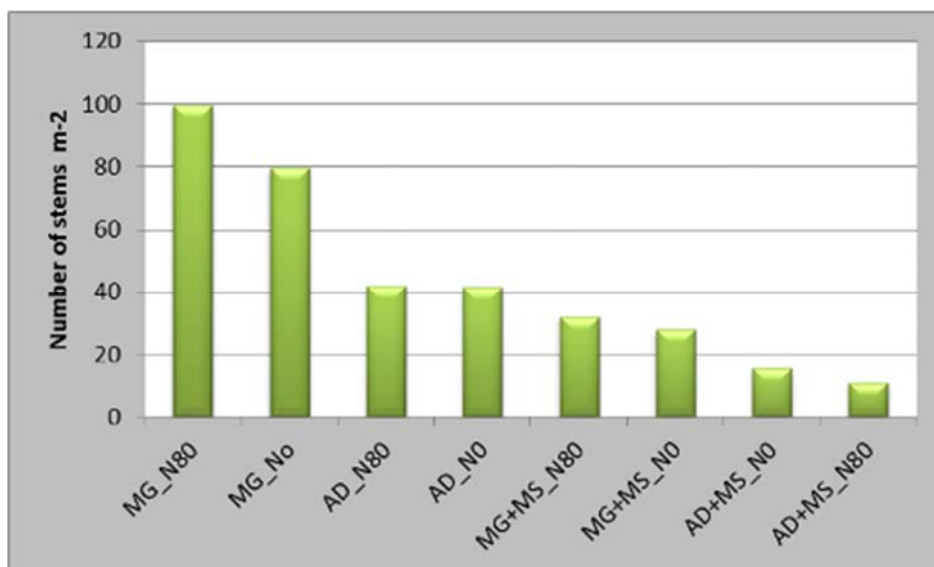
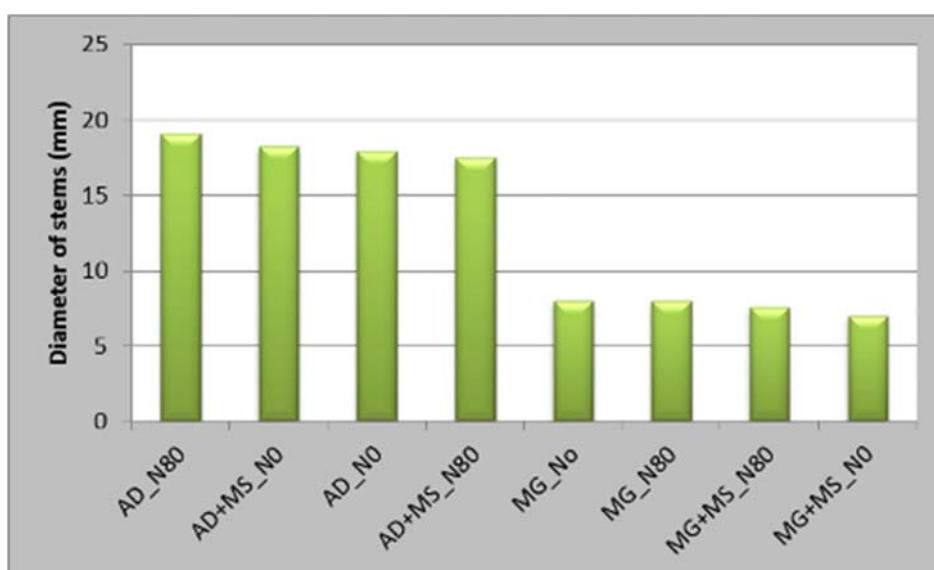


Figure 2 – CRES. Height of stems for *Arundo donax* (AD) and *Miscanthus x Giganteus* (MG) in pure stands and in mixtures with *Medicago sativa* (MS) in two nitrogen fertilisation rates (N0 and N80).



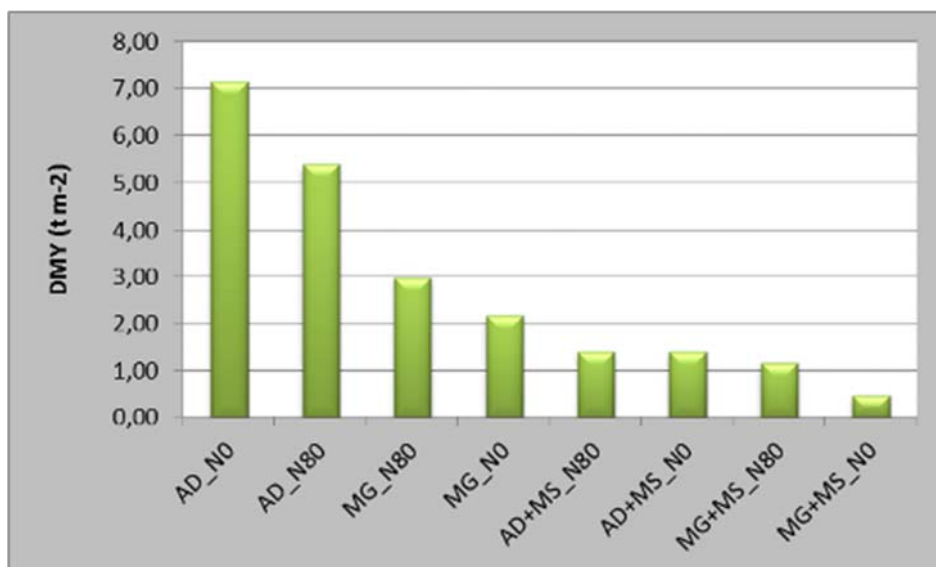


**Figure 3 – CRES.** Number of stems per square meter for Arundo donax (AD) and Miscanthus x Giganteus (MG) in pure stands and in mixtures with Medicago sativa (MS) in two nitrogen fertilisation rates (N0 and N80).

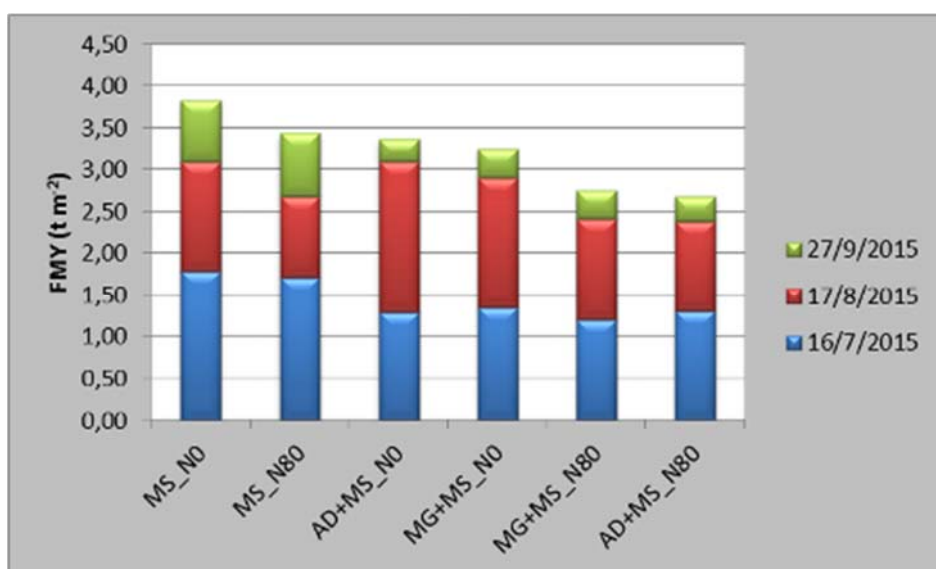


**Figure 4 – CRES.** Stem diameter for Arundo donax (AD) and Miscanthus x Giganteus (MG) in pure stands and in mixtures with Medicago sativa (MS) in two nitrogen fertilisation rates (N0 and N80).



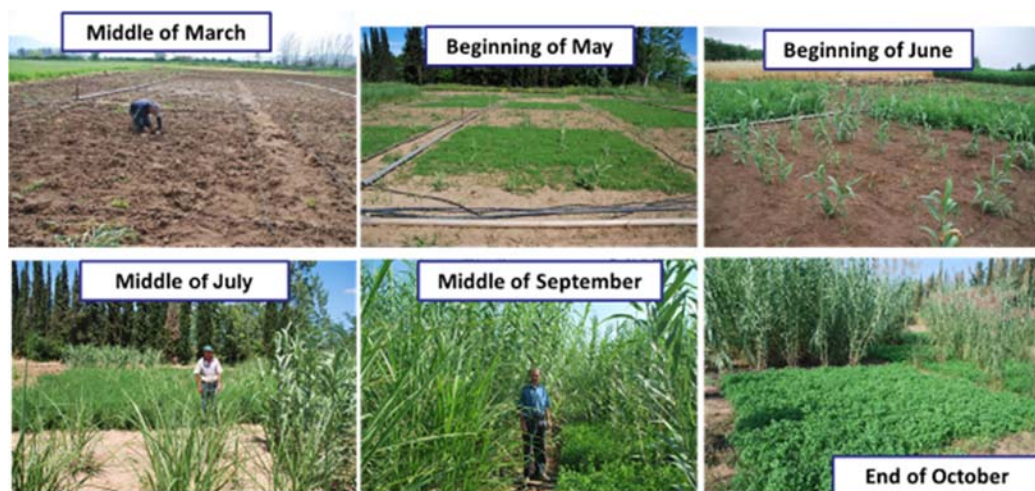


**Figure 5 – CRES.** Dry matter yields for Arundo donax (AD) and Miscanthus x Giganteus (MG) in pure stands and in mixtures with Medicago sativa (MS) in two nitrogen fertilisation rates (N0 and N80).

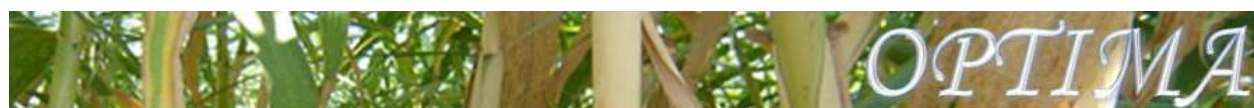


**Figure 6 – CRES.** Fresh matter yields for Medicago sativa (MS) harvested four times a year, in pure stands and in mixtures with Arundo donax (AD) and Miscanthus x Giganteus (MG), under two nitrogen fertilisation rates (N0 and N80).





**Figure 7 - CRES.** Field view at different phonological stages



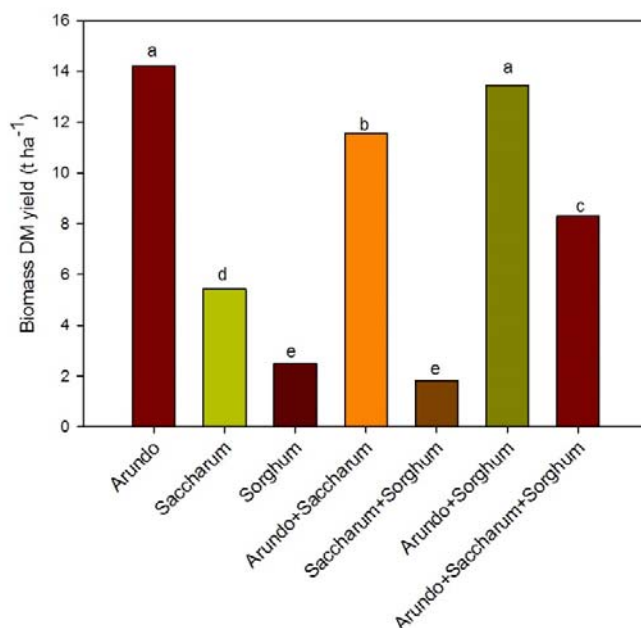
## Università di Catania (n. 1)

**Table 1 - UNICT.** Number of intercropped species in spring intercropping trial

| Number of intercropped species | Species                                           |
|--------------------------------|---------------------------------------------------|
| 3                              | <i>Arundo</i> + <i>Saccharum</i> + <i>Sorghum</i> |
| 2                              | <i>Arundo</i> + <i>Saccharum</i>                  |
| 2                              | <i>Arundo</i> + <i>Sorghum</i>                    |
| 2                              | <i>Saccharum</i> + <i>Sorghum</i>                 |
| 1                              | <i>Arundo</i>                                     |
| 1                              | <i>Saccharum</i>                                  |
| 1                              | <i>Sorghum</i>                                    |



**Picture 1 - UNICT.** *Arundo*+*Saccharum* (sx), *Sorghum*+*Saccharum* (middle) and *Sorghum*+*Arundo*+*Saccharum* (dx)



**Figure 1 - UNICT.** Aboveground biomass yield (t dry matter ha<sup>-1</sup>) of intercropping levels and pure stands in the average of fertilization rates at 3<sup>rd</sup> year harvest. Different letters indicate significant differences at  $P \leq 0.05$  by SNK Test.

**Table 2 - UNICT.** Nitrogen fertilization effect on aboveground biomass yield ( $\text{t ha}^{-1}$ ) in the average of intercropping levels at 3<sup>rd</sup> year harvest (February 2015). Different letters indicate significant differences at  $P \leq 0.05$  by SNK Test.

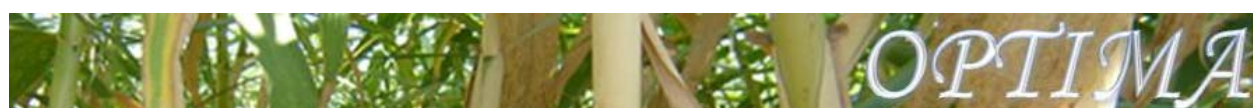
| Nitrogen fertilization ( $\text{kg ha}^{-1}$ ) | Average biomass yield ( $\text{t ha}^{-1}$ ) |
|------------------------------------------------|----------------------------------------------|
| 0                                              | 7.07c                                        |
| 40                                             | 8.27b                                        |
| 80                                             | 9.22a                                        |

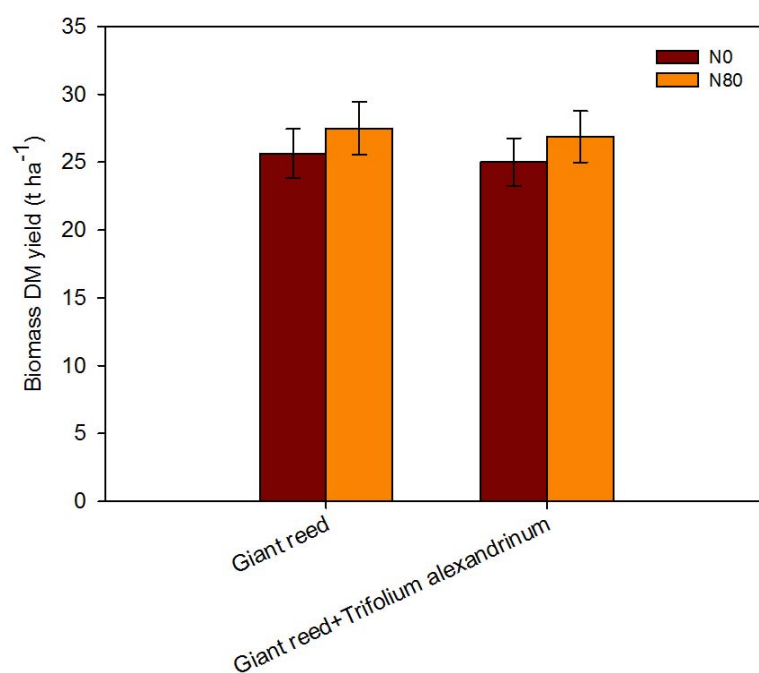
**Table 3 - UNICT.** Number of intercropped species

| Number of intercropped species | Species                                    |
|--------------------------------|--------------------------------------------|
| 2                              | <i>Miscanthus</i> + <i>T. alexandrinum</i> |
| 2                              | <i>Arundo</i> + <i>T. alexandrinum</i>     |
| 1                              | <i>Arundo</i>                              |
| 1                              | <i>Miscanthus</i>                          |
| 1                              | <i>T. alexandrinum</i>                     |



**Picture 2 - UNICT.** *Arundo* and *T. alexandrinum* in early-spring (left), middle of summer-time (center) and end-of summer after some irrigations (left)





**Figure 2 - UNICT.** Aboveground biomass yield (t dry matter ha<sup>-1</sup>) of intercropping levels and pure stands in the average of fertilization rates at the winter harvest (February 2015).

**Table 4 - UNICT.** Aboveground biomass yield (t ha<sup>-1</sup>) in relation to the different studied treatments (mixture and fertilization rate).

| Mixture                           | Fertilization rate | DM yield (tha-1) |
|-----------------------------------|--------------------|------------------|
| Giant reed                        | N0                 | 25.63±1.81       |
| Giant reed+Trifolium alexandrinum | N0                 | 25.00±1.76       |
| Giant reed                        | N80                | 27.50±1.94       |
| Giant reed+Trifolium alexandrinum | N80                | 26.88±1.90       |
| <i>Average</i>                    |                    |                  |
| N0                                |                    | 25.31±1.79       |
| N80                               |                    | 27.19±1.92       |
| Giant reed                        |                    | 26.56±1.88       |
| Giant reed+Trifolium alexandrinum |                    | 25.94±1.83       |



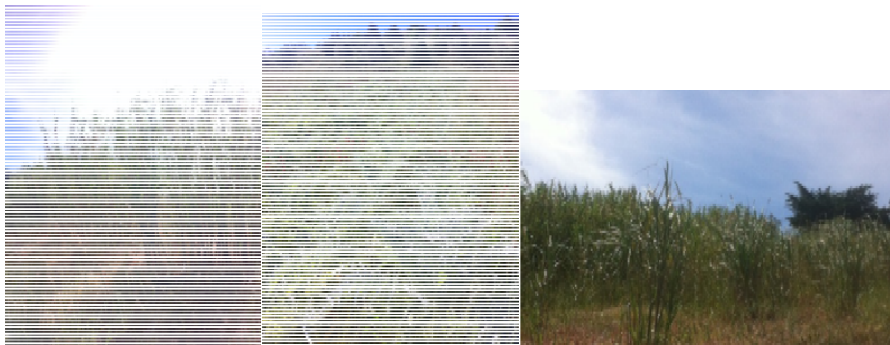
**Table 5 – UNICT.** Biometric and productive measurements of 5 ha giant reed plantation (harvest February 2015).

| Measurement                   | Unit               | Value       |
|-------------------------------|--------------------|-------------|
| # of emerged plants           | m <sup>2</sup>     | 19.7±4.3    |
| uniformity after transplant   | visual score       | Medium-high |
| height of stems               | Cm                 | 223.5±82.1  |
| Dry stems                     | %                  | 85.1±12.3   |
| Dry leaves                    | %                  | 13.1±8.7    |
| Aboveground dry biomass yield | t ha <sup>-1</sup> | 12.1±2.9    |

**Table 6 - UNICT.** Morpho-biometric and productive characters of *Arundo donax*, *Saccharum spontaneum* and cardoon in the marginal slope field.

|                   | Measurement unit   | <i>Arundo donax</i> | <i>Saccharum spontaneum</i> | <i>Cynara cardunculus</i> |
|-------------------|--------------------|---------------------|-----------------------------|---------------------------|
| Plant height      | Cm                 | 223.2±38.2          | 94.1±26.4                   | 98.6±11.4                 |
| Stems             | %                  | 87.1±4.0            | 79±5.1                      | 36.8±3.2                  |
| Leaves            | %                  | 12.9±4.0            | 21±5.1                      | 40.5±6.1                  |
| Heads             | %                  |                     |                             | 19.15±5.7                 |
| Hachenes          | %                  |                     |                             | 3.55±0.41                 |
| H <sub>2</sub> O  | %                  | 45±2.7              | 53±3.2                      | 9.3±1.3                   |
| Dry biomass yield | t ha <sup>-1</sup> | 8.1±1.5             | 4.3±1.1                     | 5.7±0.9                   |

± standard deviation of three determinations



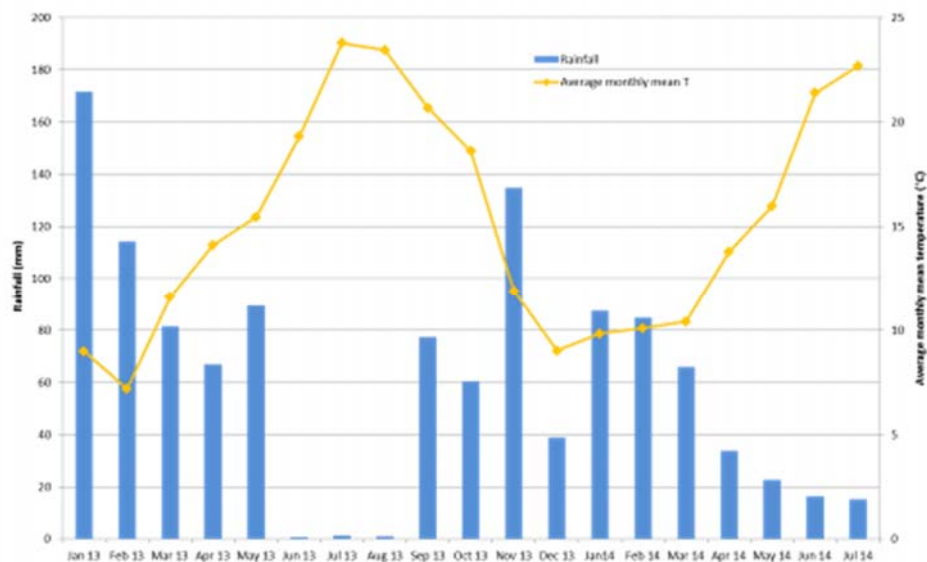
**Picture 3 - UNICT.** Giant reed (left) cardoon (center) and saccharum (right) in the in a marginal area with 26-28% slope during last growing season.



**Istituto per il Sistema Produzione Animale in Ambiente Mediterraneo – CNR-  
ISPAAM (n. 8)**

**Table 1 – CNR-ISPAAM.** Seed rates and number of germinating seeds in legume crops intercropped in biomass crop (autumn sowing).

| species                | Accession      | n. of rhizomes $m^{-2}$ /sowing rate seed $ha^{-1}$ |
|------------------------|----------------|-----------------------------------------------------|
| <i>A. donax</i>        | Local landrace | 2                                                   |
| <i>M. x giganteus</i>  | unknown        | 2                                                   |
| <i>T. alexandrinum</i> | Cv Laura       | 42 kg                                               |



**Figure 1 – CNR-ISPAAM.** The weather pattern in the period January 2013 - August 2014.

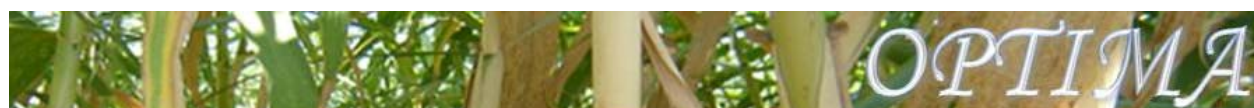


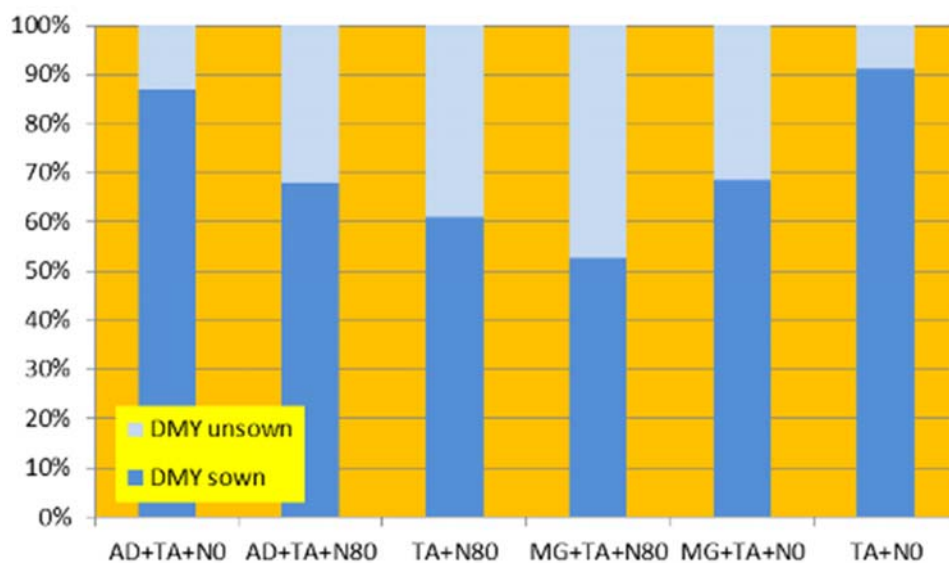


**Figure 2 - CNR-ISPAAAM.** Seedling establishment of *T. alexandrinum* (TA) intercropped with the conventional bioenergy species *A. donax* (AD) and *M. x giganteus* (MG) at two levels of nitrogen fertilization (N0= no fertilization and N80=80 kg of Nitrogen ha<sup>-1</sup>).

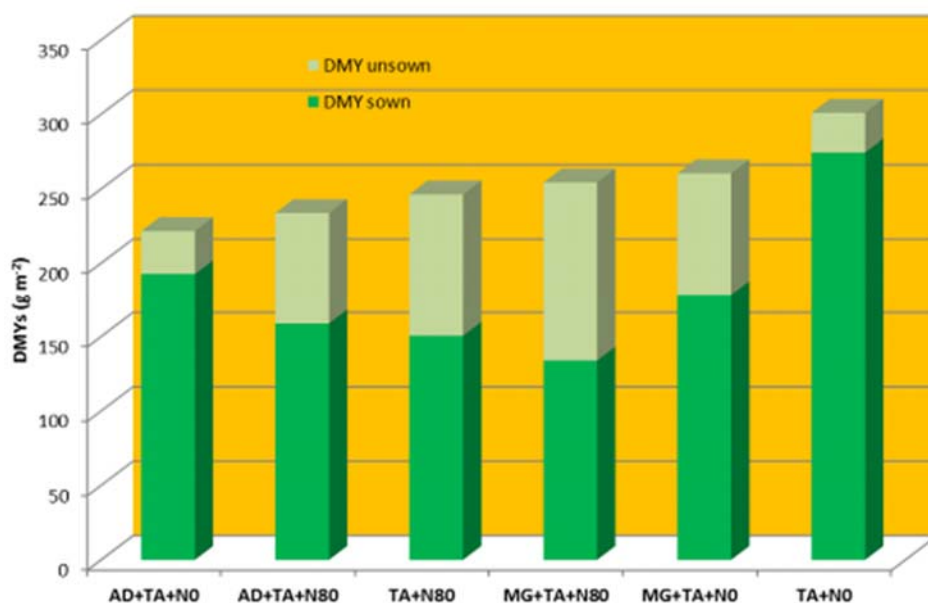


**Picture 1 – CNR-ISPAAAM.** Appearance of the pure stand of berseem clover, giant reed and giant silver reed intercropped with berseem clover at the end of summer 2014.





**Figure 3 - CNR-ISPAAM.** Rate of sown and unsown species in the DM for each treatment.



**Figure 4 - CNR-ISPAAM.** DMYs of *T. alexandrinum* and unsown species in different intercropped crops of bioenergy plants.



Indian Institute of Technology, Delhi – CRDT (n. 13)



(a)



(b)

**Picture 1 – CRDT.** Germination of Switchgrass with a) Moist cotton treatment (after 12 days) b) with different seed treatment methods.

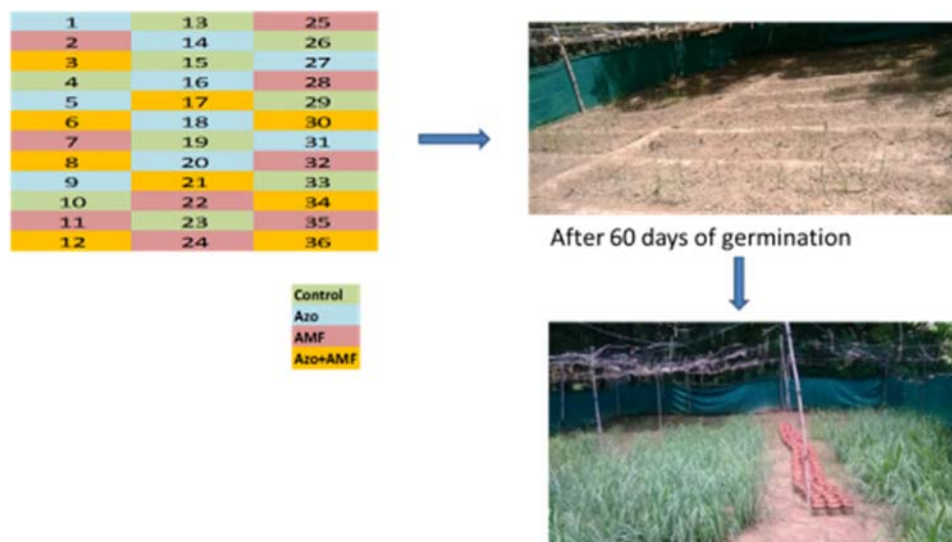
**Table 1 - CRDT.** Seed viability/germination (%) (at lab level in petri plates)

| Methods Used         |  | No. of seeds germinated (No. of seeds tested = 25) |                     |                     |                      |                      |               |
|----------------------|--|----------------------------------------------------|---------------------|---------------------|----------------------|----------------------|---------------|
|                      |  | 1 <sup>st</sup> Day                                | 4 <sup>th</sup> day | 8 <sup>th</sup> Day | 12 <sup>th</sup> Day | 15 <sup>th</sup> Day | % Germination |
| Moist blotting paper |  | 0                                                  | 0                   | 0                   | 1                    | 9                    | 36            |
| Moist cotton         |  | 0                                                  | 0                   | 0                   | 5                    | 15                   | 60            |
| 2% Water agar        |  | 0                                                  | 0                   | 0                   | 2                    | 7                    | 28            |

**Table 2 – CRDT.** Germination % of seeds of *P. virgatum*

| Treat-ments   | Hot water (80°C, 4 min) | Cold water (4min) | Cold water (over-night) | 1% H <sub>2</sub> SO <sub>4</sub> (5 min) | 0.1% H <sub>2</sub> SO <sub>4</sub> (5min) | 10 ppm GA (5min) | Bioinoculants ( <i>Azospirillum</i> ) | Bioinoculants (AMF) | Bioinoculants ( <i>Azospirillum</i> +AMF) | Control |
|---------------|-------------------------|-------------------|-------------------------|-------------------------------------------|--------------------------------------------|------------------|---------------------------------------|---------------------|-------------------------------------------|---------|
| Germination % | 10±0.5                  | 6±0.7             | 36±0.7                  | 9±0.5                                     | 15±1.0                                     | 54±0.5           | 58±1.0                                | 59±1.1              | 61±0.9                                    | 60±1.0  |





**Picture 2 – CRDT.** Switchgrass Cultivation at Micromodel, IIT Delhi (with RBD)



**Picture 3 – CRDT.** Cultivation of Napier grass at Micromodel, IIT Delhi

**Table 3 – CRDT.** Fresh biomass yield (t/ha/year) of grasses and legume under silvipastoral models at Micromodel, IIT Delhi and Farmers land (Block Farrukhnagar)

| Treatments                            | <i>C.setigerus</i> (cc337)<br>(Dhaman grass) |          | <i>C.setigerus</i> (cc58)<br>(Dhaman grass) |          | <i>P. pedicellatum</i><br>(Dinanath Grass) |          | <i>P. maximum</i><br>(Guinae Grass) |          |
|---------------------------------------|----------------------------------------------|----------|---------------------------------------------|----------|--------------------------------------------|----------|-------------------------------------|----------|
|                                       | I Yr                                         | II Yr    | I Yr                                        | II Yr    | I Yr                                       | II Yr    | I Yr                                | II Yr    |
| <b>At Micromodel, IIT Delhi</b>       |                                              |          |                                             |          |                                            |          |                                     |          |
| <b>Control (T1)</b>                   | 28.96±1.1                                    | 28.5±0.9 | 25.63±1.1                                   | 25.1±1.5 | 23.7±0.16                                  | 20.1±0.7 | 27.54±1.1                           | 27.1±1.0 |
| <b><i>Azospirillum</i> (T2)</b>       | 30.5±0.9                                     | 30.1±1.0 | 28.0±0.9                                    | 27.4±1.0 | 26.81±0.11                                 | 24.5±0.5 | 29.4±0.9                            | 28.8±0.9 |
| <b>AMF (T3)</b>                       | 31.9±1.0                                     | 31.4±0.5 | 28.2±1.0                                    | 27.5±1.0 | 26.78±0.12                                 | 24.4±0.4 | 29.5±1.0                            | 28.8±0.9 |
| <b><i>Azospirillum</i>+AMF (T4)</b>   | 33.7± 1.1                                    | 33.3±1.0 | 32.0±1.5                                    | 31.6±1.0 | 28.35±0.07                                 | 26.1±1.0 | 33.1±2.0                            | 32.9±1.0 |
| <b>At Vill. Farrukhnagar, Haryana</b> |                                              |          |                                             |          |                                            |          |                                     |          |
| <b>Control (T1)</b>                   | 15.1±1.1                                     | 14.5±0.5 | 14.9±1.5                                    | 14.1±0.5 | 22.9.0±2.0                                 | 19.4±1.0 | -                                   | 24.5±1.1 |
| <b><i>Azospirillum</i> (T2)</b>       | 16.1±1.5                                     | 15.7±0.5 | 15.93±0.5                                   | 15.5±0.3 | 25.11±1.25                                 | 23.1±1.1 | -                                   | 26.2±1.0 |
| <b>AMF (T3)</b>                       | 15.8±0.7                                     | 15.1±0.4 | 15.4±0.5                                    | 14.9±0.4 | 25.18±1.1                                  | 23.3±0.9 | -                                   | 26.3±1.0 |
| <b><i>Azospirillum</i>+AMF (T4)</b>   | 18.2±1.7                                     | 17.8±0.4 | 17.7±0.8                                    | 17.2±0.6 | 27.20±1.18                                 | 25.4±1.0 | -                                   | 30.0±1.0 |

***C. setigerus*:** 6 cuts in I year

**I Year:** Sowing - 14<sup>th</sup> April 2012; 1<sup>st</sup> Cut - 10<sup>th</sup> June 2012; 2<sup>nd</sup> Cut - 25<sup>th</sup> July 2012; 3<sup>rd</sup> cut - 17<sup>th</sup> August 2012; 4<sup>th</sup> cut – 25<sup>th</sup> September 2012; 5<sup>th</sup> cut – 10<sup>th</sup> November 2012; 6<sup>th</sup> cut – 13<sup>th</sup> March 2013

**II year:** 1<sup>st</sup> Cut - 28<sup>th</sup> May 2013; 2<sup>nd</sup> Cut – 3<sup>rd</sup> July 2013; 3<sup>rd</sup> cut - 10<sup>th</sup> August 2013; 4<sup>th</sup> cut – 12<sup>th</sup> September 2013; 5<sup>th</sup> cut – 31<sup>st</sup> Oct. 2013; 6<sup>th</sup> cut – 2<sup>nd</sup> March 2014

***P maximum*:** 4 cuts in I year

**I year:** Sowing – 17<sup>th</sup> July 2012; 1<sup>st</sup> Cut - 10<sup>th</sup> Sept 2012; 2<sup>nd</sup> Cut – 10<sup>th</sup> Oct. 2012; 3<sup>rd</sup> cut – 13<sup>th</sup> Jan. 2013; 4<sup>th</sup> cut: 6<sup>th</sup> May 2013

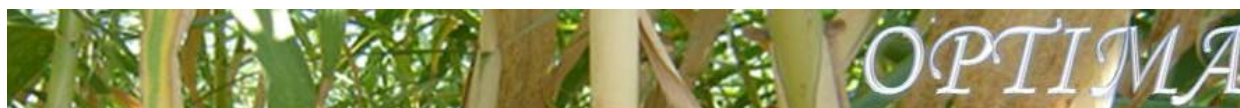
**II year:** 1<sup>st</sup> Cut - 18<sup>th</sup> Aug. 2013; 2<sup>nd</sup> Cut – 28<sup>th</sup> Sept. 2013; 3<sup>rd</sup> cut – 26<sup>th</sup> Dec. 2013; 4<sup>th</sup> cut: 23<sup>rd</sup> Apr. 2014

-*P. maximum* seeds did not germinate at village Farrukhnagar in the first year. In II year, root slips of the grass were transplanted.

***P. pedicellatum*:** 2 cuts/year

**I Year:** Sowing – 17<sup>th</sup> July 2012; 1<sup>st</sup> Cut: 10<sup>th</sup> Sept. 2012; 2<sup>nd</sup> cut: 15<sup>th</sup> November 2012

**II year:** ; Sowing – 10<sup>th</sup> July 2013; 1<sup>st</sup> Cut: 14<sup>th</sup> Sept. 2013; 2<sup>nd</sup> cut: 20<sup>th</sup> November 2013



**Table 4 – CRDT.** Fresh biomass yield (t/ha/year) of legumes under silvipastoral models at Micromodel, IIT Delhi and at field level (Block Farrukhnagar, Haryana)

| Treatments                   | <i>Trifolium alexandrinum</i> (Barseem)<br>At Micromodel, IIT Delhi |                | <i>Trifolium resupinatum</i> (Rijheka)<br>On Farmer's field (Block Farrukhnagar) |
|------------------------------|---------------------------------------------------------------------|----------------|----------------------------------------------------------------------------------|
|                              | <i>I Year</i>                                                       | <i>II Year</i> | <i>I Year</i>                                                                    |
| <b>Control</b>               | 3.5±0.4                                                             | 3.6±0.2        | 2.7±0.05                                                                         |
| <b><i>Rhizobium</i></b>      | 4.1±0.2                                                             | 4.5±0.4        | 3.1±0.2                                                                          |
| <b>AMF</b>                   | 4.3±0.2                                                             | 4.8±0.4        | 3.2±0.3                                                                          |
| <b><i>Rhizobium</i> +AMF</b> | 4.9±0.09                                                            | 5.1±0.5        | 3.7±0.1                                                                          |

*Trifolium alexandrinum*: *I year*: Sowing: 19th Jan.. 2013 (3 cuts till May 2013); *II Year*: Sowing: 25th Jan.. 2014 (3 cuts till May 2014)

*Trifolium resupinatum*: *I year*: Sowing: 24th Jan.. 2013 (3 cuts till May 2013);

**Table 5 – CRDT.** Effect of different biofertilizer treatments on survival % and growth parameters of tree sp. at Micromodel, IIT Delhi (after two years)

| Treatments               | Ber ( <i>Z. mauritiana</i> ) |                       |                   |                                         |                  | Kinnow ( <i>Citrus nobilis</i> ) |                       |                   |                                         |                  | Lemon ( <i>C. limon</i> ) |                       |                   |                                         |                  |
|--------------------------|------------------------------|-----------------------|-------------------|-----------------------------------------|------------------|----------------------------------|-----------------------|-------------------|-----------------------------------------|------------------|---------------------------|-----------------------|-------------------|-----------------------------------------|------------------|
|                          | S %                          | Avg shoot length (cm) |                   | Avg girth (cm) (30 cm above the ground) |                  | S%                               | Avg shoot length (cm) |                   | Avg girth (cm) (30 cm above the ground) |                  | S %                       | Avg shoot length (cm) |                   | Avg girth (cm) (30 cm above the ground) |                  |
|                          |                              |                       |                   |                                         |                  |                                  |                       |                   |                                         |                  |                           |                       |                   |                                         |                  |
|                          |                              | A                     | B                 | A                                       | B                |                                  | A                     | B                 | A                                       | B                |                           | A                     | B                 | A                                       | B                |
| Control                  | 62                           | 19                    | 73.4 <sup>a</sup> | 0.8                                     | 2.0 <sup>a</sup> | 54                               | 17                    | 65.7 <sup>a</sup> | 0.8                                     | 2.1 <sup>a</sup> | 50                        | 19                    | 68.3 <sup>a</sup> | 0.7                                     | 2.0 <sup>a</sup> |
| AMF                      | 73                           | 18                    | 96.7 <sup>b</sup> | 0.7                                     | 2.8 <sup>b</sup> | 69                               | 16                    | 90.4 <sup>b</sup> | 0.7                                     | 2.7 <sup>b</sup> | 57                        | 17                    | 85.4 <sup>b</sup> | 0.6                                     | 2.9 <sup>b</sup> |
| <i>Azotobacter</i>       | 74                           | 19                    | 95.3 <sup>b</sup> | 0.7                                     | 2.7 <sup>b</sup> | 70                               | 17                    | 89.3 <sup>b</sup> | 0.7                                     | 2.7 <sup>b</sup> | 59                        | 18                    | 85.0 <sup>b</sup> | 0.7                                     | 2.8 <sup>b</sup> |
| AMF + <i>Azotobacter</i> | 76                           | 18                    | 109 <sup>c</sup>  | 0.8                                     | 3.1 <sup>c</sup> | 73                               | 17                    | 103 <sup>c</sup>  | 0.8                                     | 3.5 <sup>c</sup> | 60                        | 18                    | 99.5 <sup>c</sup> | 0.8                                     | 3.7 <sup>c</sup> |

same letter within each column indicates no significant difference among treatments ( $p < 0.05$ ) as determined by DMRT. value are means of  $n = 4$ . S: Survival; A: during transplantation (July, 2012); B: after two years, in July, 2014

**Table 6 – CRDT.** Effect of different biofertilizer treatments on survival % and growth parameters of tree sp. at Project site, Farrukhnagar, Haryana (after two years)

| Treatments               | Ber ( <i>Z. mauritiana</i> ) |                       |                   |                                         |                  | Kinnow ( <i>Citrus nobilis</i> ) |                       |                   |                                         |                  | Lemon ( <i>C. limon</i> ) |                       |                   |                                         |                  |
|--------------------------|------------------------------|-----------------------|-------------------|-----------------------------------------|------------------|----------------------------------|-----------------------|-------------------|-----------------------------------------|------------------|---------------------------|-----------------------|-------------------|-----------------------------------------|------------------|
|                          | S %                          | Avg shoot length (cm) |                   | Avg girth (cm) (30 cm above the ground) |                  | S %                              | Avg shoot length (cm) |                   | Avg girth (cm) (30 cm above the ground) |                  | S %                       | Avg shoot length (cm) |                   | Avg girth (cm) (30 cm above the ground) |                  |
|                          |                              |                       |                   |                                         |                  |                                  |                       |                   |                                         |                  |                           |                       |                   |                                         |                  |
|                          |                              | A                     | B                 | A                                       | B                |                                  | A                     | B                 | A                                       | B                |                           | A                     | B                 | A                                       | B                |
| Control                  | 60                           | 18                    | 79.2 <sup>a</sup> | 0.7                                     | 2.4 <sup>a</sup> | 50                               | 17                    | 76.4 <sup>a</sup> | 0.8                                     | 2.5 <sup>a</sup> | 48                        | 18                    | 73.4 <sup>a</sup> | 0.6                                     | 2.4 <sup>a</sup> |
| AMF                      | 70                           | 18                    | 96.7 <sup>b</sup> | 0.8                                     | 3.8 <sup>b</sup> | 65                               | 16                    | 94.3 <sup>b</sup> | 0.7                                     | 3.2 <sup>b</sup> | 55                        | 17                    | 85.6 <sup>b</sup> | 0.7                                     | 3.4 <sup>b</sup> |
| <i>Azotobacter</i>       | 72                           | 19                    | 96 <sup>b</sup>   | 0.8                                     | 3.6 <sup>b</sup> | 70                               | 16                    | 93.1 <sup>b</sup> | 0.7                                     | 3.0 <sup>b</sup> | 57                        | 17                    | 85 <sup>b</sup>   | 0.8                                     | 3.2 <sup>b</sup> |
| AMF + <i>Azotobacter</i> | 72                           | 18                    | 125 <sup>c</sup>  | 0.8                                     | 4.2 <sup>c</sup> | 71                               | 16                    | 118 <sup>c</sup>  | 0.8                                     | 3.6 <sup>c</sup> | 60                        | 19                    | 99.8 <sup>c</sup> | 0.7                                     | 3.7 <sup>c</sup> |

same letter within each column indicates no significant difference among treatments ( $p < 0.05$ ) as determined by DMRT. value are means of  $n = 4$ . S: Survival; A: during transplantation (July, 2012); B: after two years, in July, 2014

**Table 7 – CRDT.** Chlorophyll, fibre and mineral contents of *Cenchrus setigerus* (cc337) grass in response to biofertilizers in silvipastoral system (Micromodel, IIT Delhi and Farrukhnagar, Haryana) after two years (*I year: May 2012-May 2013; II Year: May 2013-May 2014*)

| Parameters                 | Silvipastoral Model at Micromodel, IIT Delhi |           |           |           | Silvipastoral Model developed at Field (Vill. Farrukhnagar, Haryana) |           |           |           |
|----------------------------|----------------------------------------------|-----------|-----------|-----------|----------------------------------------------------------------------|-----------|-----------|-----------|
|                            | Control                                      | AMF       | Az        | AMF + Az  | Control                                                              | AMF       | Az        | AMF + Az  |
| <b>Chlorophyll content</b> |                                              |           |           |           |                                                                      |           |           |           |
| Chl a (mg/g)               | 2.45±0.02                                    | 2.56±0.01 | 2.57±0.1  | 2.6±0.02  | 2.40±0.01                                                            | 2.47±0.02 | 2.47±0.02 | 2.56±0.01 |
| Total Chl (mg/g)           | 2.9±0.04                                     | 3.0±0.02  | 3.01±0.2  | 3.11±0.02 | 2.8±0.02                                                             | 2.95±0.02 | 2.98±0.03 | 3.08±0.01 |
| <b>Fibre content</b>       |                                              |           |           |           |                                                                      |           |           |           |
| Cellulose(%)               | 35.4±0.2                                     | 37.2±0.2  | 35.6±0.2  | 36.4±0.2  | 33.8±0.33                                                            | 34.6±0.3  | 34.2±0.4  | 34.9±0.1  |
| ADF (%)                    | 46.3±0.3                                     | 44.3±0.2  | 43.9±0.3  | 42.9±0.2  | 49.8±0.19                                                            | 48.3±0.3  | 48.2±0.3  | 47.3±0.2  |
| NDF (%)                    | 58.2±0.2                                     | 54.6±0.3  | 54.2±0.4  | 50.2±0.   | 61.1±0.25                                                            | 60.1±0.5  | 60.0±0.3  | 59.3±0.2  |
| <b>Mineral content</b>     |                                              |           |           |           |                                                                      |           |           |           |
| N (%)                      | 1.28±0.01                                    | 1.27±0.02 | 1.26±0.03 | 1.49±0.01 | 0.91±0.01                                                            | 1.18±0.01 | 1.19±0.01 | 1.47±0.01 |
| P (%)                      | 0.5±0.04                                     | 0.61±0.01 | 0.54±0.02 | 0.67±0.02 | 0.45±0.02                                                            | 0.48±0.02 | 0.47±0.02 | 0.56±0.01 |
| K (%)                      | 1.35±0.02                                    | 1.42±0.01 | 1.43±0.02 | 1.48±0.02 | 1.29±0.02                                                            | 1.37±0.01 | 1.38±0.01 | 1.41±0.02 |
| Ca (mg/Kg)                 | 3.18±0.03                                    | 3.21±0.02 | 3.2±0.01  | 3.27±0.01 | 3.1±0.01                                                             | 3.18±0.01 | 3.17±0.02 | 3.21±0.02 |
| Mg (mg/Kg)                 | 3.35±0.03                                    | 3.4±0.02  | 3.39±0.01 | 3.45±0.01 | 3.29±0.02                                                            | 3.37±0.02 | 3.38±0.02 | 3.41±0.02 |

**Table 8 - CRDT** Chlorophyll, fibre and mineral contents of *Cenchrus setigerus* (cc58) grass in response to biofertilizers in silvipastoral system (Micromodel, IIT Delhi and Farrukhnagar, Haryana) after two years (*I year: May 2012-May 2013; II Year: May 2013-May 2014*)

| Parameter s                | Silvipastoral Model at Micromodel, IIT Delhi |           |           |           | Silvipastoral Model developed at Field (Vill. Farrukhnagar, Haryana) |           |           |           |
|----------------------------|----------------------------------------------|-----------|-----------|-----------|----------------------------------------------------------------------|-----------|-----------|-----------|
|                            | Control                                      | AMF       | Az        | AMF + Az  | Control                                                              | AMF       | Az        | AMF + Az  |
| <b>Chlorophyll content</b> |                                              |           |           |           |                                                                      |           |           |           |
| Chl a (mg/g)               | 2.65±0.02                                    | 2.69±0.02 | 2.7±0.02  | 2.75±0.03 | 2.59±0.03                                                            | 2.61±0.03 | 2.62±0.04 | 2.67±0.02 |
| Total Chl (mg/g)           | 3.1±0.02                                     | 3.14±0.02 | 3.15±0.01 | 3.19±0.02 | 3.0±0.02                                                             | 3.08±0.03 | 3.08±0.04 | 3.12±0.03 |
| <b>Fibre content</b>       |                                              |           |           |           |                                                                      |           |           |           |
| Cellulose(%)               | 36.8±0.1                                     | 37.7±0.2  | 37.6±0.1  | 38.1±0.2  | 34.9±0.3                                                             | 35.8±0.4  | 35.8±0.3  | 36.9±0.4  |
| ADF (%)                    | 47.1±0.2                                     | 46.4±0.2  | 46.1±0.3  | 45.0±0.1  | 50.1±0.4                                                             | 49.1±0.5  | 49.7±0.2  | 48.0±0.5  |
| NDF (%)                    | 59.2±0.1                                     | 55.3±0.2  | 54.9±0.2  | 51.4±0.2  | 62.1±0.5                                                             | 59.2±0.4  | 59.9±0.2  | 58.1±0.5  |
| <b>Mineral content</b>     |                                              |           |           |           |                                                                      |           |           |           |
| N (%)                      | 1.26±0.03                                    | 1.28±0.02 | 1.29±0.02 | 1.45±0.02 | 1.07±0.01                                                            | 1.18±0.02 | 1.2±0.03  | 1.3±0.04  |
| P (%)                      | 0.5±0.02                                     | 0.62±0.01 | 0.62±0.02 | 0.69±0.03 | 0.47±0.02                                                            | 0.52±0.02 | 0.52±0.03 | 0.58±0.03 |
| K (%)                      | 1.38±0.01                                    | 1.46±0.01 | 1.47±0.01 | 1.5±0.03  | 1.29±0.01                                                            | 1.35±0.02 | 1.36±0.02 | 1.41±0.02 |
| Ca                         | 3.2±0.02                                     | 3.26±0.0  | 3.26±0.0  | 3.31±0.0  | 3.09±0.0                                                             | 3.14±0.0  | 3.15±0.0  | 3.2±0.02  |

|         |          |          |          |          |          |          |          |          |
|---------|----------|----------|----------|----------|----------|----------|----------|----------|
| (mg/Kg) |          | 3        | 1        | 2        | 2        | 3        | 2        |          |
| Mg      | 3.41±0.0 | 3.47±0.0 | 3.48±0.0 | 3.52±0.0 | 3.31±0.0 | 3.39±0.0 | 3.4±0.01 | 3.42±0.0 |
| (mg/Kg) | 2        | 2        | 2        | 2        | 2        | 3        |          | 1        |

**Table 9 – CRDT.** Chlorophyll, fibre and mineral contents of *P. maximum* grass in response to biofertilizers in silvipastoral system (Micromodel, IIT Delhi and Farrukhnagar, Haryana) after two years (I year: May 2012-May 2013; II Year: May 2013-May 2014)

| Parameters                 | Silvipastoral Model at Micromodel, IIT Delhi |          |          |          | Silvipastoral Model developed at Field (Vill. Farrukhnagar, Haryana) |          |          |           |
|----------------------------|----------------------------------------------|----------|----------|----------|----------------------------------------------------------------------|----------|----------|-----------|
|                            | Control                                      | AMF      | Az       | AMF + Az | Control                                                              | AMF      | Az       | AMF + Az  |
| <b>Chlorophyll content</b> |                                              |          |          |          |                                                                      |          |          |           |
| Chl a (mg/g)               | 2.4±0.02                                     | 2.46±0.2 | 2.43±0.0 | 2.51±0.0 | 2.35±0.0                                                             | 2.47±0.0 | 2.36±0.0 | 2.45±0.0  |
|                            |                                              |          | 4        | 5        | 3                                                                    | 4        | 2        | 3         |
| Total Chl (mg/g)           | 3.04±0.0                                     | 3.15±0.1 | 3.10±0.0 | 3.28±0.0 | 2.76±0.0                                                             | 2.95±0.0 | 2.91±0.0 | 3.1±0.03  |
|                            | 3                                            |          | 3        | 1        | 4                                                                    | 1        | 2        |           |
| <b>Fibre content</b>       |                                              |          |          |          |                                                                      |          |          |           |
| Cellulose (%)              | 30.1±0.1                                     | 31.5±0.2 | 30.7±0.3 | 32.1±0.3 | 29.4±0.2                                                             | 30.0±0.1 | 29.8±0.2 | 30.26±0.1 |
| ADF (%)                    | 34.3±0.1                                     | 33.6±0.1 | 33.3±0.3 | 32.7±0.1 | 35.3±0.3                                                             | 34.6±0.3 | 34.6±0.3 | 34.2±0.2  |
| NDF (%)                    | 62.5±0.2                                     | 60.9±0.3 | 60.6±0.2 | 59.6±0.2 | 63.2±0.3                                                             | 62.5±0.4 | 62.4±0.2 | 62.4±0.4  |
| <b>Mineral content</b>     |                                              |          |          |          |                                                                      |          |          |           |
| N (%)                      | 1.72±0.0                                     | 1.79±0.0 | 1.85±0.0 | 2.4±0.02 | 1.6±0.01                                                             | 1.72±0.0 | 1.8±0.01 | 2.2±0.04  |
|                            | 2                                            | 2        | 1        |          |                                                                      | 3        |          |           |
| P (%)                      | 0.57±0.0                                     | 0.67±0.0 | 0.62±0.0 | 0.71±0.0 | 0.49±0.0                                                             | 0.54±0.0 | 0.52±0.0 | 0.61±0.0  |
|                            | 3                                            | 3        | 1        | 2        | 2                                                                    | 2        | 3        | 3         |
| K (%)                      | 1.38±0.0                                     | 1.45±0.0 | 1.45±0.0 | 1.5±0.03 | 1.31±0.0                                                             | 1.37±0.0 | 1.35±0.0 | 1.39±0.0  |
|                            | 3                                            | 1        | 2        |          | 1                                                                    | 1        | 3        | 3         |
| Ca (mg/Kg)                 | 3.43±0.0                                     | 3.48±0.0 | 3.52±0.0 | 3.6±0.03 | 3.36±0.0                                                             | 3.38±0.0 | 3.37±0.0 | 3.42±0.0  |
|                            | 2                                            | 2        | 1        |          | 1                                                                    | 1        | 1        | 1         |
| Mg (mg/Kg)                 | 3.54±0.0                                     | 3.6±0.01 | 3.61±0.0 | 3.65±0.0 | 3.47±0.0                                                             | 3.52±0.0 | 3.5±0.01 | 3.57±0.0  |
|                            | 2                                            |          | 1        | 1        | 1                                                                    | 1        |          | 1         |

**Table 10 – CRDT.** Chlorophyll, fibre and mineral contents of *P. pedicellatum* grass in response to biofertilizers in silvipastoral system (Micromodel, IIT Delhi and Farrukhnagar, Haryana) after two years (I year: May 2012-May 2013; II Year: May 2013-May 2014)

| Parameters                 | Silvipastoral Model at Micromodel, IIT Delhi |          |          |          | Silvipastoral Model developed at Field (Vill. Farrukhnagar, Haryana) |          |          |          |
|----------------------------|----------------------------------------------|----------|----------|----------|----------------------------------------------------------------------|----------|----------|----------|
|                            | Control                                      | AMF      | Az       | AMF + Az | Control                                                              | AMF      | Az       | AMF + Az |
| <b>Chlorophyll content</b> |                                              |          |          |          |                                                                      |          |          |          |
| Chl a (mg/g)               | 2.7±0.03                                     | 2.78±0.0 | 2.72±0.0 | 2.85±0.0 | 2.55±0.0                                                             | 2.62±0.0 | 2.61±0.0 | 2.71±0.0 |
|                            |                                              | 1        | 4        | 3        | 3                                                                    | 1        | 2        | 3        |
| Total Chl (mg/g)           | 3.18±0.0                                     | 3.31±0.0 | 3.26±0.0 | 3.38±0.0 | 3.08±0.0                                                             | 3.11±0.0 | 3.1±0.02 | 3.17±0.0 |
|                            | 2                                            | 2        | 2        | 1        | 3                                                                    | 1        |          | 3        |

| <b>Fibre content</b>   |           |           |           |           |           |           |           |           |
|------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| Cellulose(%)           | 32.15±0.3 | 32.9±0.1  | 32.56±0.2 | 33.2±0.1  | 31.2±0.2  | 32.12±0.3 | 31.89±0.3 | 32.7±0.2  |
| ADF (%)                | 41.2±0.2  | 37.2±0.2  | 38.3±0.3  | 36.3±0.2  | 43.2±0.2  | 41.1±0.2  | 41.2±0.2  | 40.2±0.1  |
| NDF (%)                | 55.7±0.3  | 54.2±0.3  | 54.2±0.1  | 52.2±0.2  | 56.3±0.1  | 55.9±0.25 | 56.2±0.2  | 55.2±0.1  |
| <b>Mineral content</b> |           |           |           |           |           |           |           |           |
| N (%)                  | 0.8±0.01  | 1.11±0.01 | 1.28±0.02 | 1.57±0.03 | 0.74±0.04 | 1.02±0.02 | 1.18±0.04 | 1.49±0.03 |
| P (%)                  | 0.41±0.03 | 0.47±0.02 | 0.48±0.02 | 0.54±0.03 | 0.36±0.02 | 0.35±0.03 | 0.4±0.03  | 0.5±0.04  |
| K (%)                  | 1.09±0.04 | 1.1±0.02  | 1.11±0.01 | 1.17±0.02 | 0.97±0.01 | 1.08±0.02 | 1.1±0.03  | 1.16±0.03 |
| Ca (mg/Kg)             | 3.08±0.01 | 3.11±0.01 | 3.1±0.01  | 3.18±0.02 | 2.98±0.02 | 3.08±0.02 | 3.07±0.01 | 3.16±0.02 |
| Mg (mg/Kg)             | 3.21±0.01 | 3.27±0.01 | 3.26±0.02 | 3.3±0.01  | 3.1±0.02  | 3.19±0.02 | 3.19±0.01 | 3.22±0.02 |

**Table 11 - CRDT.** Chlorophyll, fibre and mineral contents of *P. virgatum* grass in response to biofertilizers at Micromodel, IIT Delhi

| <b>Parameters</b>          | <b>Silvipastoral Model at Micromodel, IIT Delhi</b> |            |            |                 |
|----------------------------|-----------------------------------------------------|------------|------------|-----------------|
|                            | <b>Control</b>                                      | <b>AMF</b> | <b>Az</b>  | <b>AMF + Az</b> |
| <b>Chlorophyll content</b> |                                                     |            |            |                 |
| Chl a (mg/g)               | 2.8±0.4                                             | 2.95±0.3   | 2.97±0.25  | 3.01±0.2        |
| Total Chl (mg/g)           | 3.32±0.35                                           | 3.4±0.2    | 3.43±0.4   | 3.5±0.1         |
| <b>Fibre content</b>       |                                                     |            |            |                 |
| Cellulose(%)               | 36.5±0.29                                           | 37.9±0.15  | 37.56±0.21 | 38.1±0.1        |
| ADF (%)                    | 43.14±0.18                                          | 41.78±0.24 | 41.9±0.34  | 38.7±0.2        |
| NDF (%)                    | 76.7±0.35                                           | 72.3±0.31  | 73.5±0.11  | 68.6±0.1        |
| <b>Mineral Content</b>     |                                                     |            |            |                 |
| N (%)                      | 0.46±0.02                                           | 0.54±0.02  | 0.6±0.01   | 0.7±0.02        |
| P (%)                      | 0.57±0.04                                           | 0.67±0.01  | 0.61±0.02  | 0.74±0.03       |
| K (%)                      | 1.5±0.03                                            | 1.75±0.01  | 1.9±0.02   | 2.01±0.02       |
| Ca (mg/Kg)                 | 6922±0.02                                           | 7144±0.02  | 7168±0.01  | 8010±0.01       |
| Mg (mg/Kg)                 | 2626±0.02                                           | 2658±0.01  | 2670±0.01  | 2695±0.01       |

**Table 12 – CRDT.** Pre analysis of soil from project site (different villages) and Micromodel IIT Delhi

| <b>Sites (Villages)</b> | <b>EC (mmhos/cm)</b> | <b>pH</b> | <b>N (Kg/ha)</b> | <b>P (Kg/ha)</b> | <b>K (Kg/ha)</b> | <b>C%</b> |
|-------------------------|----------------------|-----------|------------------|------------------|------------------|-----------|
| <b>Kheda</b>            | 2.1±0.5              | 8.5±0.3   | 320±2.1          | 5.5±0.02         | 253±3.1          | 0.34±0.01 |
| <b>Bawda</b>            | 1.9±0.6              | 8.7±0.3   | 318±1.6          | 6.1±0.01         | 256±2.4          | 0.41±0.01 |
| <b>Patli</b>            | 1.9±0.5              | 8.3±0.2   | 310±2.0          | 6.0±0.01         | 241±2.0          | 0.36±0.02 |
| <b>Jodala</b>           | 2.2±0.4              | 8.5±0.3   | 291±1.1          | 6.3±0.03         | 210±1.5          | 0.33±0.03 |
| <b>MicroModel, IITD</b> | 0.15±0.1             | 7.0±0.2   | 123±1.2          | 8.9±0.04         | 239±1.5          | 0.51±0.02 |

**Table 13. CRDT.** Comparative microbial analysis of soil of Micromodel, IIT Delhi and selected villages at project site, Block Farrukhnagar, Haryana (pre analysis)

| Site (villages)  | <i>Azotobacter</i> (CFU, log10) | <i>Azospirillum</i> (CFU, log10) | AMF (spores/g) |
|------------------|---------------------------------|----------------------------------|----------------|
| Kheda            | 3.1±0.2                         | 3.0±0.1                          | 14±0.4         |
| Bawda            | 3.3±0.1                         | 3.3±0.1                          | 13±0.4         |
| Patli            | 3.1±0.1                         | 3.1±0.1                          | 13±0.5         |
| Jodala           | 2.9±0.2                         | 3.4±0.2                          | 12±0.3         |
| Micromodel, IITD | 11.0±1.1                        | 9±0.2                            | 35±1.1         |

**Table 14 – CRDT.** Post analysis of soil from project site (different villages) and Micromodel IIT Delhi

| Sites (Villages)   | EC (mmhos/cm) | pH      | N (Kg/ha) | P (Kg/ha) | K (Kg/ha) | C%        |
|--------------------|---------------|---------|-----------|-----------|-----------|-----------|
| Farrukhnagar**     | 2.24±0.02     | 8.4±0.1 | 326±2.1   | 6.57±0.5  | 269±1.5   | 0.43±0.01 |
| Mubarikpur**       | 2.2±0.03      | 8.1±0.1 | 310±2.1   | 6.11±0.6  | 256±2.0   | 0.4±0.01  |
| Mevka**            | 2.22±0.02     | 8.0±0.2 | 325±1.5   | 6.4±0.4   | 272±2.2   | 0.5±0.02  |
| Dooma**            | 3.12±0.02     | 8.8±0.2 | 253±1.3   | 5.4±0.4   | 193± 2.5  | 0.37±0.02 |
| Kheda*             | 2.0±0.6       | 8.4±0.3 | 326±1.6   | 6.2±0.01  | 259±2.4   | 0.43±0.01 |
| Bawda*             | 2.0±0.5       | 8.0±0.2 | 318±2.0   | 6.09±0.01 | 245±2.0   | 0.38±0.02 |
| Patli*             | 2.11±0.4      | 8.1±0.3 | 300±1.1   | 6.38±0.03 | 213±1.5   | 0.35±0.03 |
| MicroModel, IITD** | 0.67±0.01     | 7.0±0.1 | 142±1.5   | 9.0±0.7   | 248±2.1   | 0.57±0.02 |

\*after one year

\*\*after two year

**Table 15 – CRDT.** Comparative microbial analysis of soil of Micromodel, IIT Delhi and selected villages at project site, Block Farrukhnagar, Haryana (after one year)

| Site (villages)    | <i>Azotobacter</i> (CFU log10) | <i>Azospirillum</i> (CFU log10) | AMF (spores/g) |
|--------------------|--------------------------------|---------------------------------|----------------|
| Farrukhnagar**     | 6.0±0.4                        | 8.1±0.5                         | 23±0.5         |
| Mubarikpur**       | 4.9 ±0.5                       | 7.9±0.5                         | 27±0.6         |
| Dooma**            | 3.0±0.2                        | 3.1±0.2                         | 16±0.5         |
| Mevka**            | 7.0 ±0.2                       | 8.2±0.5                         | 29±1.5         |
| Kheda*             | 5.5±0.4                        | 7.7±0.5                         | 19±0.5         |
| Bawda*             | 4.4 ±0.5                       | 7.2±0.5                         | 20±0.6         |
| Patli*             | 2.8±0.2                        | 2.64±0.2                        | 13±0.5         |
| Micromodel, IITD** | 11.5±0.8                       | 10.4±0.6                        | 45±1.7         |

\*after one year

\*\*after two year

**Annexure I - CRDT****Training Programs Conducted this year**

|   |                                |                                                                                                                                                                        |    |
|---|--------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----|
| 1 | 28th Oct<br>2013               | Conducted a <b>Training Programme</b> on the benefits of High Yielding Fodder grasses and fodder conservation                                                          | 40 |
| 2 | 16 <sup>th</sup> Dec.<br>2013  | <b>Motivation programme</b> for villagers (Vill. Jodala and Patli) to use biofertilizers and utilizing their marginal lands for fodder grass cultivation               | 20 |
| 3 | 28th Jan.<br>2014              | <b>Taining programme</b> on fodder gass cultivation and use of biofertilizers (Vill. Patli)                                                                            | 30 |
| 4 | 10th March<br>2014             | Meeting Sarpanch, MNREGA motivators and villagers to discuss about sustainability of the project activities and dissemination of the technology to the other villages. | -  |
| 5 | 10th July<br>2014              | <b>Taining programme</b> on Hay and Silage formation at Farrukhnagar                                                                                                   | 15 |
| 6 | 24 <sup>th</sup> July.<br>2014 | <b>Taining programme</b> on Silage formation at Vill. Farrukhnagar                                                                                                     | 30 |
| 7 | 29th Aug.<br>2014              | <b>Taining programme</b> on Silage formation at Vill. Jodala                                                                                                           | 50 |
| 8 | 9th Sept.<br>2014              | <b>Taining programme</b> on Silage formation at Vill. Mubarikpur                                                                                                       | 10 |

**Glimpses of some training and interaction programmes with farmers:****(A)****(B)****(A) & (B) Visit to Farmer's Field at Vill. Mubarikpur**



**C) Silage Making Training Programme  
Farrukhnagar programme on Hay and Silage**

**D) Awareness generation cum training at Vill.**



**E) Meeting with Village Sarpanch and MNREGA Activators**

## **Annexure II - CRDT**

### **Quantity and Type of Grass provided to the Farmers this year (2013-2014)**

| <b>S. No</b> | <b>Name of beneficiary</b>         | <b>Village</b> | <b>Seeds provided</b>                     | <b>Quantity</b>       |
|--------------|------------------------------------|----------------|-------------------------------------------|-----------------------|
| 1            | Gopal                              | ---- do----    | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |
| 2            | Kudia Ram                          | ---- do----    | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |
| 3            | Lali Devi                          | ---- do----    | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |
| 4            | Tarachand Yadav                    | Joniawas       | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |
| 5            | Dharmapal Singh                    | Musedpur       | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |
| 6            | Roop Chand                         | Musedpur       | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |
| 7            | Man Singh, Chairman,<br>Kisan club | Dawoda         | Para grass<br>Karnal grass                | 100 root slips each   |
| 8            | Santosh devi                       | ---- do----    | <i>P. maximum,</i><br><i>pedicillatum</i> | <i>P.</i> 0.2 Kg each |
| 9            | Mukesh                             | ---- do----    | <i>P. maximum,</i><br><i>pedicillatum</i> | <i>P.</i> 0.2 Kg each |
| 10           | Hans Raj                           | ---- do----    | <i>P. maximum,</i><br><i>pedicillatum</i> | <i>P.</i> 0.2 Kg each |
| 11           | Fatehsingh                         | ---- do----    | <i>P. maximum</i><br>Napier grass         | 100 root slips each   |

**Annexure III - CRDT**
**Grass seeds distribution**

| S.No | Village      | No. Of farmers involved | Types pf grass seed distributed | Yield obtained (ton/ha/year) | pH  |     |
|------|--------------|-------------------------|---------------------------------|------------------------------|-----|-----|
|      |              |                         |                                 |                              | A   | B   |
| 1    | Mubarikpur   | 7                       | Gunnia grass                    | 24-28                        | 8.7 | 8.1 |
|      |              |                         | Cenchrus                        | 12-20                        |     |     |
|      |              |                         | Dinanath                        | 15-24                        |     |     |
| 2    | Meoka        | 5                       | Gunnia grass                    | 25-30                        | 8.5 | 8.0 |
|      |              |                         | Cenchrus                        | 17-20                        |     |     |
|      |              |                         | Dinanath                        | 22-27                        |     |     |
|      |              |                         | Napier bajra                    | 30-35                        |     |     |
| 3    | Dooma        | 6                       | Gunnia grass                    | Not established              | 9.0 | 8.8 |
|      |              |                         | Cenchrus                        | ---do---                     |     |     |
|      |              |                         | Dinanath                        | ---do---                     |     |     |
|      |              |                         | Napier bajra                    | ---do---                     |     |     |
|      |              |                         | Para grass                      | 4-5                          |     |     |
|      |              |                         | Karnal grass                    | Not established              |     |     |
| 4    | Farrukhnagar | 5                       | Cenchrus                        | 15-18                        | 9.0 | 8.4 |
|      |              |                         | Gunnia grass                    | 24-29                        |     |     |
|      |              |                         | Dinanath                        | 18-25                        |     |     |
|      |              |                         | Para grass                      | Not established              |     |     |
|      |              |                         | Karnal grass                    | ---do---                     |     |     |
| 5    | Patli        | 8                       | Gunnia grass                    | 15-19                        | 8.5 | 8.1 |
|      |              |                         | Cenchrus                        | 10-15                        |     |     |
|      |              |                         | Dinanath                        | 12-17                        |     |     |
|      |              |                         | Napier bajra                    | 20-30                        |     |     |
| 6    | Kheda        | 3                       | Gunnia grass                    | 16-20                        | 9.0 | 8.4 |
|      |              |                         | Dinanath                        | Not established              |     |     |
|      |              |                         | Para grass                      | ---do---                     |     |     |
|      |              |                         | Karnal grass                    | ---do---                     |     |     |
|      |              |                         | Napier grass                    | 20-27                        |     |     |
| 7    | Dawoda       | 1                       | Para grass                      | Not established              | 8.9 | 8.4 |
|      |              |                         | Karnal grass                    | ---do--                      |     |     |
|      |              |                         | Napier grass                    | 20-30                        |     |     |
| 8    | Bawda        | 4                       | Gunnia grass                    | 22-25                        | 8.6 | 8.1 |
|      |              |                         | Dinanath                        | ---do---                     |     |     |
|      |              |                         | Napier grass                    | 19-24                        |     |     |
| 9    | Jadola       | 5                       | Gunnia grass                    | 18-25                        | 8.6 |     |
|      |              |                         | Napier Grass                    | 20-29                        |     |     |
| 10   | Musedpur     | 2                       | Gunnia grass                    | 19-24                        | 8.4 |     |
|      |              |                         | Napier Grass                    | 21-30                        |     |     |
| 11   | Joniawas     | 1                       | Gunnia grass                    | 18-25                        | 8.3 |     |
|      |              |                         | Napier Grass                    | 20-287                       |     |     |

**A: Before sowing the grass**

**B: After one year of grass establishment**

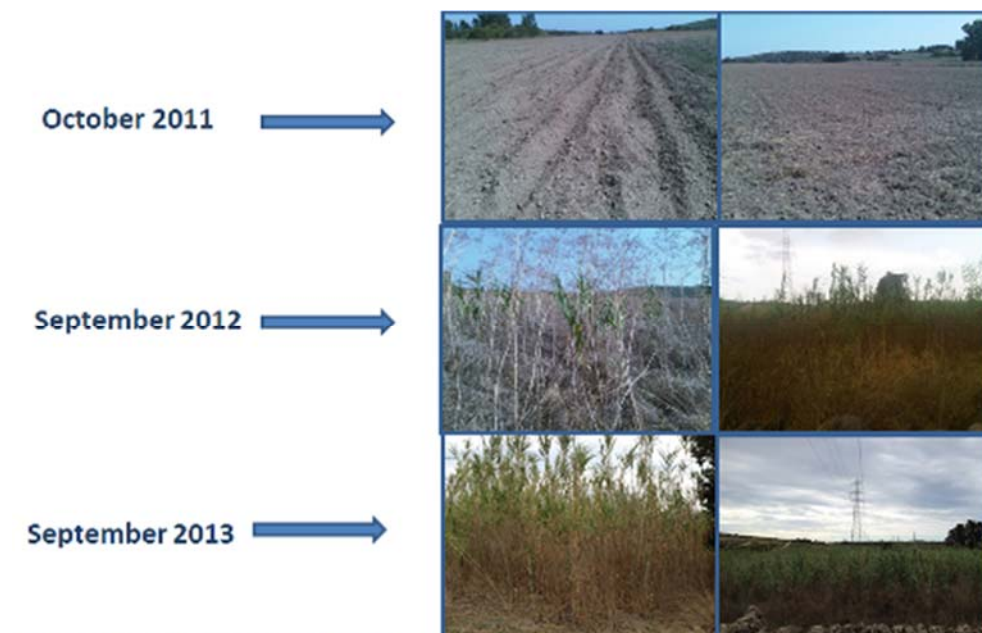
- pH of Vill. Jadola, Musedpur and Joniawas will be given after the completion of one year of grass establishment in these villages

## Task 4.2 Productivity in marginal large fields

University of Catania – UNICT (n. 1)



**Picture 1 - UNICT.** Experimental field location in Sicily, Italy. In the red marked circle in the middle of Sicily, 320 m<sup>2</sup> plot is shown, while in the South-East side the 5 ha field.





**Picture 2 - UNICT.** From field bed preparation (autumn 2011) up to last harvest time (February 2014).

**Table 1 - UNICT.** Biometric and productive measurements of 5 ha giant reed plantation (first harvest, February 2013)

| Measurement                   | Unit               | value  |
|-------------------------------|--------------------|--------|
| # of emerged plants           | m <sup>2</sup>     | 4      |
| uniformity after transplant   | visual score       | medium |
| height of stems               | cm                 | 173    |
| Dry stems                     | %                  | 68     |
| Dry leaves                    | %                  | 32     |
| Aboveground dry biomass yield | t ha <sup>-1</sup> | 1.8    |

**Table 2 - UNICT.** Biometric and productive measurements of 5 ha giant reed plantation (second harvest, February 2014)

| Measurement                   | Unit               | value       |
|-------------------------------|--------------------|-------------|
| # of emerged plants           | m <sup>2</sup>     | 15.3±4.1    |
| uniformity after transplant   | visual score       | Medium-high |
| height of stems               | cm                 | 198.4±79.1  |
| Dry stems                     | %                  | 84.1±13.3   |
| Dry leaves                    | %                  | 12.1±9.1    |
| Aboveground dry biomass yield | t ha <sup>-1</sup> | 8.4±2.1     |

**Table 3 - UNICT.** Biometric and productive measurements of 5 ha giant reed plantation (third harvest, February 2015).

| Measurement                   | Unit               | Value       |
|-------------------------------|--------------------|-------------|
| # of emerged plants           | m <sup>2</sup>     | 19.7±4.3    |
| uniformity after transplant   | visual score       | Medium-high |
| height of stems               | Cm                 | 223.5±82.1  |
| Dry stems                     | %                  | 85.1±12.3   |
| Dry leaves                    | %                  | 13.1±8.7    |
| Aboveground dry biomass yield | t ha <sup>-1</sup> | 12.1±2.9    |



**Picture 3 - UNICT.** Plots of 320 m<sup>2</sup> each in the Center of Sicily (550 m a.s.l, 37° 21'N, 14°16'E). Up on the right giant reed (*Arundo donax*), up on the left *Saccharum spontaneum*.

**Table 2 – UNICT.** Biometric and productive characters of giant reed and *Saccharum spontaneum* after the harvest of 2013.

|                           | Measurement unit   | <i>Arundo</i> | <i>Saccharum</i> |
|---------------------------|--------------------|---------------|------------------|
| Stem numbers              | n.                 | 7.2±1.0       | 1.9±1.1          |
| Weigth of 1 stem          | g                  | 261.6±45.0    | 62.2±14.4        |
| Basal stem diameter       | mm                 | 18.1±1.2      | 9.4±0.3          |
| Plant height              | cm                 | 237.0±27.7    | 91.3±11.4        |
| Stem fresh weigth         | g                  | 230.5±42.2    | 47.6±12.1        |
| Leaf fresh weigth         | g                  | 31.1±2.8      | 14.6±2.4         |
| Stem                      | %                  | 87.7±1.5      | 74.9±2.4         |
| Leaf                      | %                  | 12.3±1.5      | 25.1±2.4         |
| H2O stems                 | %                  | 49.1±1.5      | 64.4±1.9         |
| H2O leaves                | %                  | 18.8±0.9      | 42.3±6.1         |
| Stems dry matter          | %                  | 50.9±1.5      | 35.6±1.9         |
| Leaves dry matter         | %                  | 81.2±0.9      | 57.7±6.1         |
| Fresh biomass yield       | t ha <sup>-1</sup> | 16.3±1.6      | 0.8±0.4          |
| Stems fresh biomas yield  | t ha <sup>-1</sup> | 14.3±1.2      | 0.6±0.4          |
| Leaves fresh biomas yield | t ha <sup>-1</sup> | 2.0±0.4       | 0.2±0.1          |
| Stems dry biomass yield   | t ha <sup>-1</sup> | 7.3±0.8       | 0.3±0.2          |
| Leaves dry biomass yield  | t ha <sup>-1</sup> | 1.6±0.3       | 0.1±0.1          |
| Dry biomass yield         | t ha <sup>-1</sup> | 8.9±1.0       | 0.4±0.2          |



**Picture 4 - UNICT.** *Miscanthus* establishment (left) and survived plant after drought stress (right).



**Picture 5 - UNICT.** Giant reed (left) cardoon (center) and saccharum (right) in the in a marginal area with 26-28% slope.

**Table 3 – UNICT.** Biometric and productive characters of giant reed and *Saccharum spontaneum* after the harvest of 2014.

|                     | Measurement unit   | <i>Arundo</i> | <i>Saccharum</i> |
|---------------------|--------------------|---------------|------------------|
| Stem numbers        | n.                 | 6.6±1.2       | 5.4±2.3          |
| Weigth of 1 stem    | g                  | 201.8±89.8    | 65.0±31.6        |
| Basal stem diameter | mm                 | 18.7±3.6      | 8.2±2.3          |
| Plant height        | cm                 | 223.2±38.2    | 94.1±26.4        |
| Stem                | %                  | 87.1±4.0      | 79±5.1           |
| Leaf                | %                  | 12.9±4.0      | 21±5.1           |
| H2O stems           | %                  | 46.0±1.3      | 66.2±1.8         |
| H2O leaves          | %                  | 26.6±14.2     | 45.7±3.6         |
| Dry biomass yield   | t ha <sup>-1</sup> | 7.6±1.3       | 1.3±0.3          |



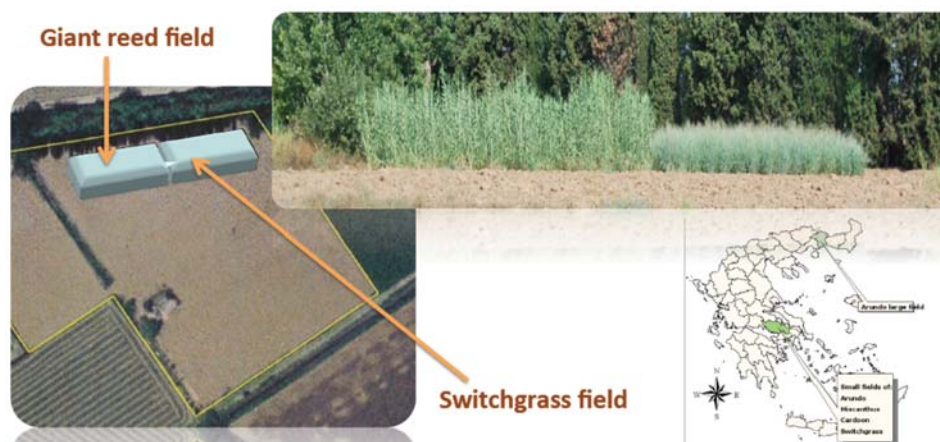
**Picture 6 - UNICT.** Giant reed (left) cardoon (center) and saccharum (right) in the in a marginal area with 26-28% slope during last growing season.

**Table 4 – UNICT.** Morpho-biometric and productive characters of *Arundo donax*, *Saccharum spontaneum* and cardoon in the marginal slope field.

|                   | Measurement<br>unit | <i>Arundo donax</i> | <i>Saccharum<br/>spontaneum</i> | <i>Cynara<br/>cardunculus</i> |
|-------------------|---------------------|---------------------|---------------------------------|-------------------------------|
| Plant height      | Cm                  | 223.2±38.2          | 94.1±26.4                       | 98.6±11.4                     |
| Stems             | %                   | 87.1±4.0            | 79±5.1                          | 36.8±3.2                      |
| Leaves            | %                   | 12.9±4.0            | 21±5.1                          | 40.5±6.1                      |
| Heads             | %                   |                     |                                 | 19.15±5.7                     |
| Hachenes          | %                   |                     |                                 | 3.55±0.41                     |
| H <sub>2</sub> O  | %                   | 45±2.7              | 53±3.2                          | 9.3±1.3                       |
| Dry biomass yield | t ha <sup>-1</sup>  | 8.1±1.5             | 4.3±1.1                         | 5.7±0.9                       |

± standard deviation of three determinations

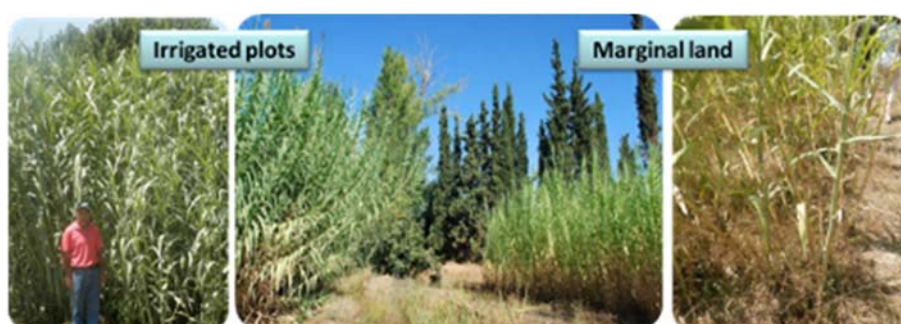
## Center for Renewable Energy Sources and Energy Saving – CRES (n. 3)



**Figure 1 - CRES.** Giant reed and switchgrass fields in Greece

**Table 1 - CRES.** Growth and productivity data of giant reed grown in non-irrigated and irrigated conditions for the first three growing periods

|                                    | Giant reed    |           |               |           |               |           |
|------------------------------------|---------------|-----------|---------------|-----------|---------------|-----------|
|                                    | Non-irrigated | Irrigated | Non-irrigated | Irrigated | Non-irrigated | Irrigated |
| Harvesting date:                   | 01.02.13      | 03.02.13  | 25.01.14      | 25.01.14  | 19.01.15      | 19.01.15  |
| Plant height (cm)                  | 391           | 414       | 400,00        | 506,00    | 540,00        | 616,00    |
| Fresh matter (t ha <sup>-1</sup> ) | 15,4          | 17,4      | 45,00         | 60,00     | 40,50         | 45,50     |
| Dry matter (t ha <sup>-1</sup> )   | 7,14          | 8,5       | 25,65         | 31,20     | 25,52         | 28,00     |
| Moisture content (%)               | 53,64         | 51,15     | 43,00         | 48,00     | 36,99         | 38,46     |
| Stem portion (% on f.m)            | 87,5          | 90        | 90,00         | 93,00     | 70,37         | 73,01     |
| Stem portion (% on d.m)            | 80            | 84,8      | 86,0          | 90,0      | 59,6          | 61,4      |



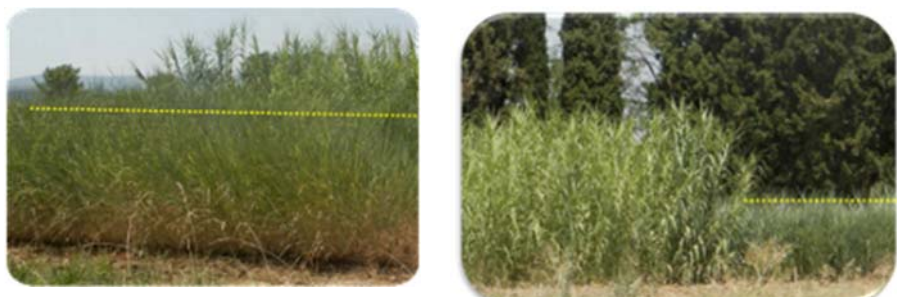
**Figure 2 – CRES.** Giant reed grown in irrigated plots (on the left) and non-irrigated plots (on the right) (19.11.2013)

**Table 2 - CRES.** Growth and productivity data of switchgrass grown in non-irrigated and irrigated conditions for the first three growing periods.

|                                    | Switchgrass   |           |               |           |               |           |
|------------------------------------|---------------|-----------|---------------|-----------|---------------|-----------|
|                                    | Non-irrigated | Irrigated | Non-irrigated | Irrigated | Non-irrigated | Irrigated |
| Harvesting date:                   | 01.02.13      | 01.02.14  | 25.01.14      | 25.01.14  | 19.01.15      | 19.01.15  |
| Plant height (cm)                  | 153           | 151       | 275           | 278       | 186,00        | 294,00    |
| Fresh matter (t ha <sup>-1</sup> ) | 7,73          | 12,47     | 17,5          | 25        | 19,50         | 23,25     |
| Dry matter (t ha <sup>-1</sup> )   | 4,95          | 7,63      | 12,95         | 20,25     | 15,19         | 17,90     |
| Moisture content (%)               | 36,00         | 38,00     | 36,00         | 36,00     | 22,10         | 23,01     |
| Stem portion (% on f.m)            | 68,7          | 70        | 68,7          | 68,7      | 72,87         | 82,37     |
| Stem portion (% on d.m)            | 63,6          | 61        | 63,6          | 63,6      | 69,0          | 61,4      |



**Figure 3 – CRES.** Switchgrass grown in irrigated plots (on the left) and in marginal lands (on the right) (15.07.2013)



**Figure 4 – CRES.** Giant reed (on the left) and switchgrass (on the right) on marginal lands (15.07.2013)

**Istituto per il Sistema Produzione Animale in Ambiente Mediterraneo – CNR-ISPAAAM (n. 8)**

**Table 1 – CNR-ISPAAAM.** Plants, stalks and shoots m<sup>-2</sup> and morphological traits. Plant height is the average height of main stalks. \*=harvested in February 2014; \*\*= harvested in August 2014.

| Species                 | Initial<br>No of<br>plants<br>(No m <sup>-2</sup> ) | Plant<br>height<br>(cm) | No of<br>stalks<br>(No m <sup>-2</sup> ) | No of<br>nodes<br>(No m <sup>-2</sup> ) | Stalk diameter (mm) |        |        |
|-------------------------|-----------------------------------------------------|-------------------------|------------------------------------------|-----------------------------------------|---------------------|--------|--------|
|                         |                                                     |                         |                                          |                                         | Basal               | Medium | Distal |
| <i>A. donax</i> *       | 2                                                   | 301                     | 18                                       | 38.5                                    | 15.6                | 12.0   | 4.6    |
| <i>M. x giganteus</i> * | 2                                                   | 201                     | 14                                       | 14.5                                    | 10.4                | 7.8    | 3.6    |

**Table 2 – CNR-ISPAAAM.** Dry matter yield, dry matter partitioning among organs and dry matter content in giant reed, miscanthus and cardoon. \*=harvested in February 2014; \*\*= harvested in August 2014.

| Species                  | Biomass yield              |                              | Biomass partitioning<br>(% of plant DM) |       |       | Dry matter content<br>(%) |       |       |
|--------------------------|----------------------------|------------------------------|-----------------------------------------|-------|-------|---------------------------|-------|-------|
|                          | DMY<br>per<br>plant<br>(g) | DMY<br>(t ha <sup>-1</sup> ) | Leaves                                  | Stems | Heads | Leaves                    | Stems | Heads |
|                          |                            |                              |                                         |       |       |                           |       |       |
| <i>A. donax</i> *        | 142.7                      | 24.6                         | 21.5                                    | 78.5  | -     | 66                        | 42    | -     |
| <i>M. x giganteus</i> *  | -                          | 8.9                          | 31.5                                    | 68.4  | 0.1   | 73                        | 43    | 76    |
| <i>C. cardunculus</i> ** | 2269.7                     | 23.0                         | 27.5                                    | 39.4  | 33.2  | 79.6                      | 49.5  | 79.6  |



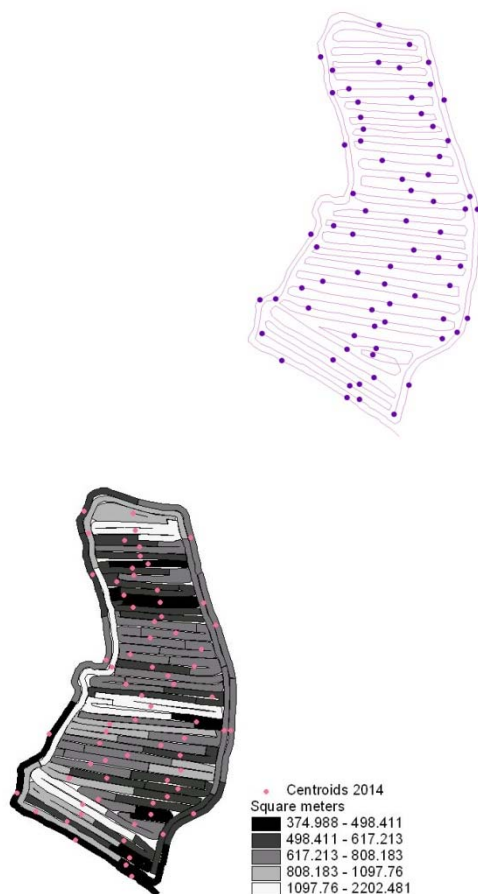
**Picture 1 – CNR-ISPAAAM.** Appearance of *M. x giganteus* (dry plants) at the end of summer 2014 in comparison with *A. donax* and *C. cardunculus* var. *altilis*.

**Table 3 – CNR-ISPAAAM.** Proximate and ultimate analysis of leaves, stalks and heads of *C. cardunculus* var. *altilis*, *A. donax* and *M. x giganteus*.

| Plant component | Calorific value (MJ kg <sup>-1</sup> ) | Volatile matter (%) | Fixed carbon (%) | Ash content (%) | C (%dw) | H (%dw) | N (%dw) | S (%dw) | O (%dw) |
|-----------------|----------------------------------------|---------------------|------------------|-----------------|---------|---------|---------|---------|---------|
| <b>Leaves</b>   |                                        |                     |                  |                 |         |         |         |         |         |
| Cardoon         | 16.0                                   | 85.9                | 4.6              | 20.2            | 41.7    | 5.9     | 2.3     | <0.3    | 48.4    |
| Arundo          | 17.0                                   | 75.0                | 3.9              | 9.1             | 48.0    | 5.9     | 1.9     | 0.8     | 44.2    |
| Miscanthus      | 16.0                                   | 73.0                | 1.7              | 9.5             | 49.0    | 6.3     | 2.7     | <0.3    | 42.0    |
| <b>Stalks</b>   |                                        |                     |                  |                 |         |         |         |         |         |
| Cardoon         | 15.8                                   | 91.6                | 7.1              | 11.9            | 41.0    | 6.2     | 1.5     | <0.3    | 49.2    |
| Arundo          | 18.0                                   | 77.0                | 10.0             | 4.6             | 49.0    | 6.1     | 2.0     | 0.4     | 42.9    |
| Miscanthus      | 17.0                                   | 77.0                | 3.6              | 6.0             | 50.0    | 6.3     | 1.7     | <0.3    | 42.0    |
| <b>Heads</b>    |                                        |                     |                  |                 |         |         |         |         |         |
| Cardoon         | 17.8                                   | 93.5                | 12.0             | 9.2             | 44.5    | 6.7     | 2.5     | <0.3    | 45.0    |



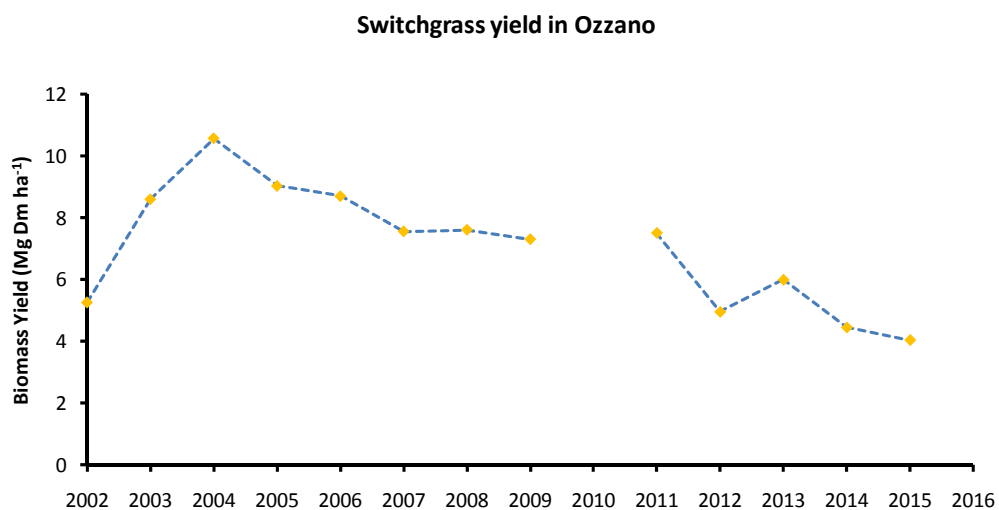
**Figure 1 - UNIBO.** Switchgrass field in Ozzano dell'Emilia. (LEFT) November 2014, just after harvesting and baling. (RIGHT) re-sprouting in April 2015.



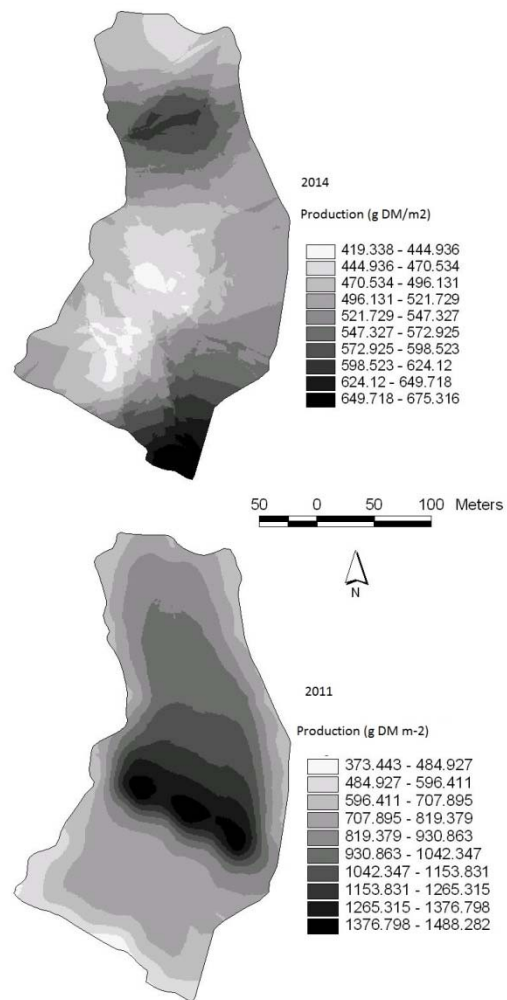
**Figure 2 - UNIBO.** Spatial distribution of bales produced from the 2014 harvest of switchgrass in the Ozzano dell'Emilia field. (LEFT) Bailer tracking (lines) and bale positioning (violet points). (RIGHT) surface (m<sup>2</sup>) from which each bale derived: the higher the surface the lower the productivity of switchgrass on that area.



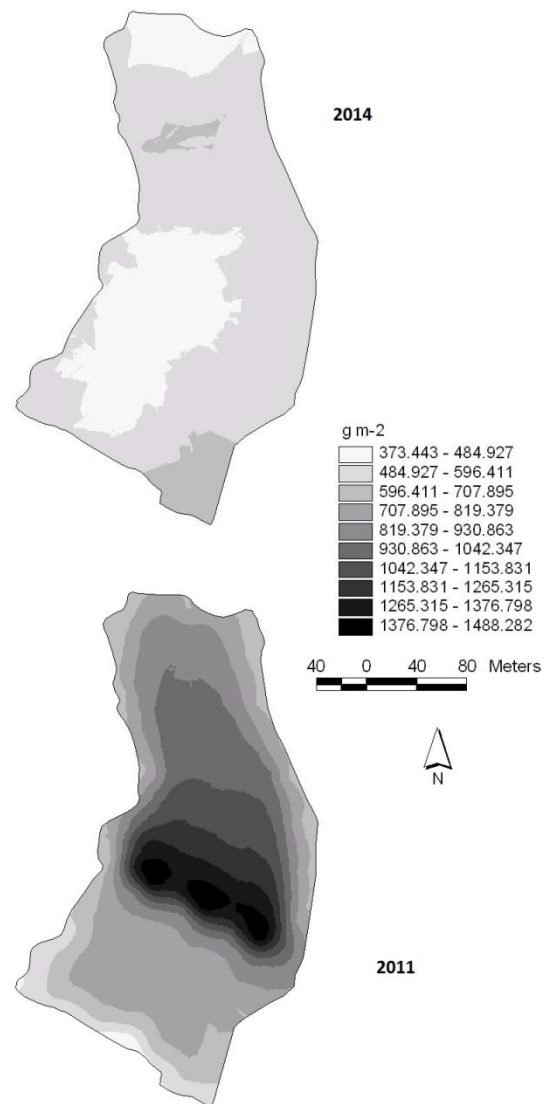
**Figure 3 - UNIBO.** Pictures of hilly field in Ozzano during harvesting in October 2015.



**Figure 4 - UNIBO.** Time course (2002-2015) of switchgrass biomass yield (Mg DM ha<sup>-1</sup>) in the 5-ha field located in Ozzano dell'Emilia (Bologna). Data for 2010 are missing because adverse environmental conditions did not allow to harvest.

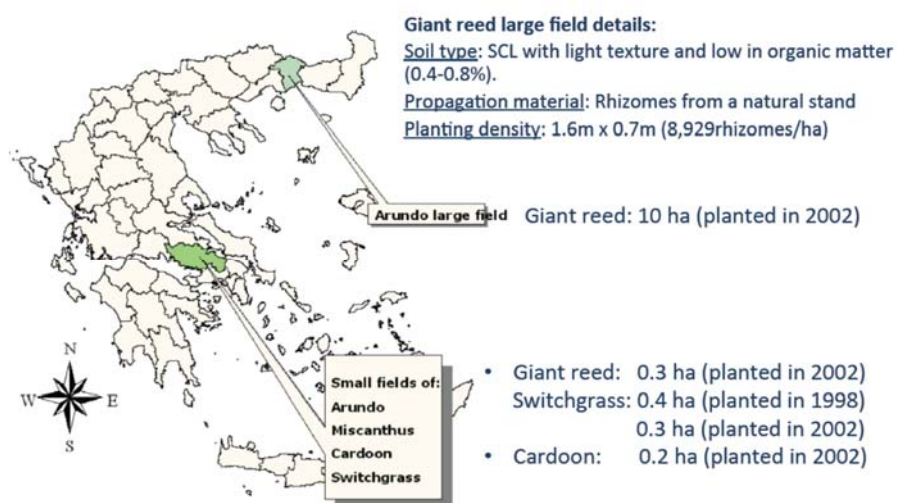


**Figure 5 - UNIBO.** Spatial distribution of switchgrass yield (g DM m<sup>-2</sup>). Above map referred to 2014 harvest; bottom map referred to 2011 harvest.

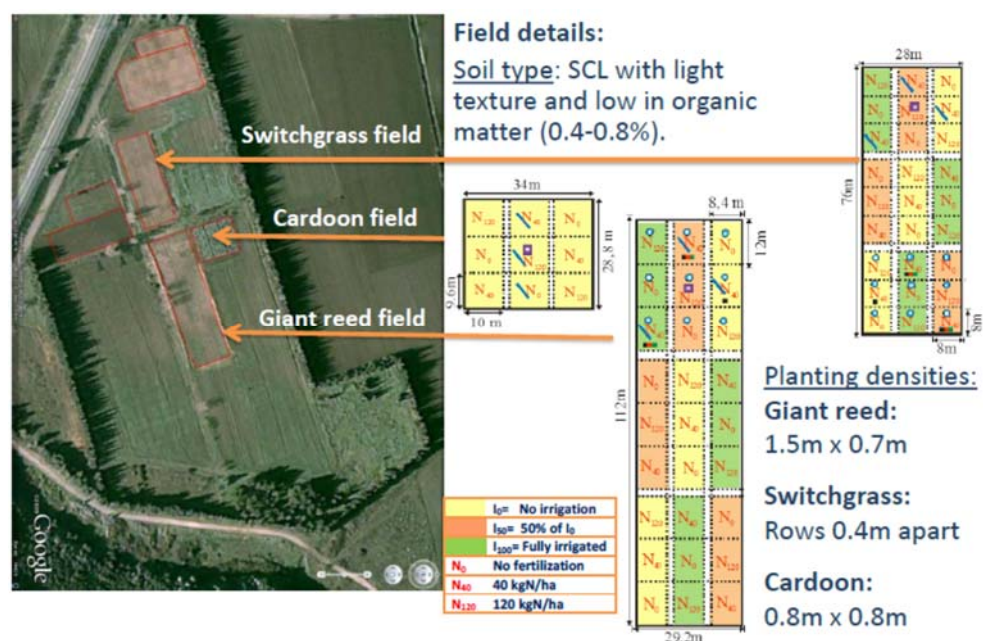


**Figure 6 - UNIBO.** Spatial distribution of switchgrass yield (g DM m<sup>-2</sup>) derived from harvest of 2014 (top map) and 2011 (bottom map) adopting the same scale.

## Task 4.3 Long term productivity of existing large-scale plantation Center for Renewable Energy Sources and Energy Saving – CRES (n. 3)



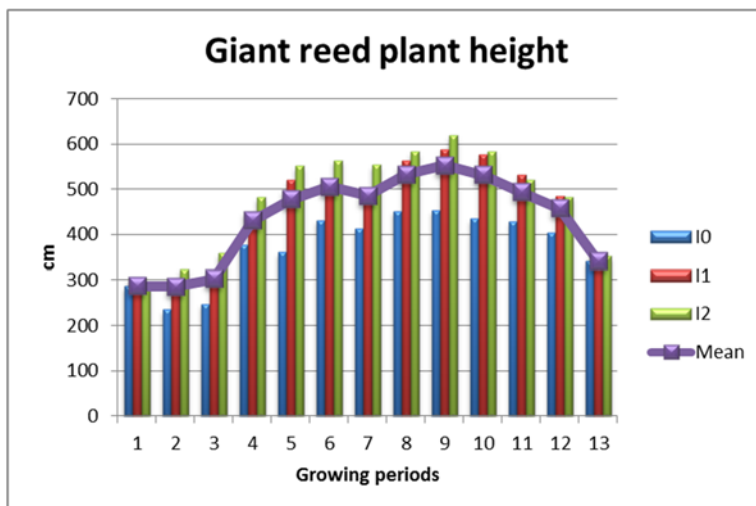
**Picture 1 - CRES.** Large and small fields of perennial crops established in North and Central Greece.



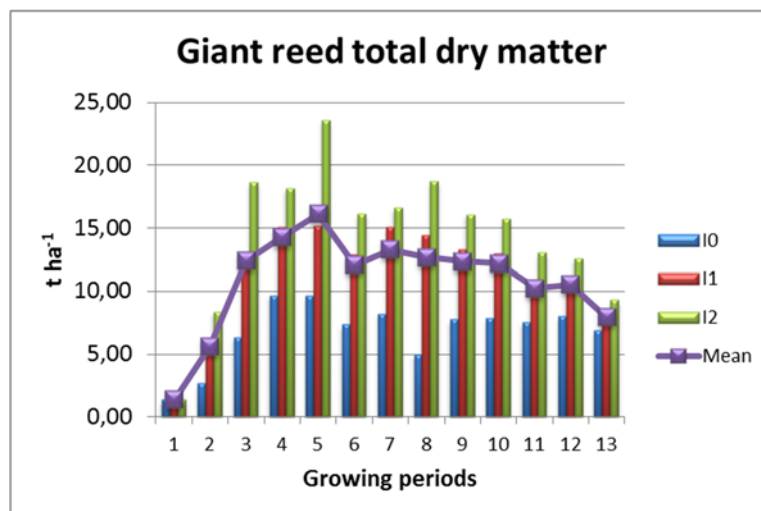
**Picture 2 - CRES.** The experimental fields of cardoon, giant reed and switchgrass



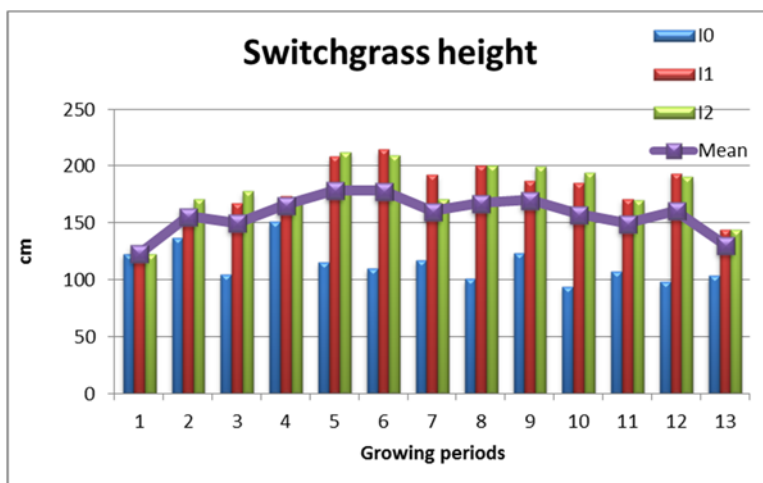
**Picture 3 - CRES.** Panoramic view of the small fields



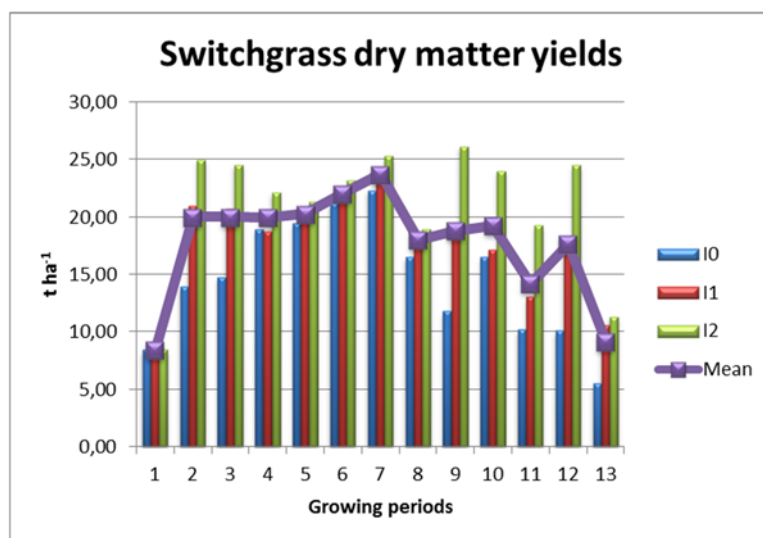
**Figure 1 - CRES.** Plant height for giant reed for three irrigation rates during 13 growing periods



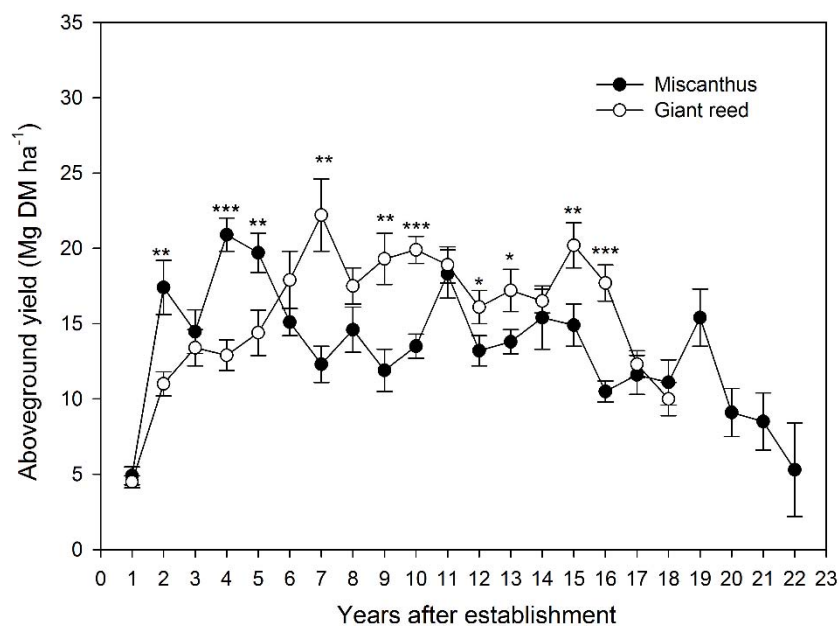
**Figure 2 - CRES.** Total dry matter yields for giant reed for three irrigation rates during 13 growing periods



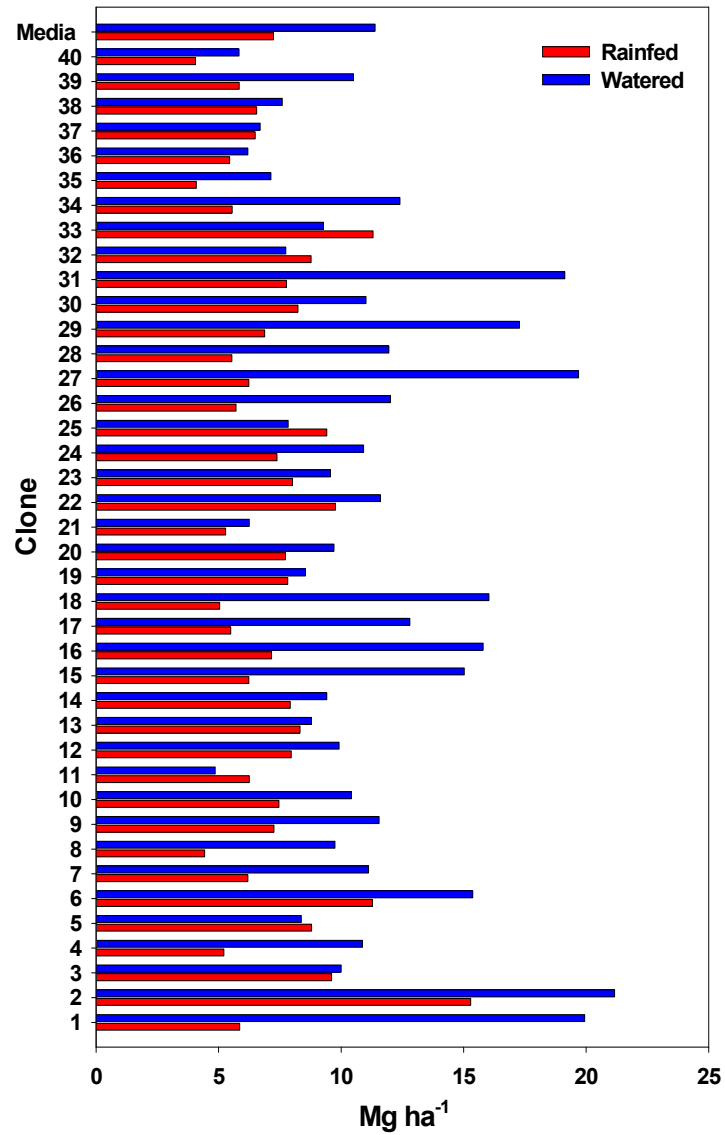
**Figure 3 – CRES.** Plant height for giant reed for three irrigation rates during 13 growing periods



**Figure 4 - CRES.** Total dry matter yields for switchgrass for three irrigation rates during 12 growing periods

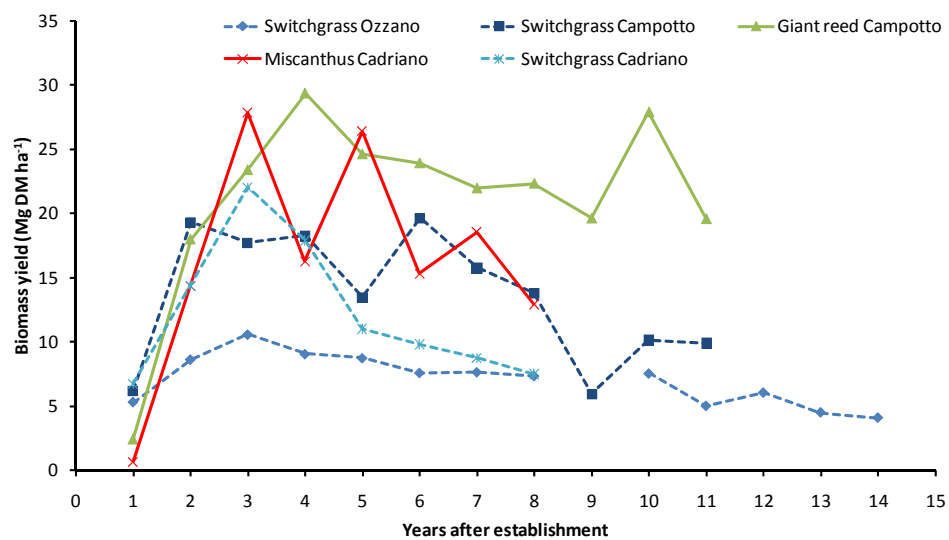


**Figure 1 - UNICT.** Long term side-by-side biomass yield of *Miscanthus x giganteus* and giant reed (*Arundo donax*) at Experimental farm of UNICT (Catania plain, 10 m a.s.l., 37°27' N, 15° 03' E).



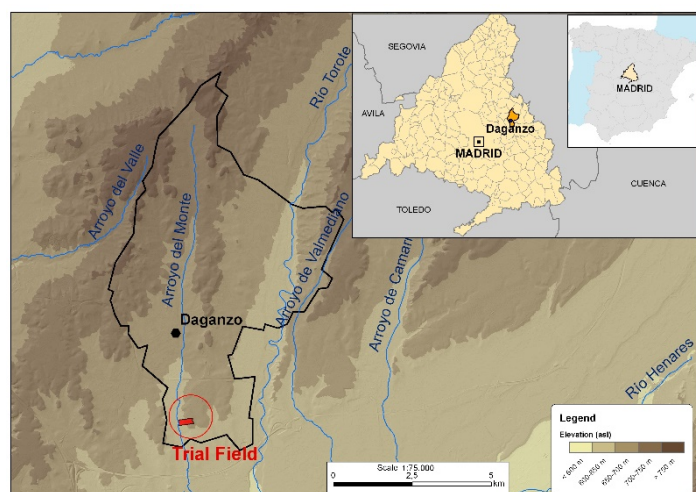
**Figure 2 - UNICT.** Aboveground biomass yield (Mg DM ha<sup>-1</sup>) of *Arundo donax* clones in rainfed ( $I_0$ ) and 100% ETm restitution conditions ( $I_{100}$ ) in semi-arid Mediterranean environment.

## University of Bologna - UNIBO (n. 2)

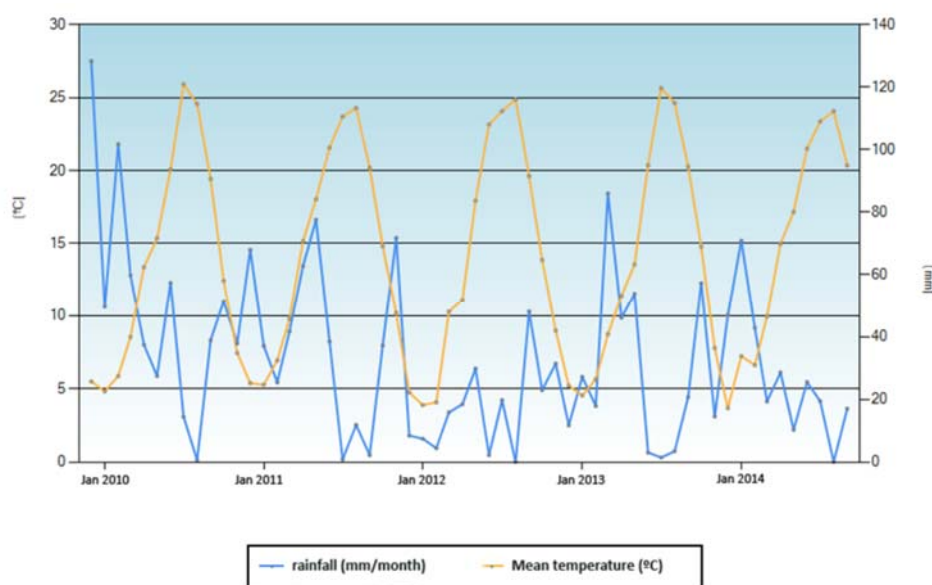


**Figure 1 - UNIBO.** Long term productivities of switchgrass, miscanthus and giant reed grown in different locations by UNIBO. Dotted lines refer to switchgrass grown in different sites.

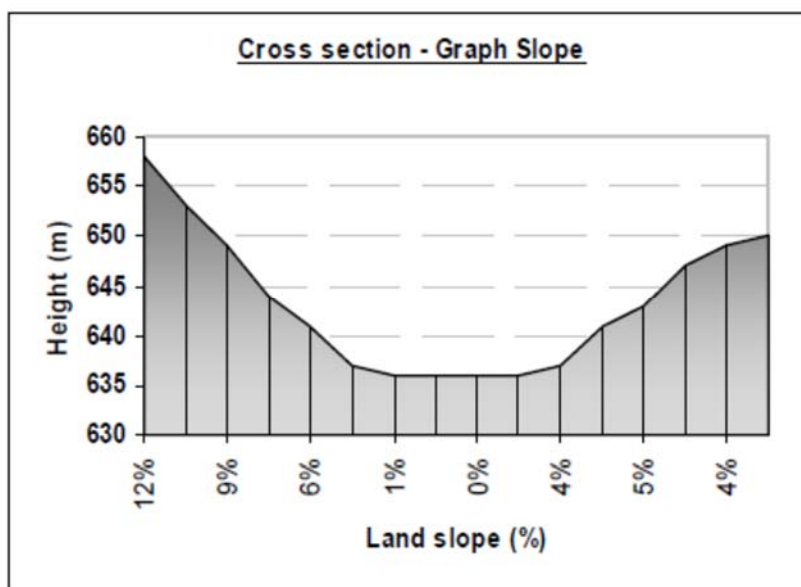
## Universidad Politecnica De Madrid – UPM (n. 5)



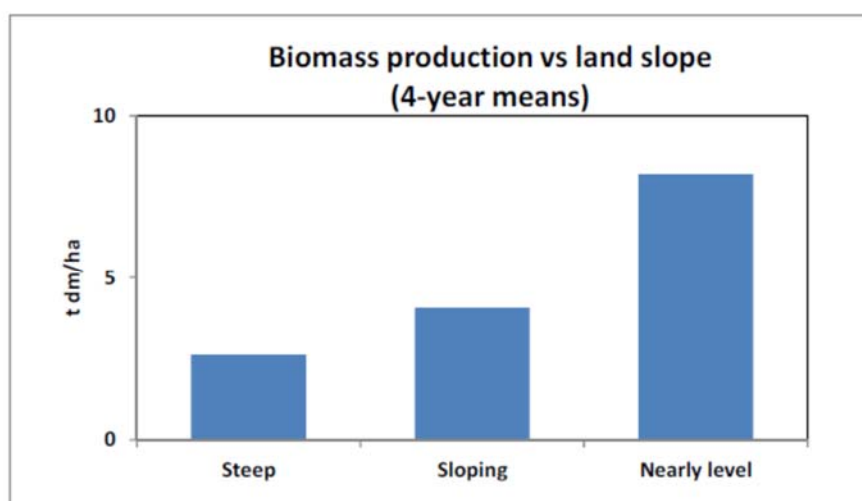
**Figure 1 - UPM.** Location map of the cynara crop under study (Daganzo, Spain).



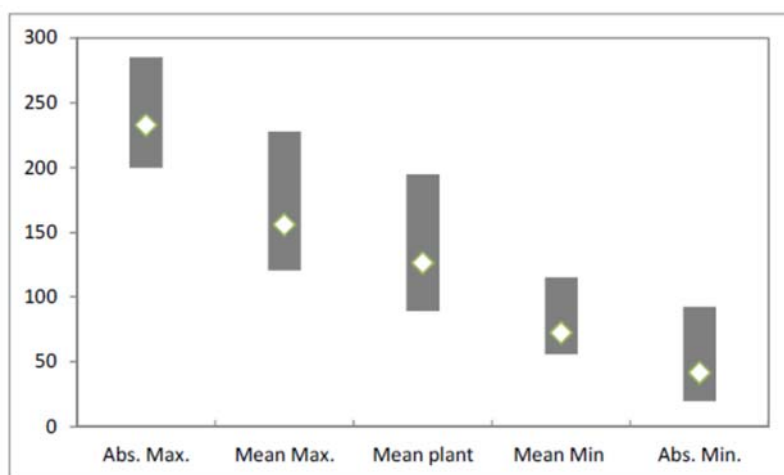
**Figure 2 - UPM.** Mean monthly temperature and monthly precipitation during the experiment. Records from the Meteorological Station located at (UTM) 30N 457867,4473610; 604 m a.s.l. Source: Ministry of Agriculture, Food and Environment of Spain.



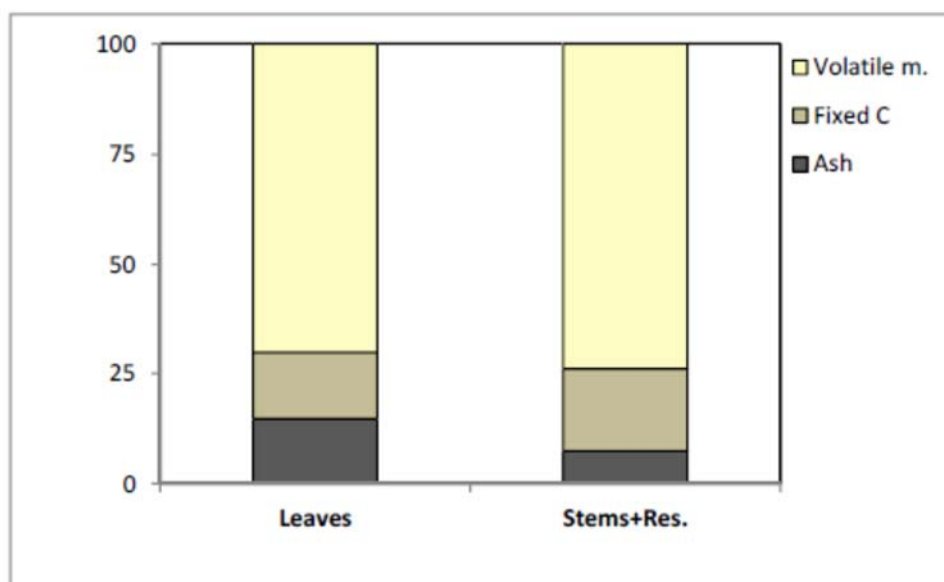
**Figure 3 - UPM.** Elevation profile and slope from one sampling spot to another.



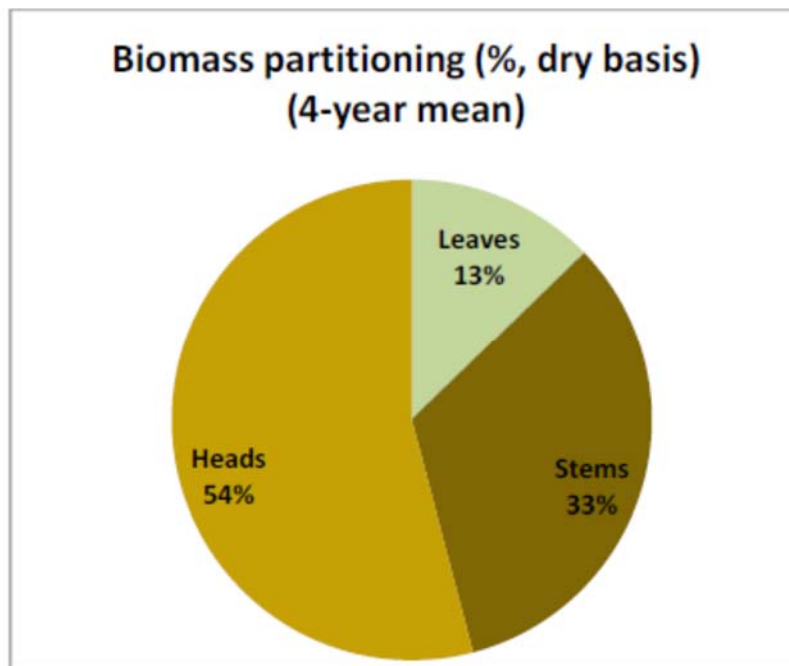
**Figure 4 - UPM.** Four-year means of biomass production of cynara grown in dry farming at the demonstrative scale in Daganzo (Spain) (355 mm rainfall per year on average).



**Figure 5 - UPM.** Range of values for canopy height in the 5 years of cynara cultivation across 8-ha crop in Daganzo (Spain), in dry farming.



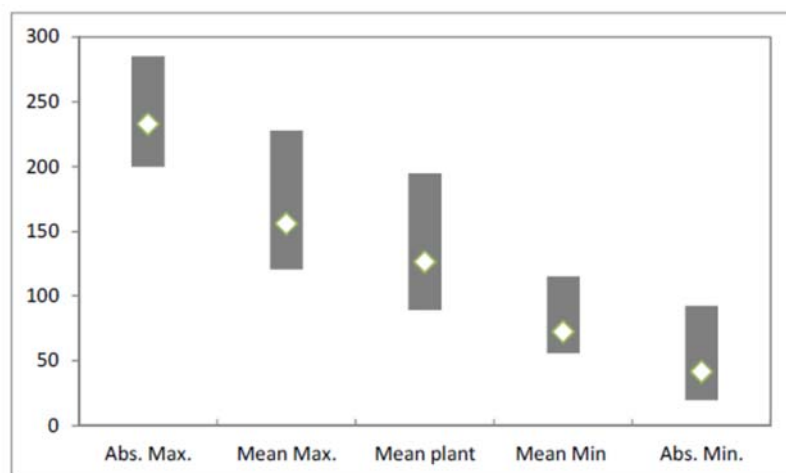
**Figure 6 - UPM.** Range of values for canopy height in the 5 years of cynara cultivation across 8-ha crop in Daganzo (Spain), in dry farming.



**Figure 5 - UPM.** Mean results of biomass partitioning (%w/w) (4 consecutive harvests) of cynara in dry farming at the demonstrative scale in Daganzo (Spain).

## Task 4.4 Reducing biomass losses during harvesting & storage (CRA-ING, SPAPPERI, UPM, SPAPPERI, CRDT)

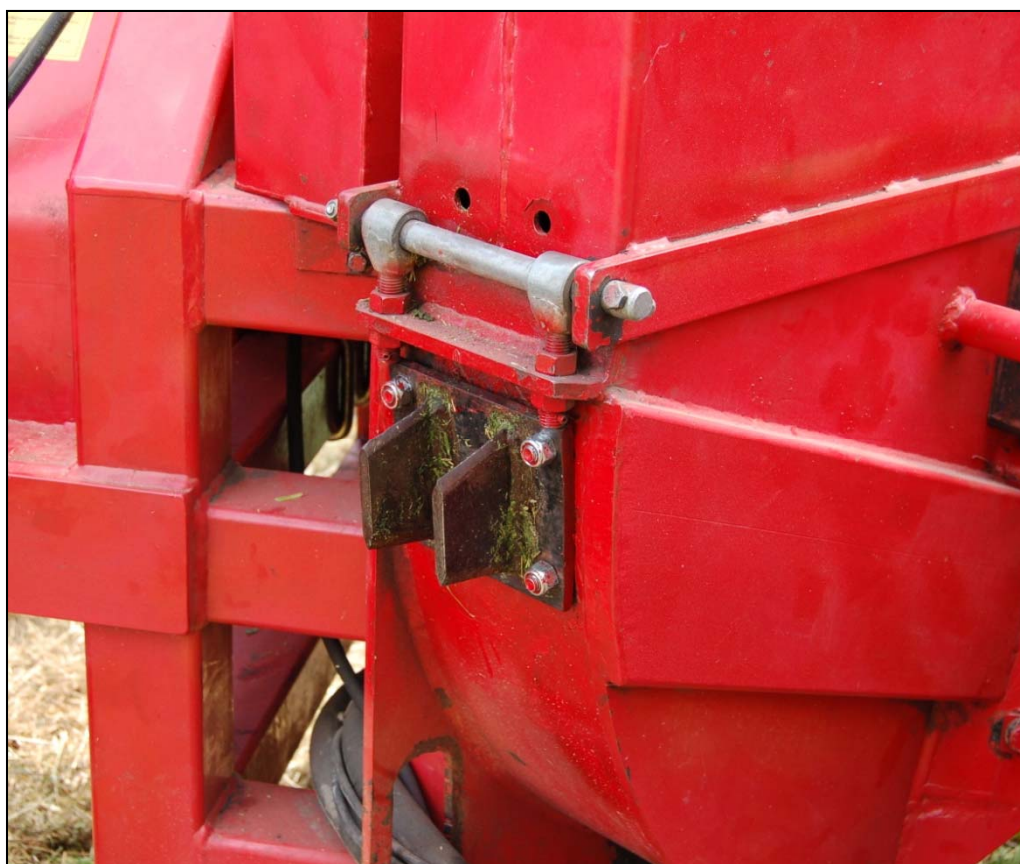
Universidad Politecnica De Madrid – UPM (n. 5)



**Figure 1 - UPM.** Range of values for canopy height in the 5 years of cynara cultivation across 8-ha crop in Daganzo (Spain), in dry farming.



**Picture 1 - UPM.** Mechanical harvest of seeds of cynara by means of a conventional John Deere W650 harvester with Kemper 206 header. September 2013..



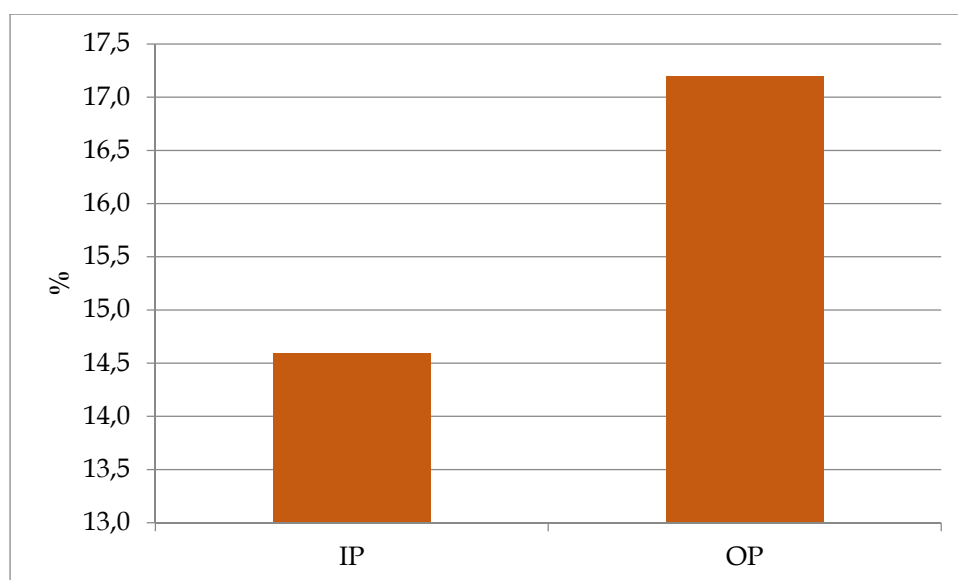
**Figure 1 - SPAPPERI.** Outer positioning of the counter-blades of the rotor.



**Figure 2 – SPAPPERI.** Details of the product obtained keeping the counter-blades in the outer or inner position. The photograph shows the cut pieces at the time of harvesting (green portions, top) or previously sieving (dried portions, bottom).

**Table 1 – SPAPPERI.** Particle size distribution (%) obtained using inner or outer position of the counter-blades. The ANOVA was performed on the arcsine square root of the percentage values. Different letters next to the averages indicate statistically significant differences (Duncan test).

| Size classes | Settings cut |          |
|--------------|--------------|----------|
|              | Inner        | Outer    |
| 100-125      | 0.00 I       | 0.00 I   |
| 75-100       | 0.00 I       | 0.43 I   |
| 50-75        | 1.18 I       | 0.90 I   |
| 25-50        | 2.44 I       | 2.59 I   |
| 12.5-25      | 10.97 G      | 13.28 G  |
| 6.3-12.5     | 39.11 A      | 37.75 A  |
| 3.15-6.3     | 26.83 D      | 24.88 DE |
| < 3.15       | 19.47 F      | 20.38 F  |



**Figure 3 – SPAPPERI.** Cumulative percentage of particles in the range 12.5-100 mm observed for the different position (inner or outer) of the counter-blades.



**Figura 4 – SPAPPERI.** Mowing and conditioning of Miscanthus  
<http://adlib.everysite.co.uk/resources/000/025/574/Miscanthusfig9.jpg>



**Figure 5 – SPAPPERI.** Mowing and conditioning of Switchgrass <https://passel.unl.edu/pages/printinformationmodule.php?idinformationmodule=1130447202&idcollectionmodule=1130274200>



**Figure 6 – SPAPPERI.** Harvesting yard on *Arundo donax* L. composed by mulcher and baler; the biomass is collected only in one tractor passage. [http://www.nobili.com/en/cat\\_serie.php?ID=411](http://www.nobili.com/en/cat_serie.php?ID=411)



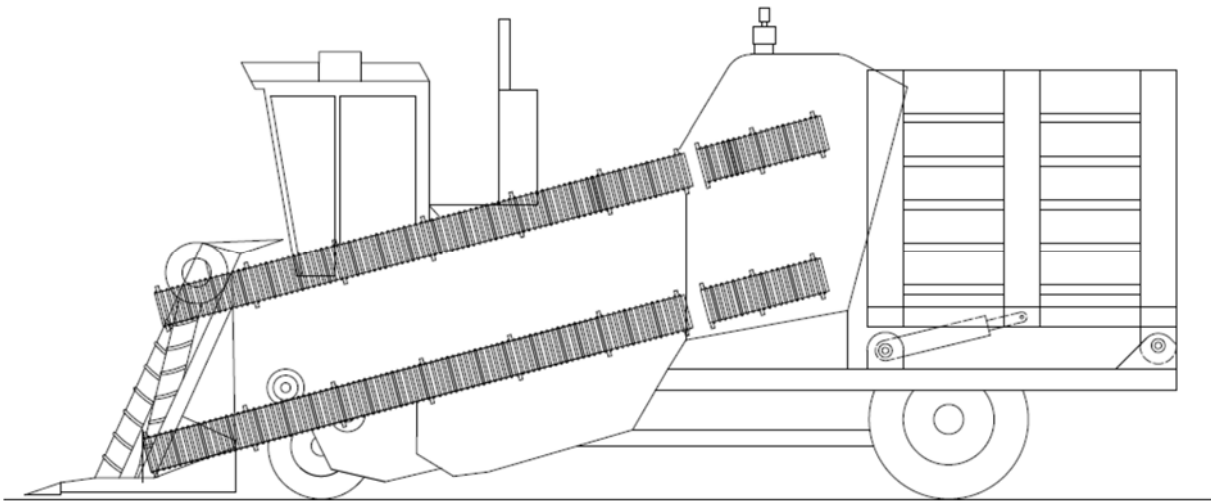
**Figure 7 – SPAPPERI.** Claas jaguar and kemper head during harvesting operation in northern Italy.  
Photo by CREA-ING

**Table 2 – SPAPPERI.** Biobaler characteristics

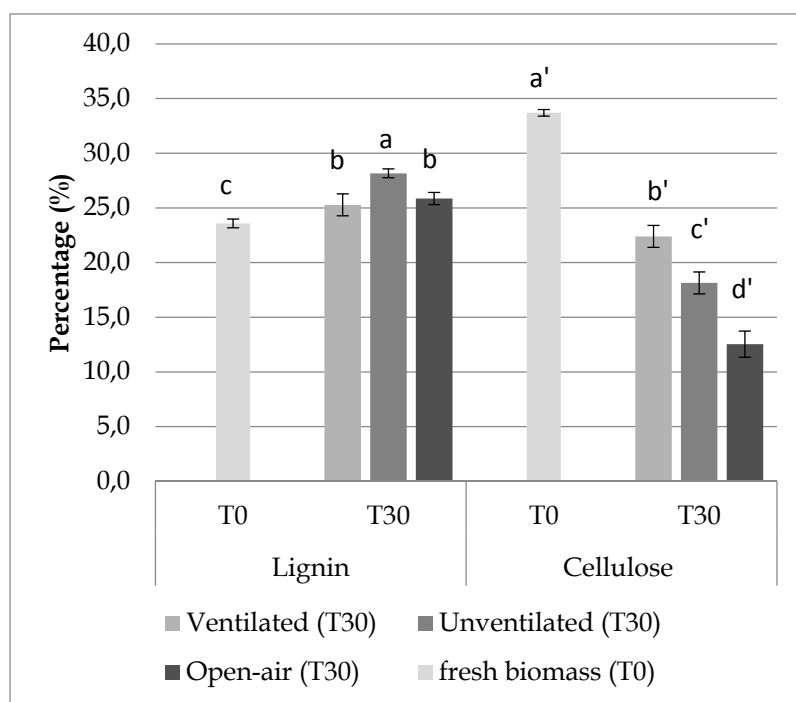
|                                                                                      |               |                                                                                                                                                                                                                              |  |
|--------------------------------------------------------------------------------------|---------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--|
|  |               | <p>The BioBaler Harvesting System is a simple concept. In a single pass, with only one operator, the BioBaler cuts and compacts biomass into dense round bale. Bales can be collected on site at any time after harvest.</p> |  |
| Manufacturer                                                                         | Anderson      |                                                                                                                                                                                                                              |  |
| Model                                                                                | Biobaler WB66 |                                                                                                                                                                                                                              |  |
| Drawbar lenght (m)                                                                   | 1.83          | 3.66                                                                                                                                                                                                                         |  |
| Overall Width (mm)                                                                   | 2585          |                                                                                                                                                                                                                              |  |

|                                     |                    |      |
|-------------------------------------|--------------------|------|
| Working width (mm)                  |                    | 2250 |
| Height (mm)*                        | Min 2460- Max 2970 |      |
| Length(mm)                          | 5460               | 7290 |
| PTO (rpm)                           | 1000               |      |
| Weight (Kg)                         | 6500               | 6820 |
| Power requirements (Kw)             | 150                |      |
| Bale size (mm x mm)                 | 1200 X 1200        |      |
| Bale weight (kg)                    | 500-600            |      |
| Number of teeth of the mulcher      | 50                 |      |
| Number of hammers of the feed rotor | 38                 |      |
| Productivity (t h <sup>-1</sup> )   | 8-20               |      |

\* Maximum and minimum height based upon the position of the lift system of the pneumatic axle



**Figure 8 – SPAPPERI.** Lateral view of the ideal prototype for mowing and baling perennial grasses for energy purposes



**Figure 1 – CREA-ING.** Lignin and cellulose contents (means  $\pm$  SD) determined on fresh biomass (T<sub>0</sub>) and after storage on the three treatments (T<sub>30</sub>). Different letters indicate a significant difference at the level of  $p < 0.05$  after HSD Tukey's test.

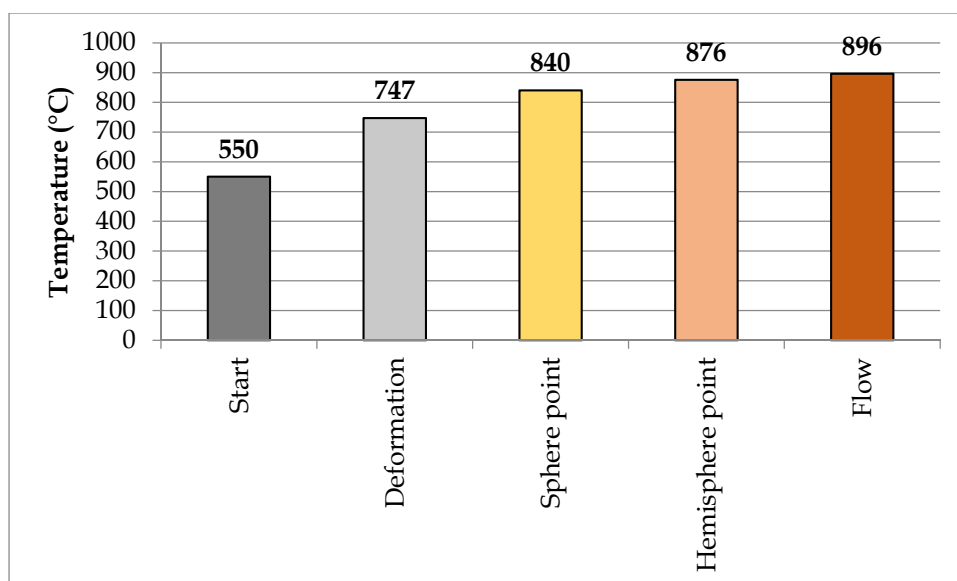
**Table 1 - Fuel quality parameters of fresh biomass (T<sub>0</sub>) or after storage (T<sub>30</sub>) under different storage conditions.**

| Parameter        | U.M.                | Ventilated       |                  | Unventilated     |                  | Open-air         |                  |
|------------------|---------------------|------------------|------------------|------------------|------------------|------------------|------------------|
|                  |                     | T <sub>0</sub>   | T <sub>30</sub>  | T <sub>0</sub>   | T <sub>30</sub>  | T <sub>0</sub>   | T <sub>30</sub>  |
| MC <sup>a</sup>  | %                   | 58.65 $\pm$ 0.2  | 10.50 $\pm$ 0.6  | 58.80 $\pm$ 0.7  | 42.2 $\pm$ 1.15  | 59.50 $\pm$ 1.8  | 60.60 $\pm$ 0.9  |
| HHV <sup>b</sup> | MJ kg <sup>-1</sup> | 17.85 $\pm$ 0.09 | 18.06 $\pm$ 0.26 | 17.89 $\pm$ 0.16 | 17.97 $\pm$ 0.39 | 17.95 $\pm$ 0.08 | 17.96 $\pm$ 0.36 |
| LHV <sup>c</sup> | MJ kg <sup>-1</sup> | 16.64 $\pm$ 0.13 | 16.85 $\pm$ 0.22 | 16.65 $\pm$ 0.14 | 16.74 $\pm$ 0.38 | 16.69 $\pm$ 0.08 | 16.71 $\pm$ 0.34 |
| Ash              | %                   | 7.4 $\pm$ 0.3    | 7.5 $\pm$ 0.3    | 7.85 $\pm$ 0.2   | 7.77 $\pm$ 0.2   | 7.9 $\pm$ 0.3    | 7.92 $\pm$ 0.2   |
| C                | %                   | 46.70            | 46.53            | 46.53            | 46.44            | 46.34            | 45.42            |
| H                | %                   | 5.38             | 5.38             | 5.82             | 5.76             | 5.58             | 5.53             |
| N                | %                   | 1.40             | 1.42             | 1.58             | 1.52             | 1.58             | 1.51             |

<sup>a</sup> MC = moisture content.

<sup>b</sup> HHV = higher heating value.

<sup>c</sup> LHV = lower heating value.



**Figure 2** - CREA-ING. Ash fusion temperatures (°C)



**Figure 3** – CREA-ING. *Arundo* harvesting in sloping degrade of riverbank

**Table 2** – CREA-ING. Main characteristics of the chopping system compared

|                 |                 |    | Excavator-mounted   | Self-propelled      |
|-----------------|-----------------|----|---------------------|---------------------|
| Driving machine |                 |    | Hitachi Zaxis 210 N | Energreen ILFS 1500 |
| Mulcher         |                 |    | Berti ECF MD/SB     | New Speed 150       |
|                 | <i>Motor</i>    |    | Hydraulic           | Hydraulic           |
|                 | <i>Weight</i>   | kg | 895                 | 480                 |
| Rotor           | <i>Lenght</i>   | mm | 1400                | 1416                |
|                 | <i>Diameter</i> | mm | 330                 | 128                 |
| Hammers         |                 | n  | 24                  |                     |
| Knives          |                 | n  |                     | 48 (16x3)           |



**Figure 4 – CREA-ING.** Hydraulic mulching head mounted on the tracked excavator



**Figure 5 - CREA-ING.** Shredder head with knives coupled with a self-propelled



**Figure 6 - CREA-ING.** River banks where the trial was carried out in February (a) and June (b)

**Table 3 - CREA-ING.** River banks analysed

|              |    | February | June     |
|--------------|----|----------|----------|
| Lenght (m)   | m  | 140      | 160      |
| Width (m)    | m  | 11,2     | 15,6     |
| Area (ha)    | ha | 0,16     | 0,25     |
|              | km | 1,14     | 1,6      |
| Previous cut |    | one year | one year |

**Table 4 - CREA-ING.** Main morpho-productive parameters.

|                 |                       |                    | February | June  |
|-----------------|-----------------------|--------------------|----------|-------|
| Plant traits    | Density               | n m <sup>-2</sup>  | 37,4     | 29,3  |
|                 | Height                | cm                 | 355,6    | 346,3 |
|                 | Diameter              | mm                 | 14,2     | 20,6  |
|                 | Weight                | kg m <sup>-2</sup> | 8,31     | 17,3  |
| Productive data | Yield <sub>f.w.</sub> | t km <sup>-1</sup> | 93,3     | 270,0 |
|                 |                       | t ha <sup>-1</sup> | 83,3     | 172,6 |
|                 | Moisture              | %                  | 47,9     | 65,8  |
|                 | Yield <sub>d.w.</sub> | t km <sup>-1</sup> | 48,6     | 92,3  |
|                 |                       | t ha <sup>-1</sup> | 43,4     | 59,0  |

**Table 5 - CREA-ING.** Average bulk density observed for month, implement and their interaction (month x implement).

| Month    | Implement |         | Mean    |
|----------|-----------|---------|---------|
|          | Hammer    | Knife   |         |
| February | 77.9 B    | 107.2 A | 92.6 B  |
| June     | 116.3 A   | 110.0 A | 113.2 A |
| Mean     | 97.1 B    | 108.6 A |         |

Different letters indicate statistical significance at the 1% (capital) level

**Table 6 - CREA-ING.** Working time (%) calculated in February and June for the mulcher with hammers and in June for the knives.

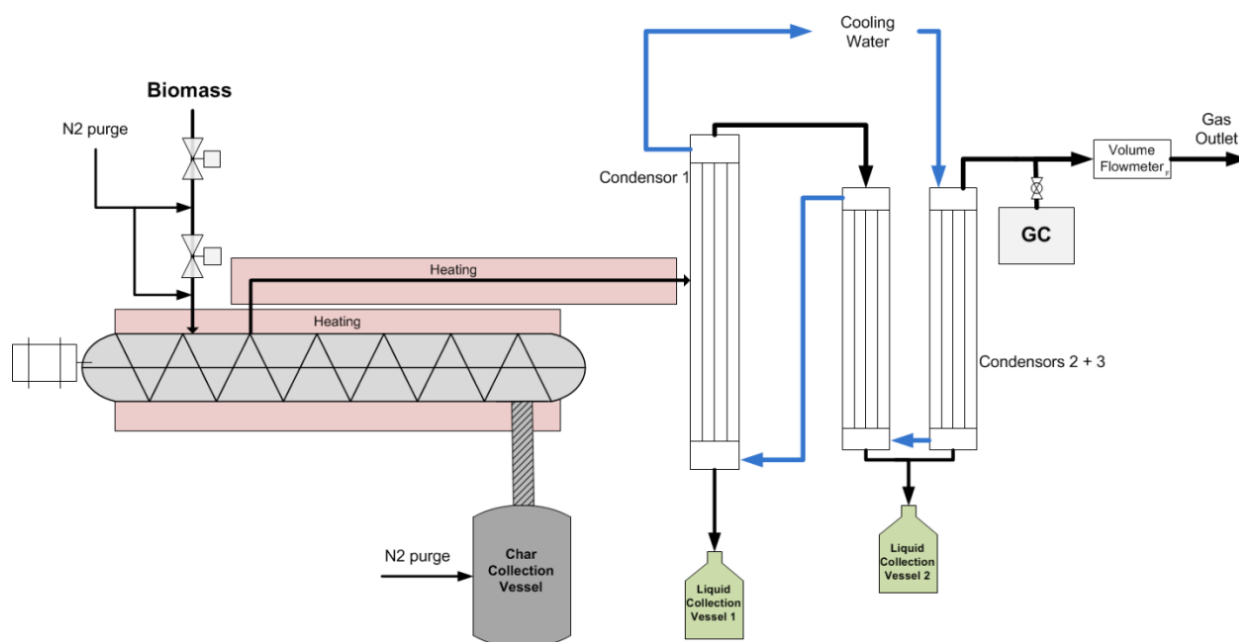
|                               | Hammers  |        | Knives |
|-------------------------------|----------|--------|--------|
|                               | February | June   | June   |
| TE - Effective operative time | 86,45    | 78,41  | 92,67  |
| TA - Time for turning         | 13,55    | 21,59  | 7,33   |
| TO - Operative time           | 100,00   | 100,00 | 100,00 |

**Table 7 - CREA-ING.** Performance of the machines observed in February and June for the heading with hammers and in June for the knives

|                                  |                    | <b>Hammers</b>  |             | <b>Knives</b> |
|----------------------------------|--------------------|-----------------|-------------|---------------|
|                                  |                    | <b>February</b> | <b>June</b> | <b>June</b>   |
| Field efficiency                 | %                  | 86,45           | 78,41       | 92,67         |
| Effective speed                  | m s <sup>-1</sup>  | 0,64            | 0,67        | 0,62          |
| Operative speed                  | m s <sup>-1</sup>  | 0,55            | 0,52        | 0,58          |
| Theoretical field capacity (TFC) | ha h <sup>-1</sup> | 0,09            | 0,11        | 0,15          |
| Effective field capacity (EFC)   | ha h <sup>-1</sup> | 0,07            | 0,09        | 0,14          |
| Work productivity (WP)           | t h <sup>-1</sup>  | 6,70            | 15,05       | 25,16         |



**Figure 1 – BTG.** photograph of the Arundo (left) and Miscanthus (right) biomass before and after size reduction.



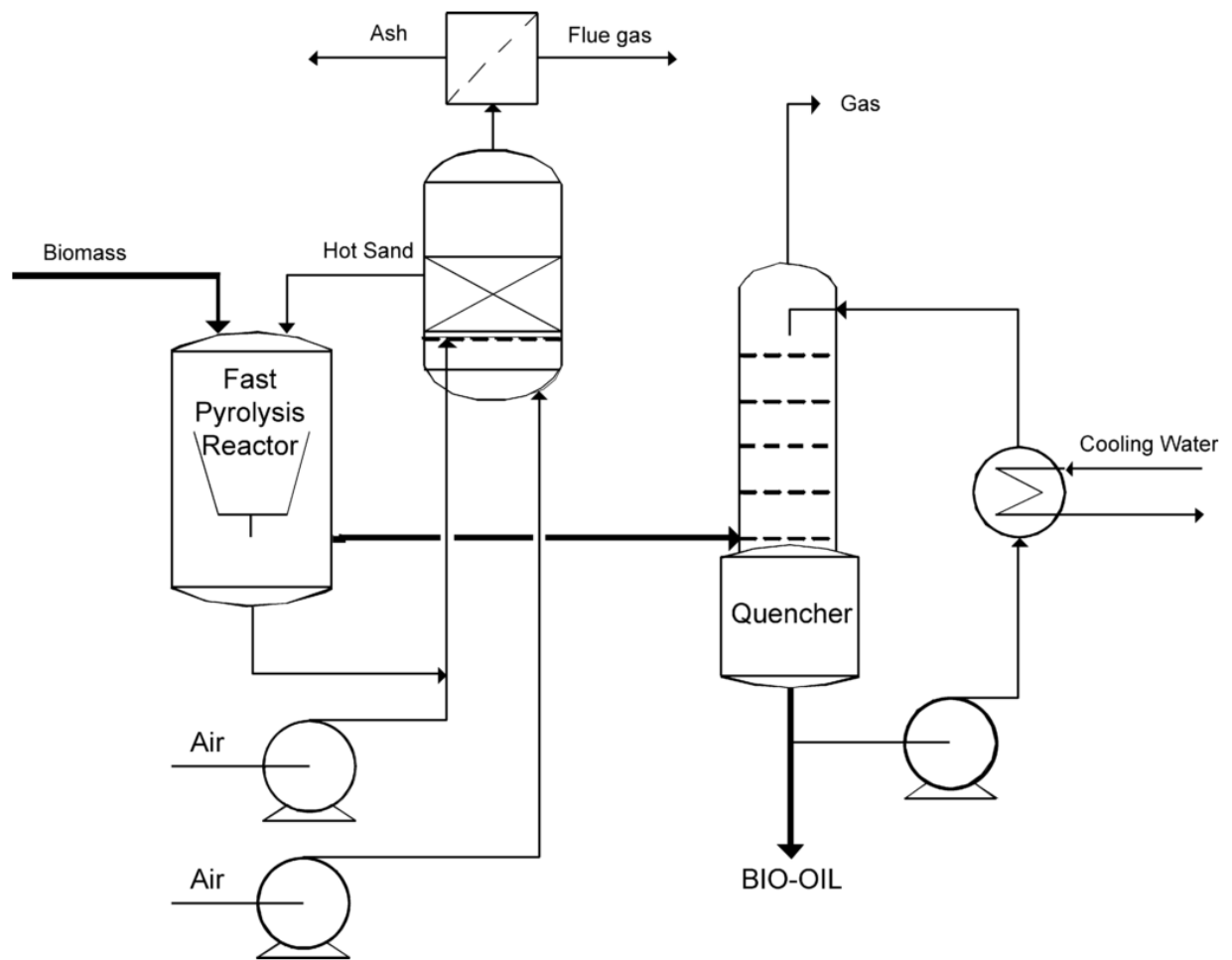
**Figure 2 – BTG.** Schematic representation of the torrefaction setup

**Table 2 – BTG.** Torrefaction operating conditions

| Parameter     | Arundo test | Miscanthus test | Unit   |
|---------------|-------------|-----------------|--------|
| Temperature   | 283         | 282             | [°C]   |
| Feed capacity | 0.35        | 0.32            | [kg/h] |
| Runtime       | 1.90        | 1.85            | [h]    |



**Figure 3 – BTG.** Photograph of the torrefaction setup



**Figure 4 – BTG.** Schematic representation of the fast pyrolysis setup



**Figure 5 – BTG.** Photograph of the fast pyrolysis setup

**Table 3 – BTG.** Pyrolysis operating conditions.

| Parameter     | Arundo test | Miscanthus test | Unit   |
|---------------|-------------|-----------------|--------|
| Temperature   | 529         | 520             | [°C]   |
| Feed capacity | 3.42        | 3.57            | [kg/h] |
| Runtime       | 1.7         | 1.6             | [h]    |

**Table 4 – BTG.** Mass, energy and carbon balance for the torrefaction tests

|               | Mass yield |            | Energy yield |            | Carbon yield |            |
|---------------|------------|------------|--------------|------------|--------------|------------|
|               | Arundo     | Miscanthus | Arundo       | Miscanthus | Arundo       | Miscanthus |
| <b>Char</b>   | 69%        | 74%        | 90%          | 87%        | 87%          | 87%        |
| <b>Liquid</b> | 18%        | 15%        | 5%           | 4%         | 7%           | 6%         |
| <b>Gas</b>    | 6%         | 4%         | 3%           | 2%         | 5%           | 3%         |
| <b>Total</b>  | 94%        | 93%        | 98%          | 93%        | 99%          | 95%        |

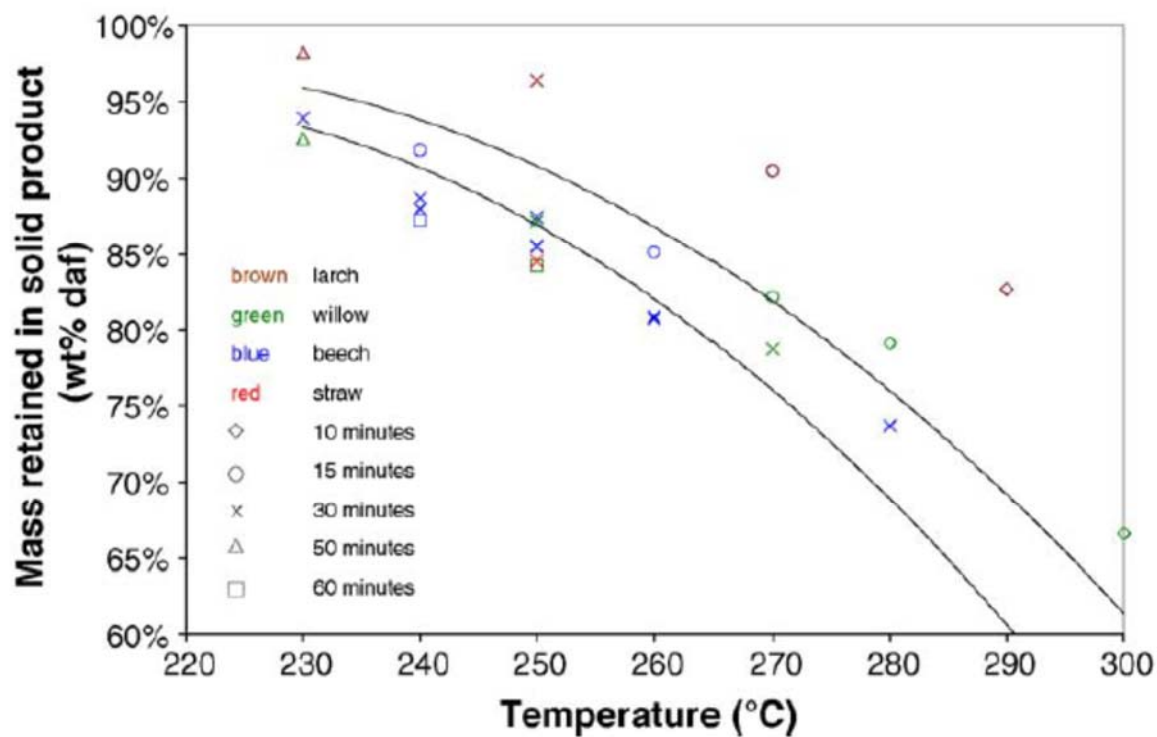


Figure 6 – BTG. Percentage char from product as result of the torrefaction temperature (Prins, 2006).

Table 5 – BTG. Mass, energy and carbon balance for the pyrolysis tests

|        | Mass yield |            | Energy yield |            | Carbon yield |            |
|--------|------------|------------|--------------|------------|--------------|------------|
|        | Arundo     | Miscanthus | Arundo       | Miscanthus | Arundo       | Miscanthus |
| Liquid | 55%        | 63%        | 55%          | 58%        | 55%          | 60%        |
| Gas    | 21%        | 19%        | 8%           | 6%         | 19%          | 17%        |
| Char   | 19%        | 15%        | 22%          | 18%        | 22%          | 18%        |
| Ash    | 6%         | 3%         | -            | -          | -            | -          |
| Total  | 100%       | 100%       | 86%          | 82%        | 95%          | 95%        |

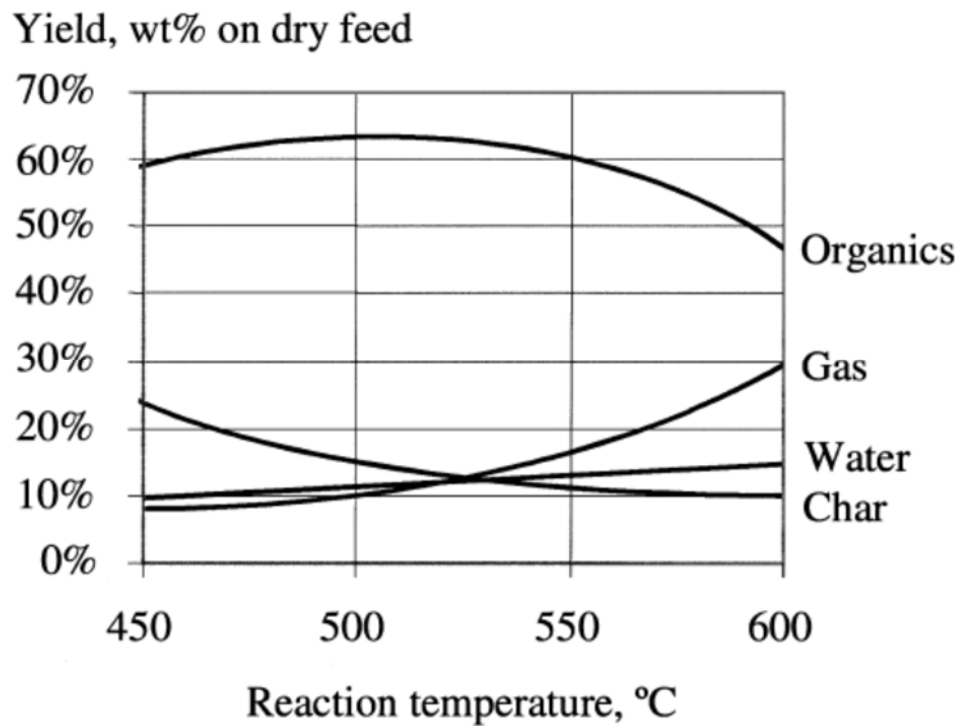


Figure 7 – BTG. Yields pyrolysis dependent on temperature (Bridgewater, 1999).

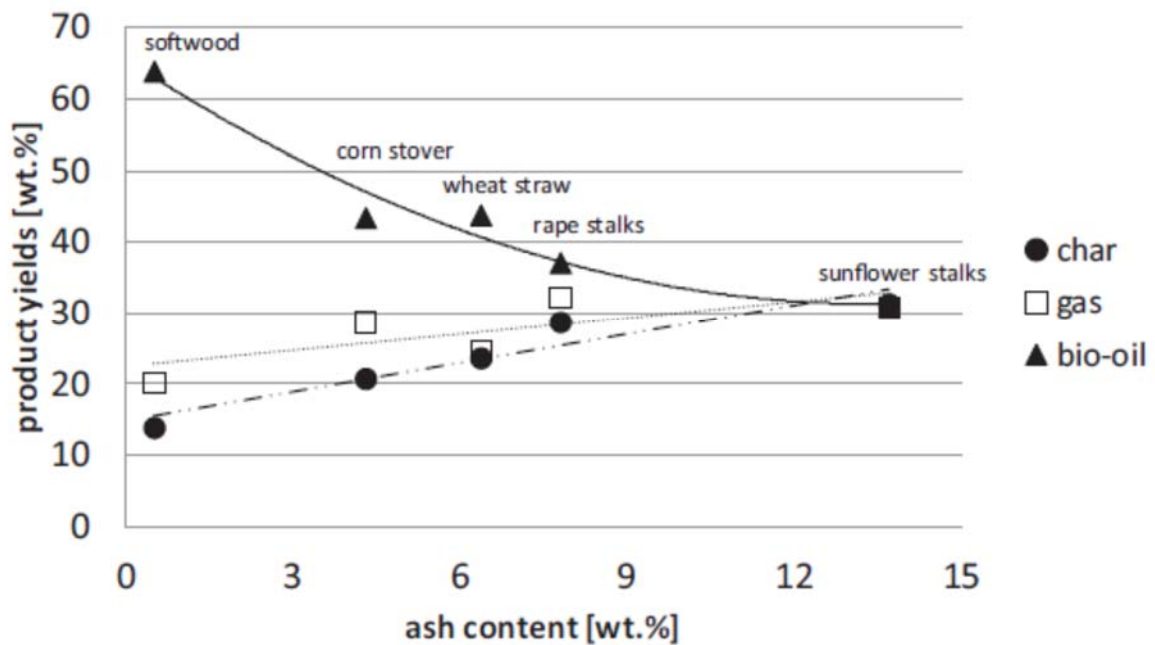


Figure 8 – BTG. Pyrolysis yields mass based of several feedstock Troger (2013).

**Table 6 – BTG.** Proximate and Elemental Analysis Miscanthus and Arundo from BTG laboratory. The green cells are measured, grey cells are calculated from the measurements.

| Feedstock                   | As received basis  |               |             |             |             |                    | Dry basis     |             |             |             |                       | Calculated (Milne)    |                    |                   |
|-----------------------------|--------------------|---------------|-------------|-------------|-------------|--------------------|---------------|-------------|-------------|-------------|-----------------------|-----------------------|--------------------|-------------------|
|                             | Moisture<br>[wt.%] | Ash<br>[wt.%] | C<br>[wt.%] | H<br>[wt.%] | N<br>[wt.%] | O (diff)<br>[wt.%] | Ash<br>[wt.%] | C<br>[wt.%] | H<br>[wt.%] | N<br>[wt.%] | O<br>(diff)<br>[wt.%] | HHV<br>dry<br>[MJ/kg] | LHV dry<br>[MJ/kg] | LHV ar<br>[MJ/kg] |
| Miscanthus feedstock (torr) | 3.4%               | 3.48%         | 43.6%       | 5.9%        | 0.5%        | 43.1%              | 3.60%         | 45.09%      | 6.16%       | 0.53%       | 44.6%                 | 18.0                  | 16.7               | 16.0              |
| Miscanthus feedstock (pyro) | 4.6%               | 3.43%         | 43.0%       | 5.9%        | 0.5%        | 42.6%              | 3.60%         | 45.09%      | 6.16%       | 0.53%       | 44.6%                 | 18.0                  | 16.7               | 15.8              |
| Arundo feedstock (torr)     | 2.7%               | 5.53%         | 42.4%       | 5.7%        | 1.2%        | 42.5%              | 5.68%         | 43.54%      | 5.90%       | 1.25%       | 43.6%                 | 17.2                  | 15.9               | 15.4              |
| Arundo feedstock (pyro)     | 2.4%               | 5.55%         | 42.5%       | 5.8%        | 1.2%        | 42.6%              | 5.68%         | 43.54%      | 5.90%       | 1.25%       | 43.6%                 | 17.2                  | 15.9               | 15.5              |

**Table 7 – BTG.** Proximate and Elemental Analysis Miscanthus and Arundo from Literature.

| Feedstock                             | As received basis  |               |             |             |             |                    | Dry basis     |             |             |             |                    | Calculated (Milne)    |                    |                   |
|---------------------------------------|--------------------|---------------|-------------|-------------|-------------|--------------------|---------------|-------------|-------------|-------------|--------------------|-----------------------|--------------------|-------------------|
|                                       | Moisture<br>[wt.%] | Ash<br>[wt.%] | C<br>[wt.%] | H<br>[wt.%] | N<br>[wt.%] | O (diff)<br>[wt.%] | Ash<br>[wt.%] | C<br>[wt.%] | H<br>[wt.%] | N<br>[wt.%] | O (diff)<br>[wt.%] | HHV<br>dry<br>[MJ/kg] | LHV dry<br>[MJ/kg] | LHV ar<br>[MJ/kg] |
| Miscanthus feedstock (Guanxing, 1999) | 3.40%              | 2.70%         | 46.27%      | 6.09%       | 0.68%       | 42.31%             | 2.8%          | 47.90%      | 6.30%       | 0.70%       | 43.80%             | 16.52                 | 15.15              | 14.55             |
| Miscanthus feedstock (Van Ree, 1997)  | 3.40%              | 1.45%         | 47.29%      | 5.84%       | 0.67%       | 41.30%             | 1.50%         | 48.95%      | 6.05%       | 0.69%       | 42.75%             | 19.68                 | 18.36              | 17.65             |
| Arundo feedstock (Miles, 1995)        | 3.4%               | 3.34%         | 45.79%      | 5.68%       | 0.58%       | 41.60%             | 3.43%         | 45.79%      | 5.84%       | 0.6%        | 42.75%             | 18.85                 | 17.58              | 17.04             |
| Arundo feedstock (Hallgren, 1999)     | 3.4%               | 3.48%         | 42.60%      | 5.02%       | 0.29%       | 45.79%             | 3.60%         | 44.10%      | 5.20%       | 0.29%       | 47.40%             | 18.30                 | 17.17              | 16.50             |

**Table 8 – BTG.** Analysis results of the feedstocks and various products. The green cells are measured, grey cells are calculated from the measurements.

|                         | As received basis |       |        |       |       |          | Dry basis |        |       |       |          | Calculated (Milne) |         |         |
|-------------------------|-------------------|-------|--------|-------|-------|----------|-----------|--------|-------|-------|----------|--------------------|---------|---------|
|                         | Moisture          | Ash   | C      | H     | N     | O (diff) | Ash       | C      | H     | N     | O (diff) | HHV dry            | LHV dry | LHV ar  |
| Torrefaction products   |                   |       |        |       |       |          |           |        |       |       |          |                    |         |         |
| Miscanthus liquid       | 65.1%             | 0.0%  | 17.0%  | 2.0%  | 0.0%  | 81.0%    | 0.00%     | 48.50% | 5.81% | 0.00% | 45.7%    | 18.7               | 17.5    | 4.5     |
| Miscanthus char         | 0%                | 5.3%  | 51.2%  | 5.6%  | 0.5%  | 37.4%    | 5.26%     | 51.19% | 5.60% | 0.53% | 37.4%    | 20.2               | 19.0    | 19.0    |
| Arundo liquid           | 61.7%             | 0.0%  | 16.4%  | 2.5%  | 0.0%  | 81.1%    | 0.00%     | 42.85% | 6.42% | 0.00% | 50.7%    | 17.0               | 15.6    | 4.5     |
| Arundo char             | 0%                | 8.5%  | 53.3%  | 5.4%  | 1.2%  | 31.6%    | 8.49%     | 53.30% | 5.43% | 1.19% | 31.6%    | 21.3               | 20.1    | 20.1    |
| Pyrolysis products      |                   |       |        |       |       |          |           |        |       |       |          |                    |         |         |
| Miscanthus liquid       | 22.9%             | 0%    | 41.4%  | 6.96% | 0.44% | 51.2%    | 0%        | 54%    | 6%    | 0.6%  | 40%      | 21.0               | 19.8    | 14.7    |
| Miscanthus char         | 0%                | 33.2% | 50.9%  | 3.2%  | 0.6%  | 12.1%    | 33%       | 51%    | 3%    | 0.61% | 12.1%    | 19.5               | 18.8    | 18.8    |
| Arundo liquid           | 28.0%             | 0%    | 41.9%  | 7.50% | 1.08% | 49.5%    | 0%        | 58%    | 6%    | 1.5%  | 34%      | 23.6               | 22.3    | 15.4    |
| Arundo char             | 0%                | 39.3% | 48.4%  | 3.0%  | 1.2%  | 8.1%     | 39.3%     | 48.4%  | 3.0%  | 1.2%  | 8.1%     | 18.8               | 18.1    | 18.1    |
| Gas composition         |                   |       |        |       |       |          |           |        |       |       |          |                    |         |         |
|                         |                   |       | CO2    |       |       |          |           |        |       |       |          |                    |         |         |
|                         |                   |       | [kg/h] |       |       |          |           |        |       |       |          |                    |         |         |
| Miscanthus Torrefaction |                   |       | 0.010  |       |       | 0.002    |           |        |       |       | 0.001    |                    |         |         |
| Arundo Torrefaction     |                   |       | 0.018  |       |       | 0.004    |           |        |       |       | 0.001    |                    |         |         |
| Miscanthus Pyrolysis    |                   |       | 0.319  |       |       | 0.261    |           |        |       |       | 0.054    |                    |         |         |
| Arundo Pyrolysis        |                   |       | 0.366  |       |       | 0.252    |           |        |       |       | 0.072    |                    |         |         |
|                         |                   |       |        |       |       |          |           |        |       |       |          | LHV                |         | Carbon  |
|                         |                   |       |        |       |       |          |           |        |       |       |          | [MJ/kg]            |         | [wt. %] |
|                         |                   |       |        |       |       |          |           |        |       |       |          | 7.2                |         | 32%     |
|                         |                   |       |        |       |       |          |           |        |       |       |          | 6.6                |         | 32%     |
|                         |                   |       |        |       |       |          |           |        |       |       |          | 5.2                |         | 38%     |
|                         |                   |       |        |       |       |          |           |        |       |       |          | 6.0                |         | 38%     |

**Table 1 – 2ZK.** The market volumes and price levels for different applications were defined and assessed by the market players.

|                                                                                                    | <b>Market volume</b> | <b>Product value<br/>(€/ton)</b> | <b>Enabling market<br/>introduction</b>    |
|----------------------------------------------------------------------------------------------------|----------------------|----------------------------------|--------------------------------------------|
| <b>Pulp&amp; paper (cellulose<br/>Properties requested (<u>market<br/>value</u>/market volume)</b> | +200 million tons    | 500-1000                         | Price - performance                        |
| <b>PVAc/cellulose</b>                                                                              | + 100 kt             | 700-1500                         | Renewable -<br>performance                 |
| <b>Phenol</b>                                                                                      | + 9 million tons     | ~1400                            | Price – renewable -<br>quality             |
| <b>Cresols</b>                                                                                     | + 100 kt             | ~2500                            | Price - quality                            |
| <b>Antioxidants</b>                                                                                | + 200 kt             | 1500-7000                        | Price – renewable -<br>performance         |
| <b>Phenol resins</b>                                                                               | + million ton        | 1000-4000                        | Price – renewable<br>– performance,<br>SHE |
| <b>Bi(o)tumen</b>                                                                                  | 95 millions          | 300-400, but rising              | Price - performance                        |
| <b>Woody Plastic Composite</b>                                                                     | +260 kT              | Broad range                      | Price - performance                        |
| <b>Fuel additives</b>                                                                              | Million tons         | ~1000                            | Price - performance                        |
| <b>Ethanol</b>                                                                                     | + 70 million tons    | ~ 500                            | Price – renewable -                        |

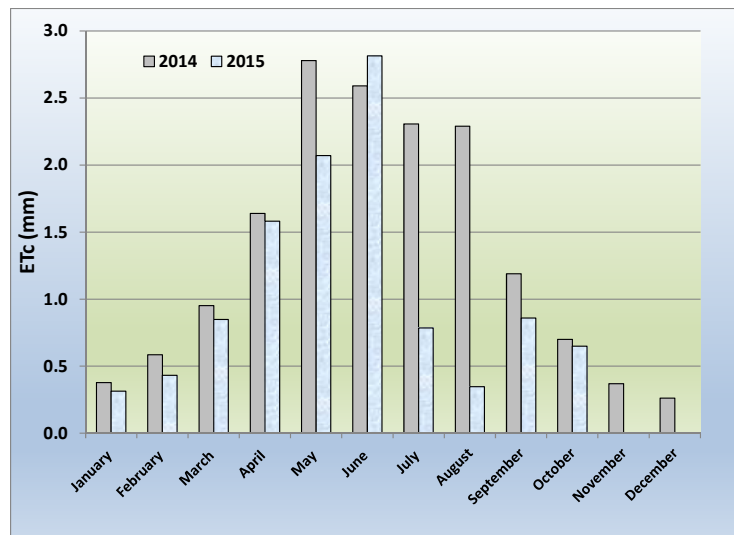
## **Task 6.1 Assessment of CO<sub>2</sub> and water exchange between canopy and atmosphere through micrometeorological “eddy covariance” measurement**

**University of Bologna - UNIBO (n. 2) / Istituto per il Sistema Produzione Animale in Ambiente Mediterraneo – CNR-ISPAAM (n. 8)**

**Table 1.** Monthly value of NEE, GPP and Re (Mg C ha<sup>-1</sup>) during the growing seasons 2014 (left table) and 2015 (right table).

| <b>2014</b>  | <b>NEE<br/>(t C ha<sup>-1</sup>)</b> | <b>GPP<br/>(t C ha<sup>-1</sup>)</b> | <b>ER<br/>(t C ha<sup>-1</sup>)</b> | <b>T air (°C)</b> |
|--------------|--------------------------------------|--------------------------------------|-------------------------------------|-------------------|
| January      | 0.17                                 | -0.29                                | 0.46                                | 5.3               |
| February     | 0.03                                 | -0.52                                | 0.55                                | 7.3               |
| March        | -0.21                                | -0.98                                | 0.77                                | 9.7               |
| April        | -0.73                                | -1.84                                | 1.10                                | 12.6              |
| May          | -1.84                                | -3.11                                | 1.27                                | 16.0              |
| June         | -2.13                                | -3.43                                | 1.29                                | 20.7              |
| July         | -1.94                                | -2.98                                | 1.04                                | 21.1              |
| August       | -1.56                                | -2.66                                | 1.10                                | 21.4              |
| September    | -0.49                                | -1.31                                | 0.82                                | 19.1              |
| October      | 0.45                                 | -0.57                                | 1.02                                | 16.2              |
| November     | 0.34                                 | -0.25                                | 0.59                                | 11.7              |
| December     | 0.08                                 | -0.20                                | 0.28                                | 6.3               |
| <b>Total</b> | <b>-7.82</b>                         | <b>-18.14</b>                        | <b>10.28</b>                        | <b>14.0</b>       |

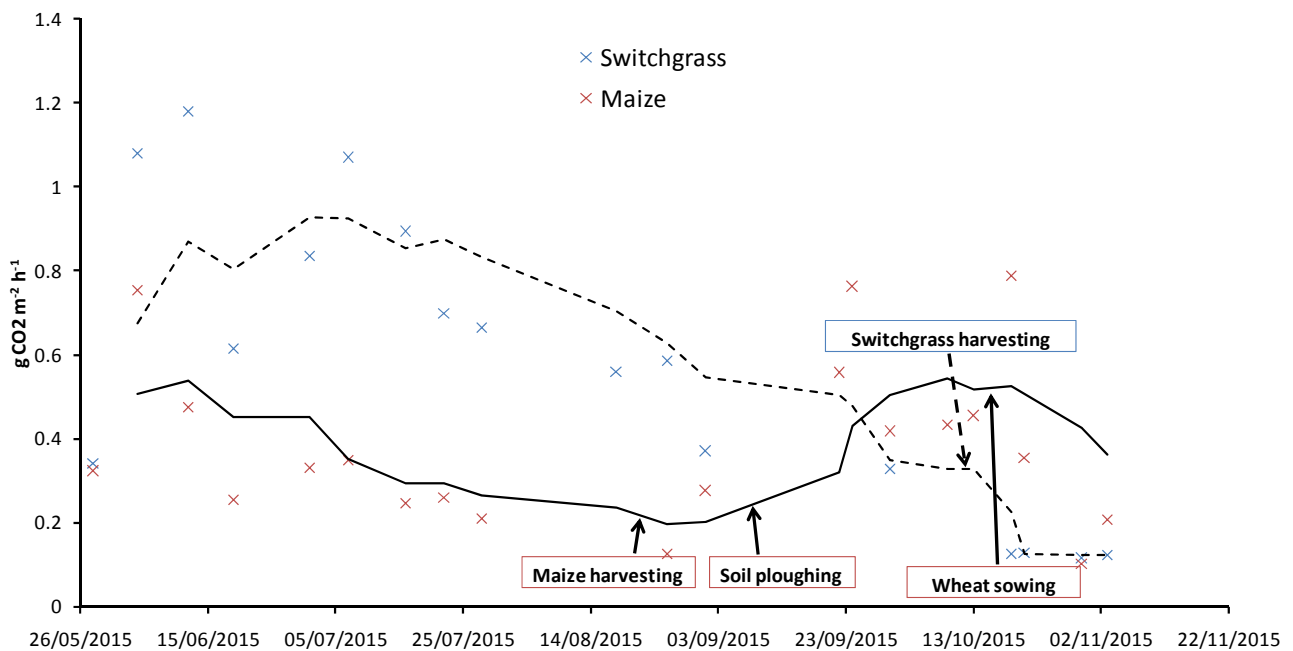
| <b>2015</b>  | <b>NEE<br/>(t C ha<sup>-1</sup>)</b> | <b>GPP<br/>(t C ha<sup>-1</sup>)</b> | <b>ER<br/>(t C ha<sup>-1</sup>)</b> | <b>T air (°C)</b> |
|--------------|--------------------------------------|--------------------------------------|-------------------------------------|-------------------|
| January      | 0.09                                 | -0.23                                | 0.32                                | 3.8               |
| February     | 0.02                                 | -0.31                                | 0.32                                | 4.4               |
| March        | -0.11                                | -0.70                                | 0.59                                | 9.0               |
| April        | -0.48                                | -1.17                                | 0.69                                | 13.7              |
| May          | -1.06                                | -2.10                                | 1.04                                | 18.5              |
| June         | -2.05                                | -3.26                                | 1.21                                | 22.7              |
| July         | -1.75                                | -2.81                                | 1.06                                | 27.6              |
| August       | -1.13                                | -2.20                                | 1.07                                | 25.0              |
| September    | -0.64                                | -1.24                                | 0.60                                | 20.1              |
| October      | 0.26                                 | -0.58                                | 0.84                                | 13.8              |
|              |                                      |                                      |                                     |                   |
|              |                                      |                                      |                                     |                   |
| <b>Total</b> | <b>-6.85</b>                         | <b>-14.59</b>                        | <b>7.74</b>                         | <b>15.9</b>       |



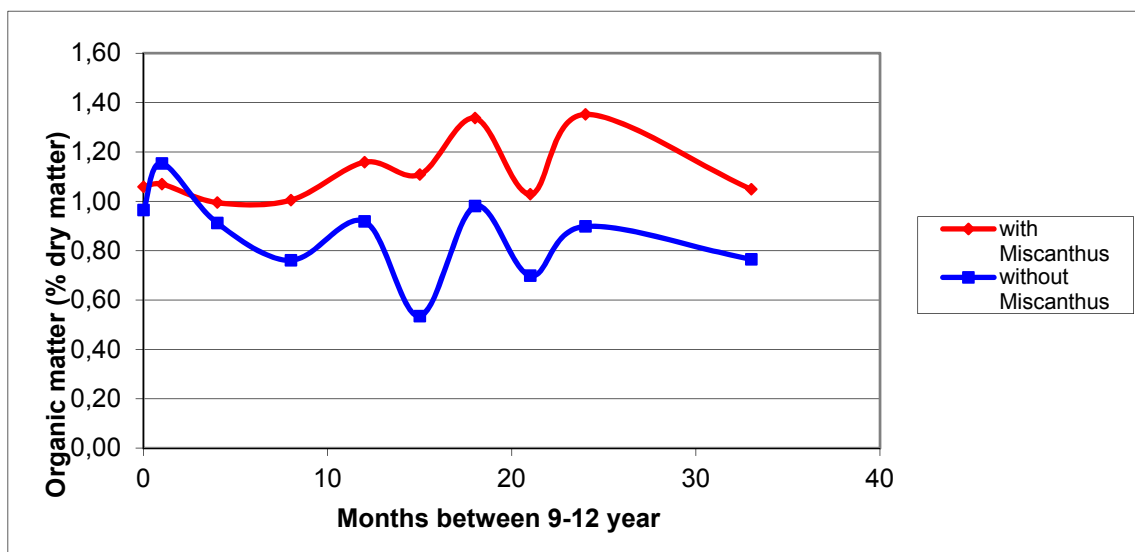
**Figure 1.** Monthly crop evapotranspiration (ET<sub>c</sub>) during 2014 (grey histograms) and 2015 (blue histograms) seasons.

## Task 6.2 - Root development and carbon storage in the soil

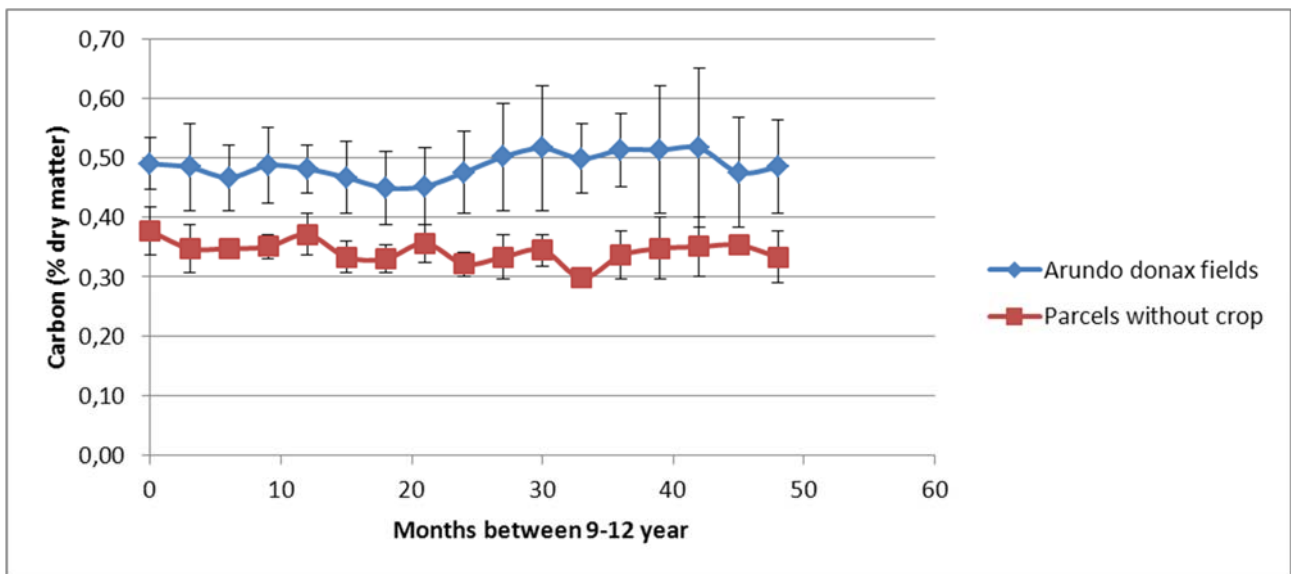
University of Bologna - UNIBO (n. 2) / Faculdade de Ciências e Tecnologia-  
Universidade Nova de Lisboa (n. 4)



**Figure 2.** Soil respiration in the period May-October 2015 for switchgrass (dashed line) and a maize (continuous line) at the experimental farm of UNIBO. Remarkable phases of the two crop cycles are highlighted: blue squares for switchgrass, red squares for maize-wheat rotation.



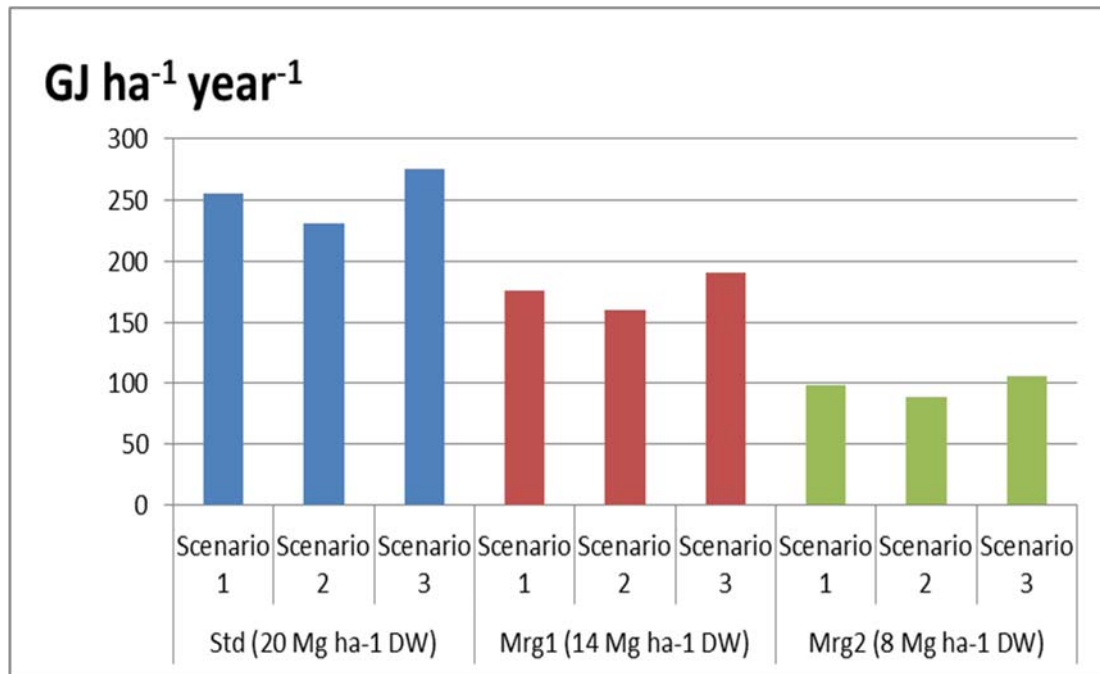
**Figure 3.** Organic matter content in the soil in *Miscanthus* (red line) and fallow fields (blue line). Average results from 33 months, between 9-12 year of *Miscanthus* plantations in FCT-UNL.



**Figure 4.** Carbon organic matter content in the soil of *A. donax* plots (blue line) and fallow ones (red line) at the experimental farm of FCT-UNL.

### Task 6.3 - Energy balance

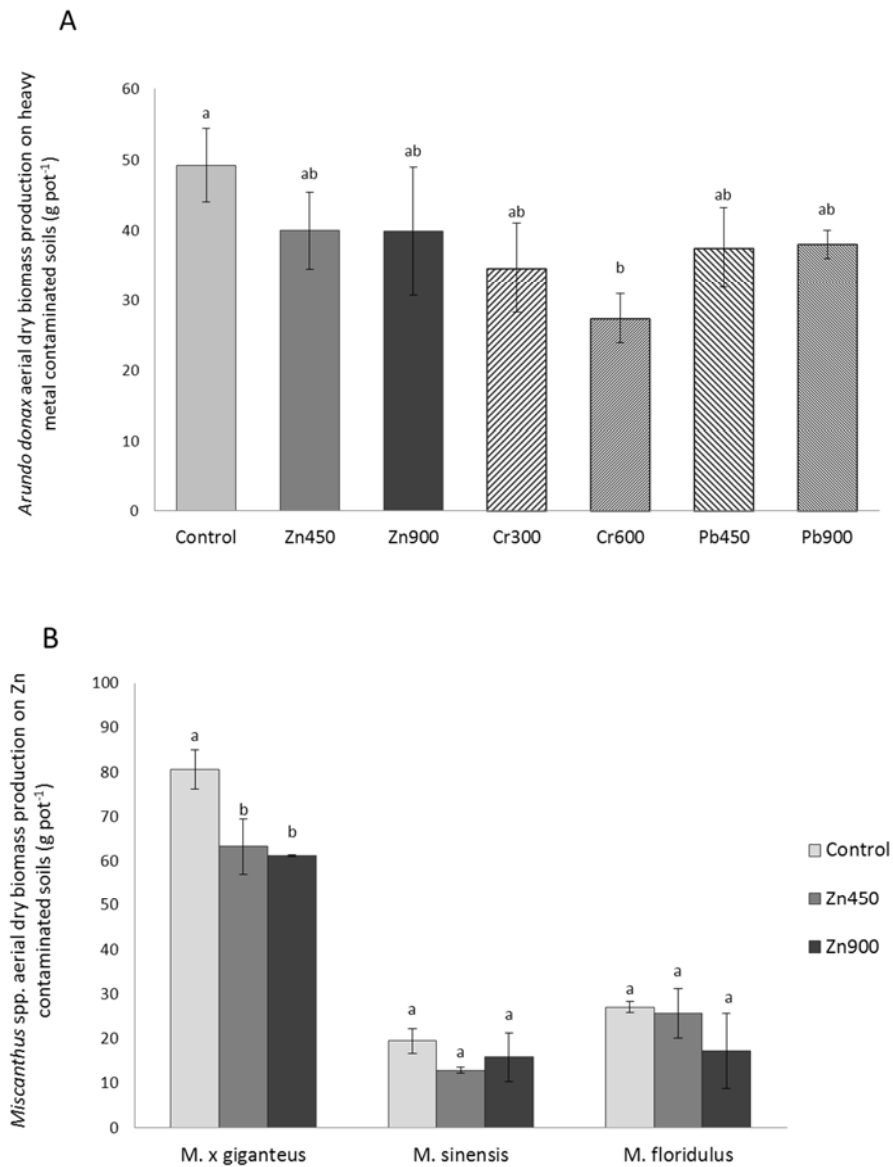
Faculdade de Ciências e Tecnologia-Universidade Nova de Lisboa (n. 4) / University of Bologna - UNIBO (n. 2)



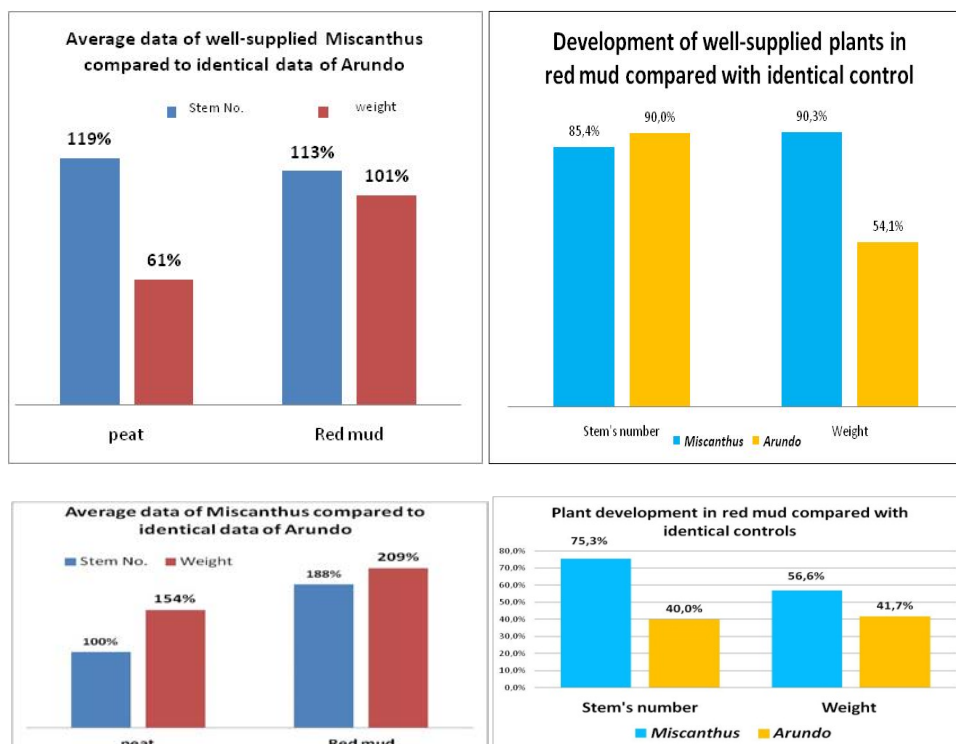
**Figure 5.** Influence of combustion scenarios in the energy balance of the use of *Miscanthus* for energy production, in Portugal (FCT-UNL). Results are from the standard and marginal soils, considering that the crop was harvested in January

## Task 6.4 – Phytoremediation issue

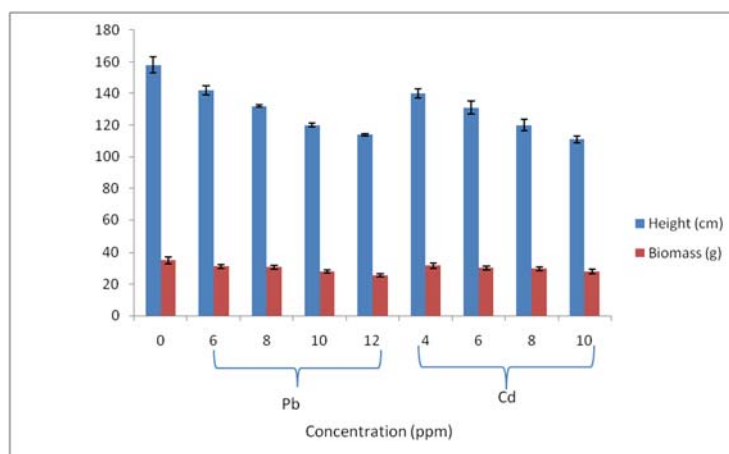
Faculdade de Ciências e Tecnologia-Universidade Nova de Lisboa (n. 4), Indian Institute of Technology, Delhi – CRDT (n. 13), Primus Ltd. – PRIMUS (n. 20)



**Figure 6.** Aerial biomass production of *Arundo donax* L. in Zn, Cr and Pb contaminated soils (A) and aerial biomass production of *Miscanthus* spp. under Zn contaminated soils (B) grown in pots by FCT-UNL. For each species, different lower-case letters indicate statistical significance ( $p < 0.05$ ) among treatments.



**Figure 7.** Results from arundo and miscanthus grown in red mud and peat (control) filled pots at PRIMUS experimental station. **Left:** Average stem number (per pot) and average weight of stems of *Miscanthus* and *Arundo* plants grown in peat and red-mud. Values of miscanthus have been standardized over values of arundo grown in the same conditions. Above graph referred to plants grown in well nourished condition, bottom graph to poorly nourished condition. **Right:** Average stem number and average weight of stems of red-mud-grown *Miscanthus* and *Arundo* plants were compared with the identical controls (same plants grown in peat). Values are presented as standardized over peat grown miscanthus (100%). Above graph in well nourished condition, bottom graph in poorly nourished condition

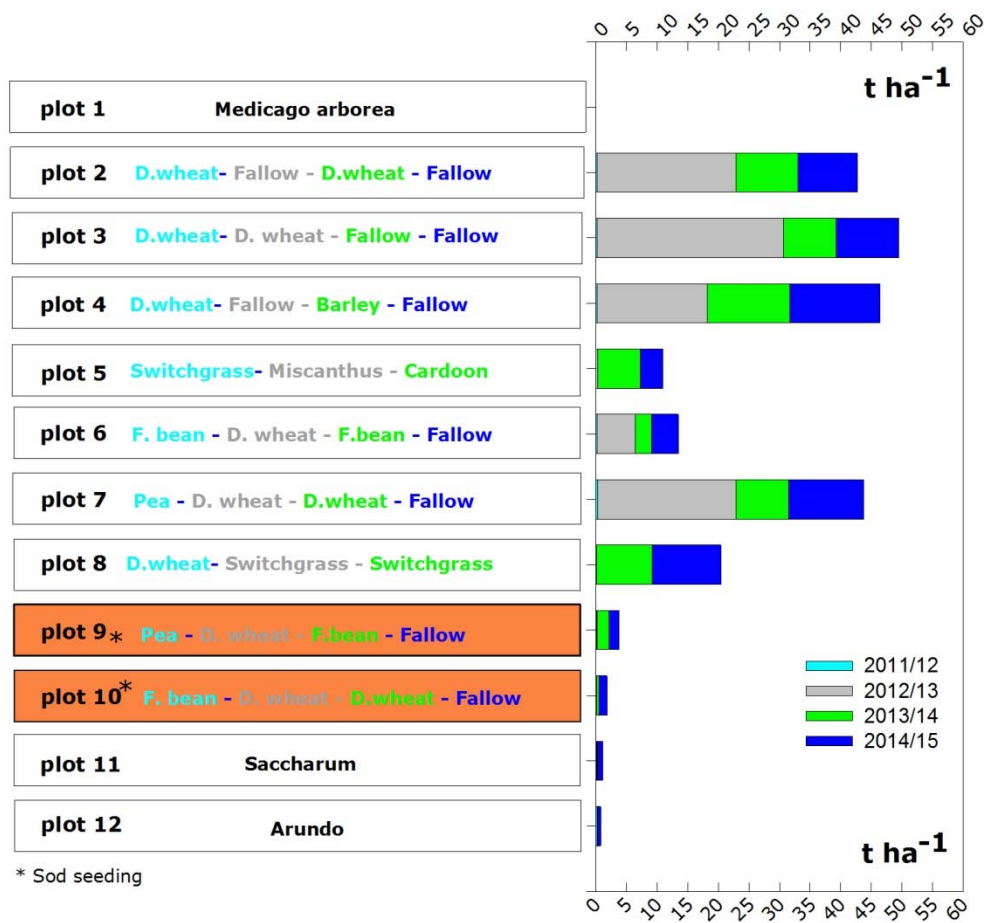


**Figure 8.** Height and biomass of switchgrass subjected to various concentrations of heavy metals (Pb and Cd) in plot experiment at CRDT- IITD.

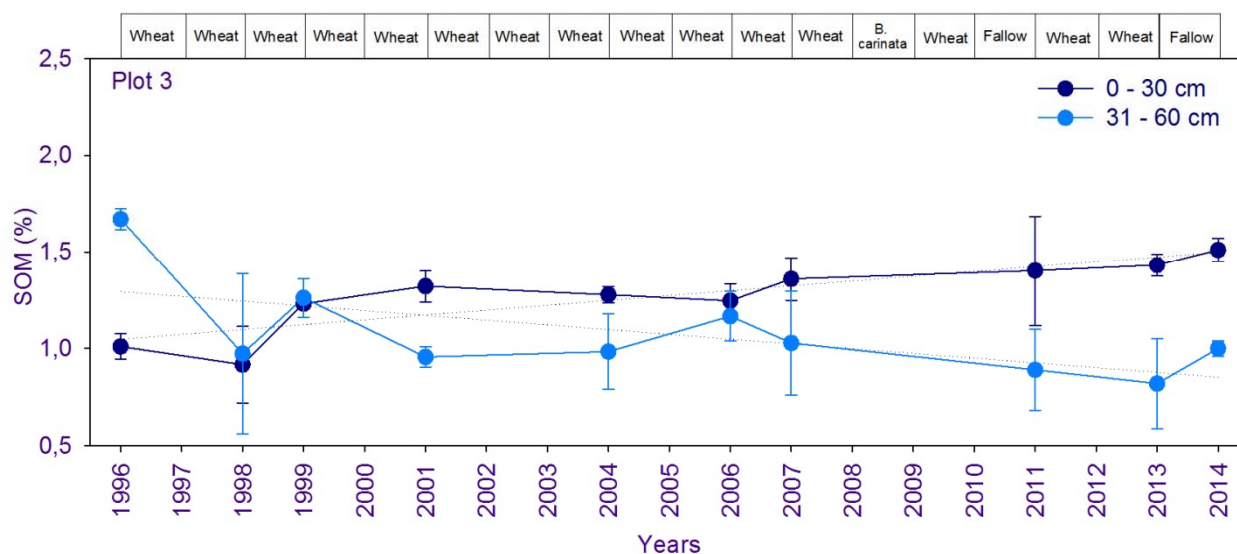
**Table 2.** Bioconcentration factor (BCF) and translocation index ( $\square\square$ ) of switchgrass treated with Pb and Cd at different concentration at CRDT- IITD experimental farm.

| Treat<br>ment | Heavy metal concentration(ppm) |      |           |      |           |      |           |      |           |      |
|---------------|--------------------------------|------|-----------|------|-----------|------|-----------|------|-----------|------|
|               | 4                              |      | 6         |      | 8         |      | 10        |      | 12        |      |
|               | BCF                            | Ti % | BCF       | Ti % | BCF       | Ti % | BCF       | Ti % | BCF       | Ti % |
| <b>Pb</b>     | -                              | -    | 0.19±0.03 | 11.6 | 0.16±0.03 | 10.5 | 0.16±0.02 | 9.9  | 0.09±0.02 | 8.7  |
| <b>Cd</b>     | 0.15±0.02                      | 11.7 | 0.13±0.04 | 9.9  | 0.11±0.02 | 9.3  | 0.08±0.02 | 9.0  | -         | -    |

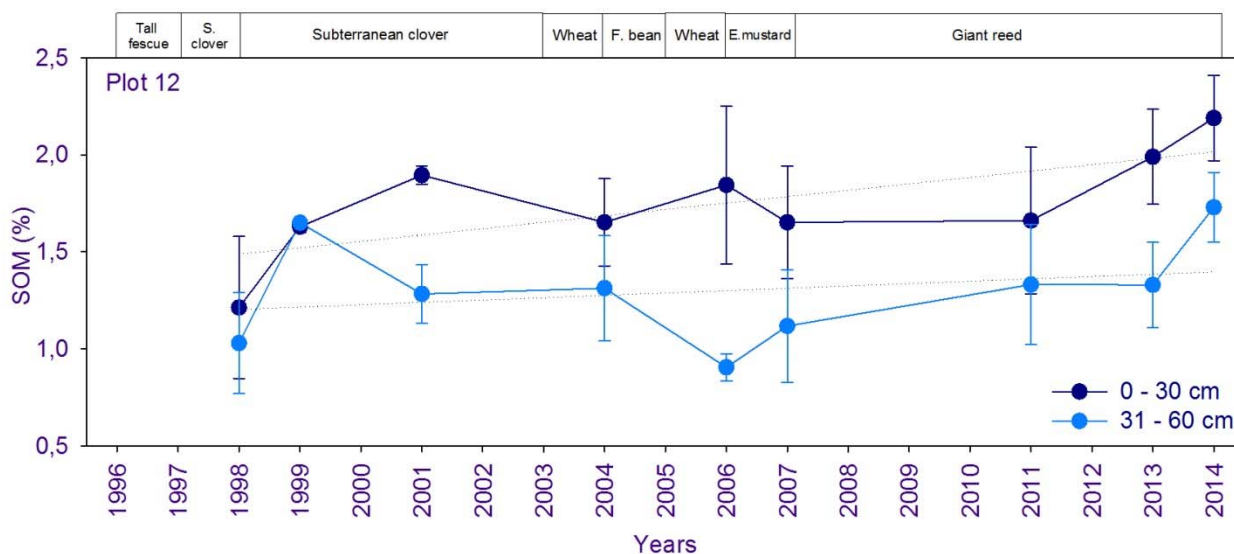
## Task 6.5 - Soil Erosion mitigation by perennial grasses



**Figure 9.** Soil losses (t ha<sup>-1</sup>) in the twelve plots established at UNICT experimental farm with perennial grasses, perennial legumes and annual species managed both with conventional and no tillage cropping systems.



**Figure 10.** Soil organic matter (SOM) content (%) in a plot managed with a typical Mediterranean cropping system (wheat-wheat-fallow) by UNICT.



**Figure 11.** Soil organic matter (SOM) content (%) in a plot managed with a typical Mediterranean cropping system (s.clover-D.wheat-F.bean-D.wheat-B.carinata) and a new perennial crops (*Arundo donax*) at the experimental farm of UNICT.